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(12) **United States Patent**
McLeod et al.

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(45) **Date of Patent: Apr. 23, 2024**

(54) **COMPOUNDS AND METHODS FOR TREATING, DETECTING, AND IDENTIFYING COMPOUNDS TO TREAT APICOMPLEXAN PARASITIC DISEASES**

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(73) Assignees: **The University of Chicago**, Chicago, IL (US); **J. Craig Venter Institute, Inc.**, Ajolla, CA (US); **The University of Sheffield** (GB); **Institute for Systems Biology**, Seattle, WA (US); **The University of Leeds** (GB); **The University of Strathclyde** (GB); **The Government of the United States**, Fort Detrick, MD (US)

(*) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 0 days.

(21) Appl. No.: **17/831,049**

(22) Filed: **Jun. 2, 2022**

(65) **Prior Publication Data**

US 2023/0140413 A1 May 4, 2023

Related U.S. Application Data

(62) Division of application No. 16/063,877, filed as application No. PCT/US2016/067795 on Dec. 20, 2016, now Pat. No. 11,414,385.

(60) Provisional application No. 62/306,385, filed on Mar. 10, 2016, provisional application No. 62/270,264, filed on Dec. 21, 2015.

(51) **Int. Cl.**

C07D 215/233	(2006.01)
A61P 33/06	(2006.01)
C07D 401/12	(2006.01)
C07D 405/12	(2006.01)
C07D 471/04	(2006.01)
C07D 487/04	(2006.01)

(52) **U.S. Cl.**

CPC ***C07D 215/233*** (2013.01); ***A61P 33/06*** (2018.01); ***C07D 401/12*** (2013.01); ***C07D 405/12*** (2013.01); ***C07D 471/04*** (2013.01); ***C07D 487/04*** (2013.01)

(58) **Field of Classification Search**

CPC C07D 215/233

See application file for complete search history.

(56) **References Cited**

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Primary Examiner — Paul V Ward

(74) *Attorney, Agent, or Firm* — McDonnell Boehnan Hulbert & Berghoff LLP

(57) **ABSTRACT**

Disclosed herein are novel compounds for treating apicomplexan parasite related disorders, methods for their use; cell line and non-human animal models of the dormant parasite phenotype and methods for their use in identifying new drugs to treat apicomplexan parasite related disorders, and biomarkers to identify disease due to the parasite and its response to treatment.

17 Claims, 22 Drawing Sheets

Specification includes a Sequence Listing.

Fig. 1A

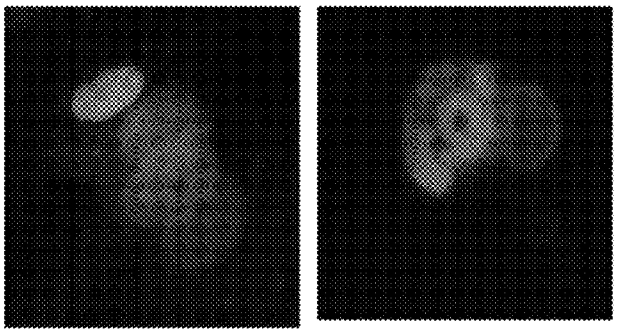


Fig. 1B

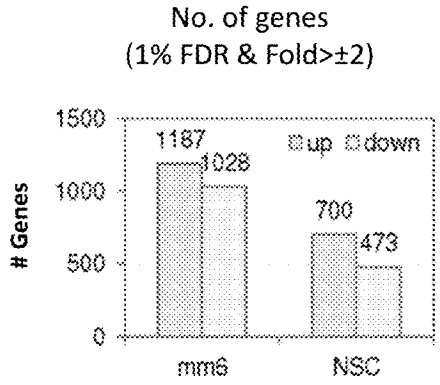


Fig. 1C

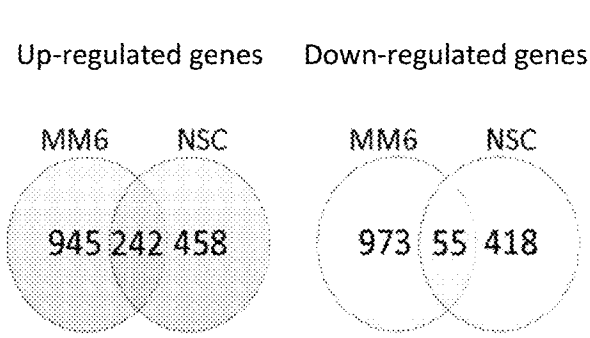


Fig. 1D

MiRs most altered

MiR	logFC (cont)	Log cpm	P value	FDR
hsa-mir-708	-11.46	5.67	6.11E-22	2.54E-18
hsa-mir-29b	-5.46	5.99	1.85E-15	2.56E-12
ENST00000474173	-9.95	4.25	7.09E-15	7.34E-12
hsa-mir-32	-5.48	7.42	8.85E-15	7.34E-12
hsa-mir-142	-4.62	10.48	1.77E-13	1.23E-10
hsa-mir-3656	-9.66	4.01	2.28E-11	1.05E-08

Fig. 2A

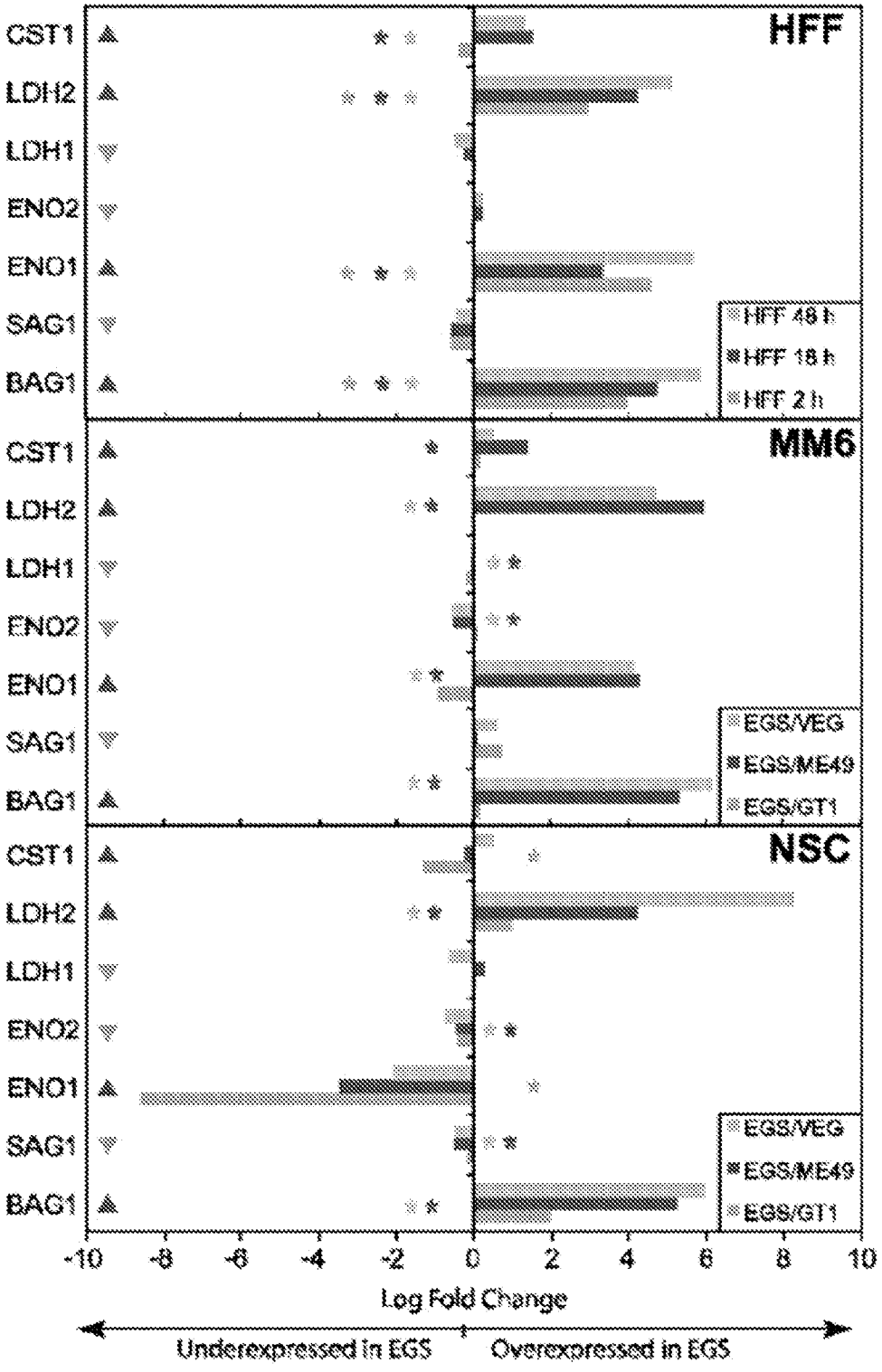
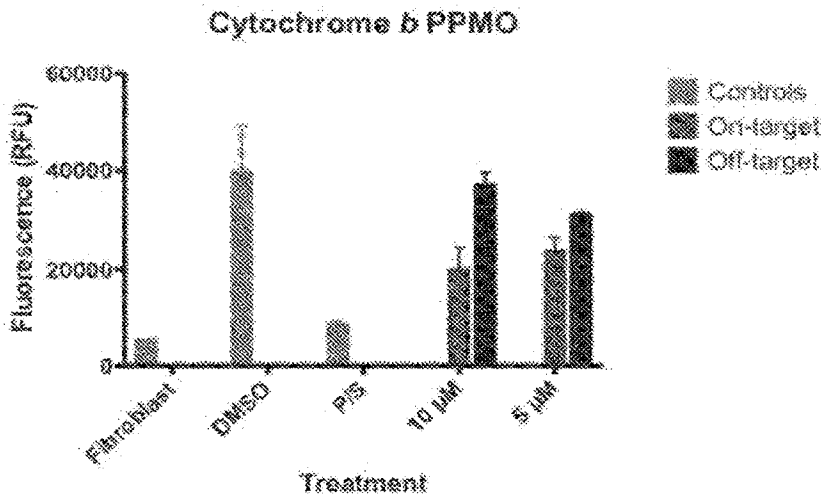

HFF
CST1
LDH2
LDH1
ENO2
ENO1
SAG1
BAG1
HFF 48 h
HFF 18 h
HFF 2 h
MM6
EGS/VEG
EGS/ME49
EGS/GT1
NSC
-10
-8
-6
-4
-2
0
2
4
6
8
10
Log Fold Change
Underexpressed in EGS
Overexpressed in EGS

Fig. 2B


Cytochrome b PPMO
Fluorescence (RFU)
60000
40000
20000
0
Controls
On-target
Off-target
FibroBlast
DMSO
PIS
10 μM
5 μM
Treatment

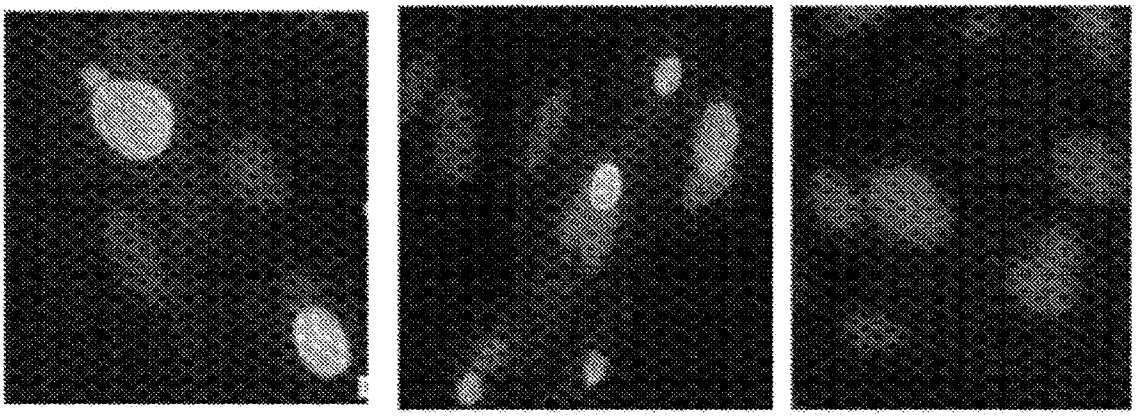
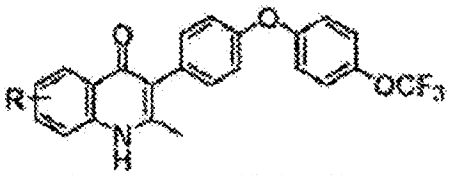
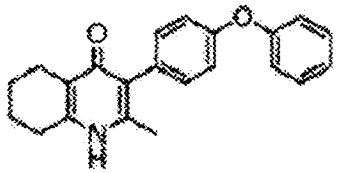


Fig. 3A



R = H ELQ-271 (1)
R = 6-Cl, 7-OMe ELQ-300 (2)
R = 6-F, 7-OMe ELQ-316 (3)



Tetrahydroquinolone MJM170 (4)

Fig. 3B

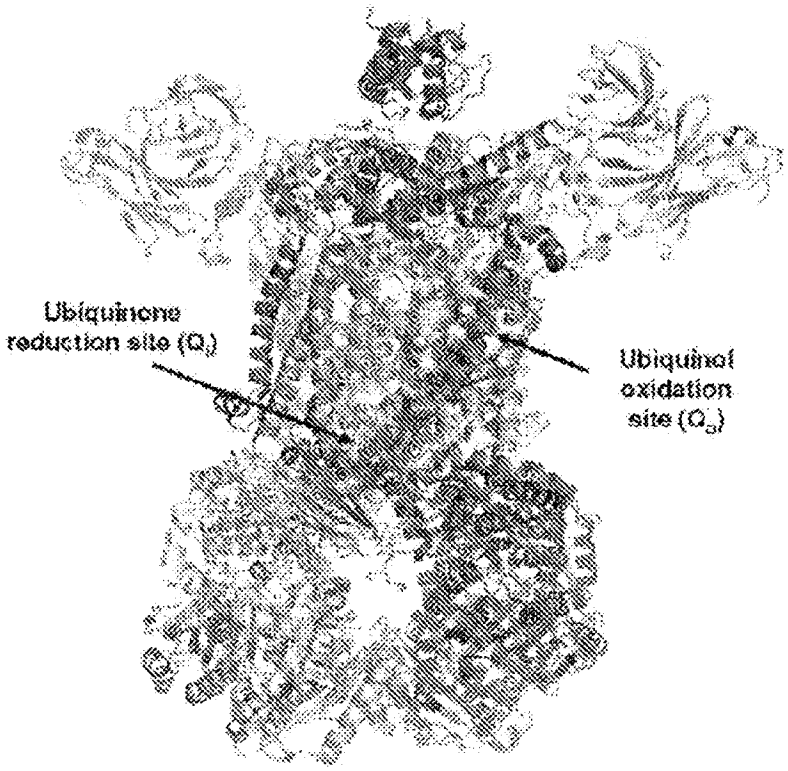
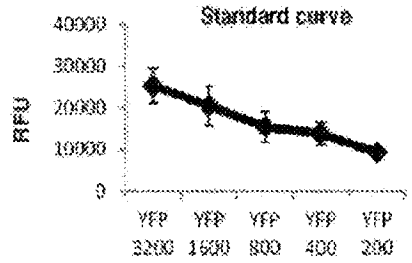


Fig. 4A



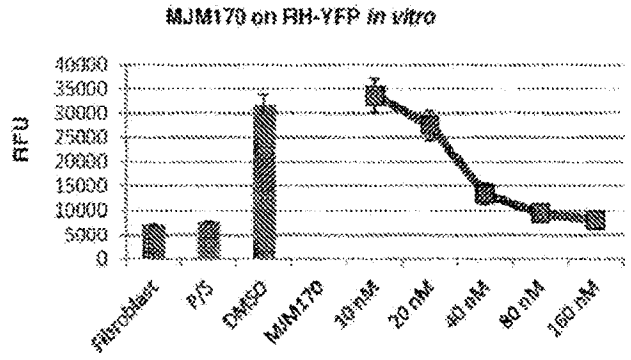


Fig. 4B

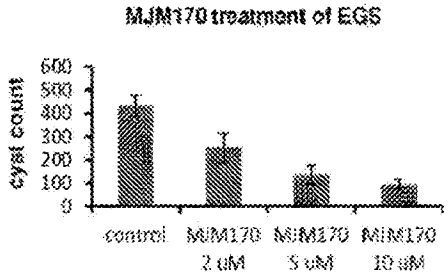


Fig. 4C

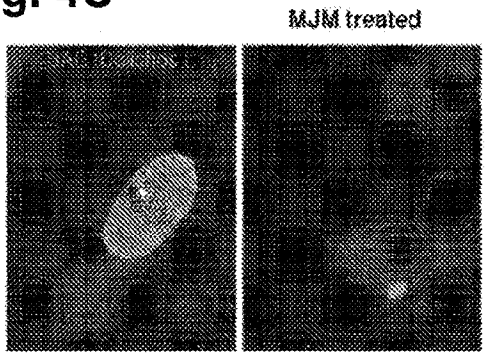


Fig. 4D

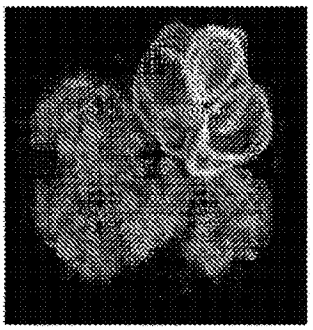


Fig. 4E

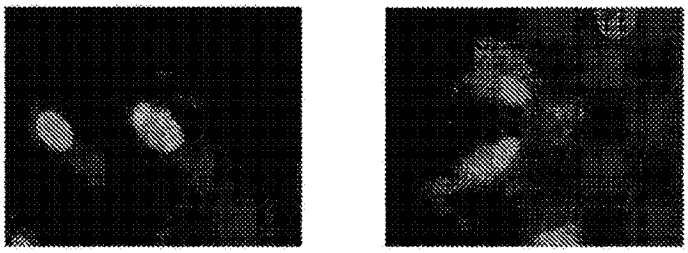


Fig. 5A

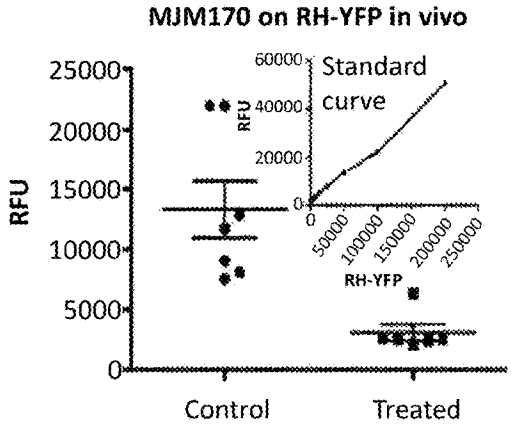


Fig. 5B

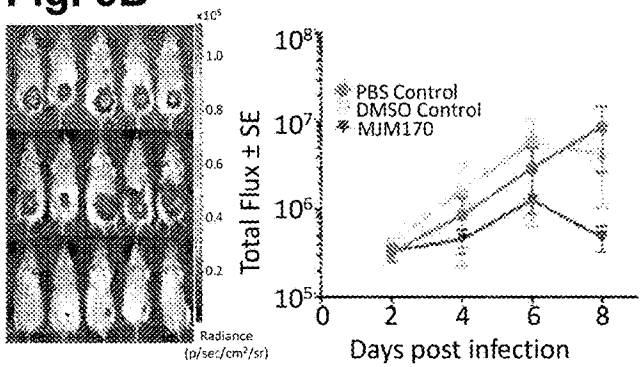


Fig. 5C

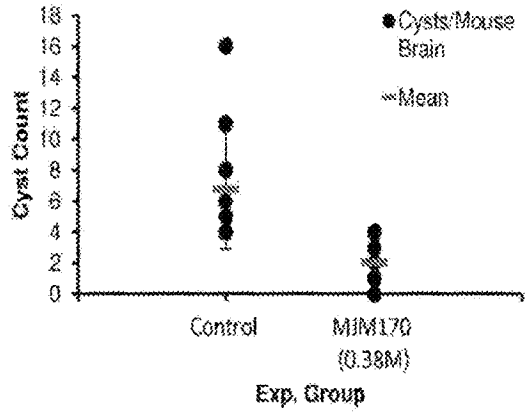


Fig. 5D

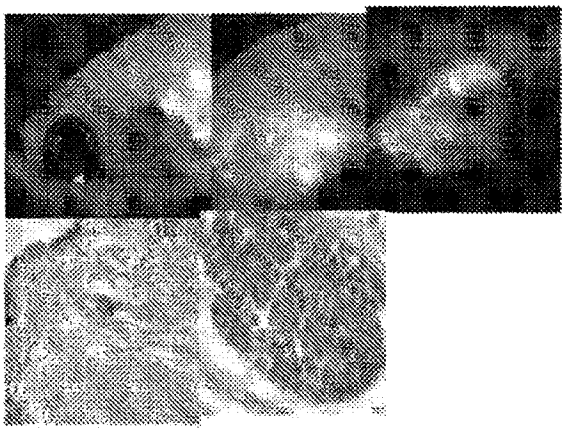


Fig. 6A

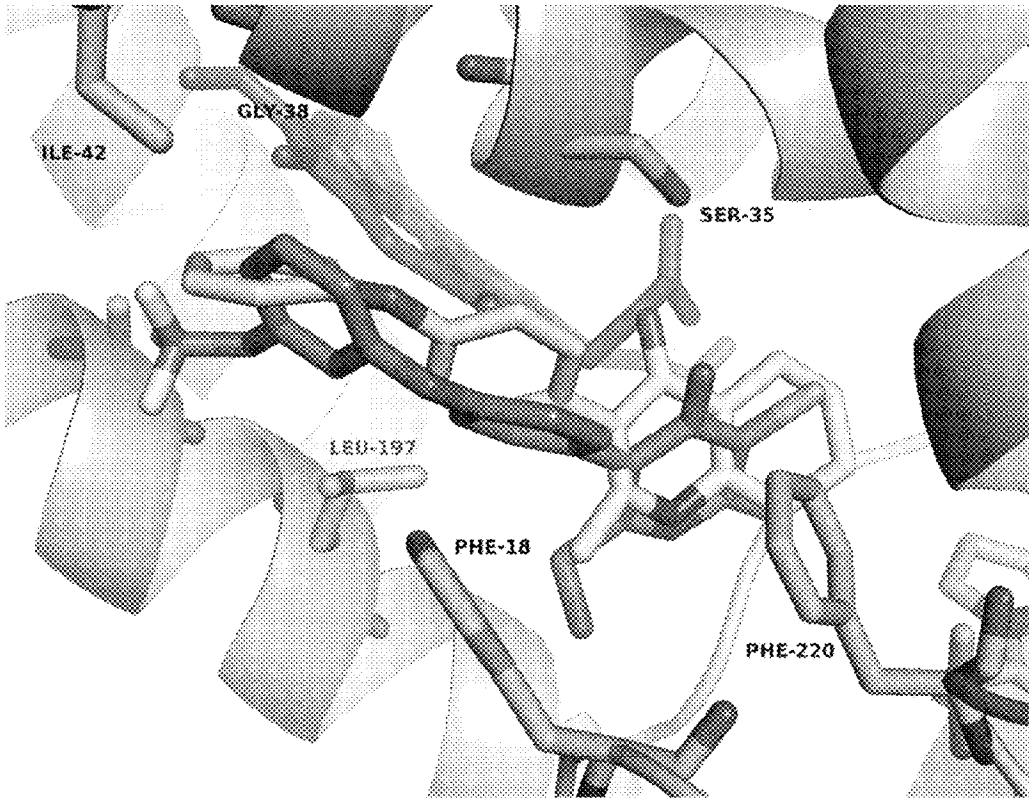

GLY-38
ILE-42
SER-35
LEU-197
PHE-18
PHE-220

Fig. 6B

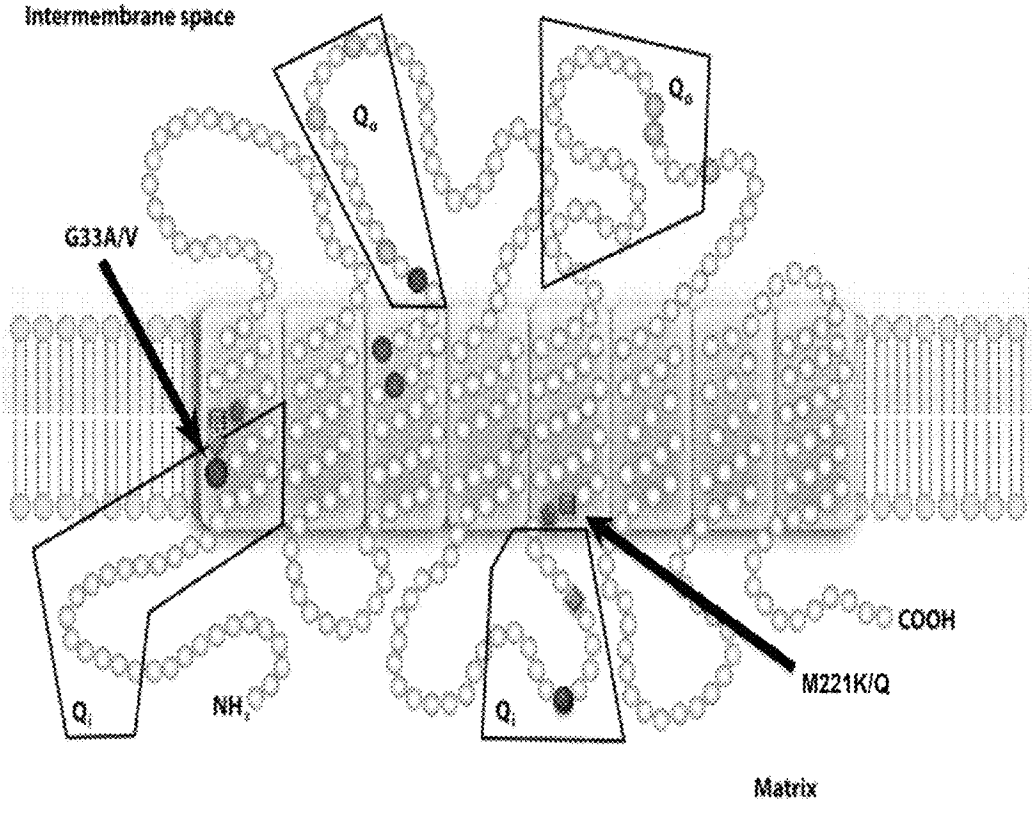

Intermembrane space
Q_o
Q_o
G33A/V
COOH
M221K/Q
Q_i
NH_2
Q_i
Matrix

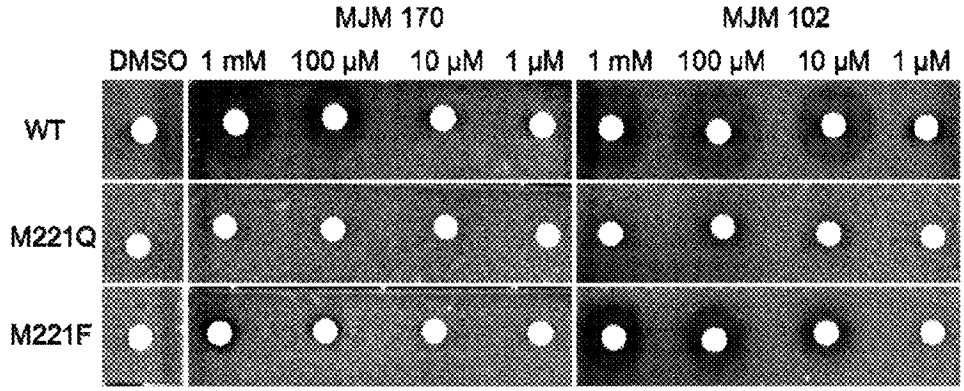
Fig. 6C

Cytochrome b Qi mutants

P. falciparum	Yeast	*T. gondii*	Bovine
G33A G33V	G37	A37	G38
F210	**M221K M221Q**	F215	F220

Cytochrome b sequences (UniProtKb):Toxo: O20672; Human: P00156;Pf: Q02768;Yeast: P00163; Bovine: P00157. Both reported Qi resistant mutants (G33A and M221K) steric clash with Qi inhibitors for a loss of binding affinity. Both are useful indicators for target confirmation. Adapted and modified from reference ... with permission

Fig. 6D



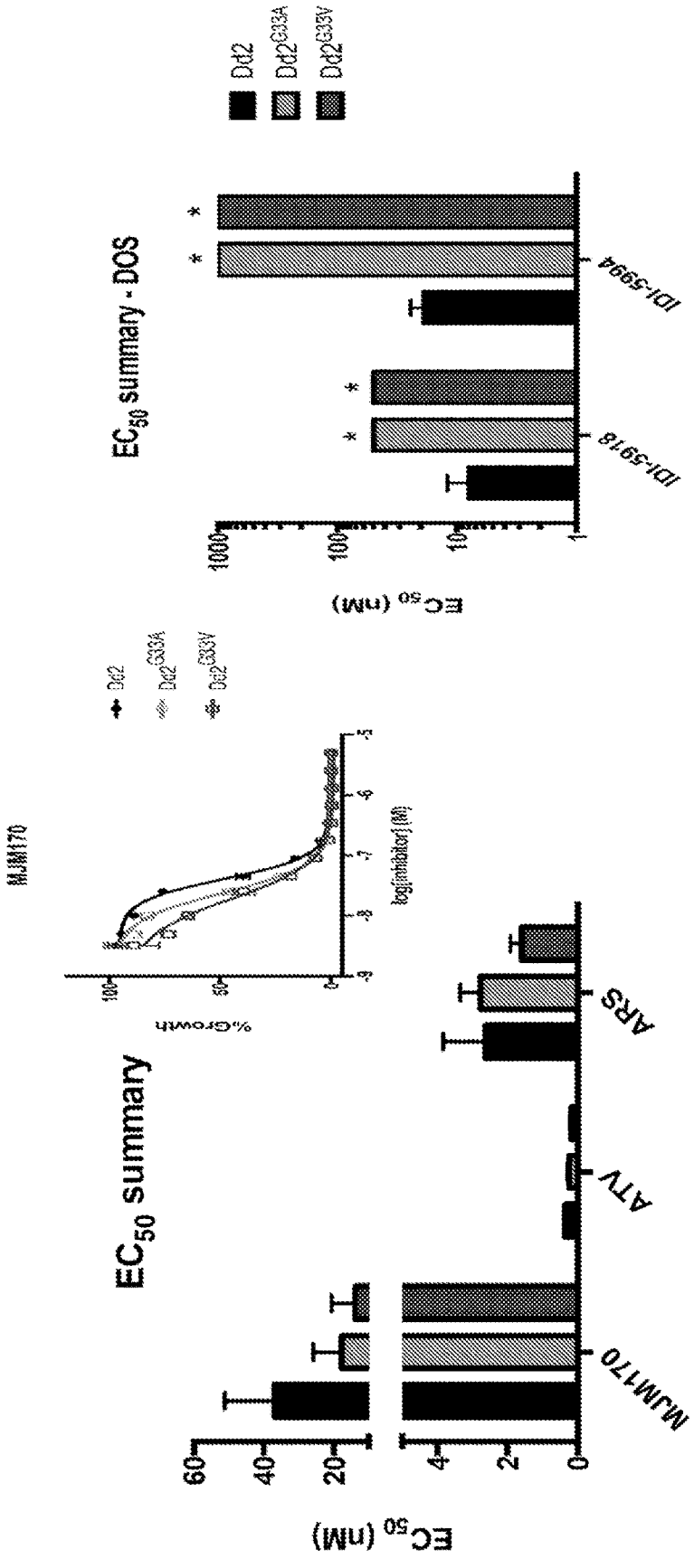

Dd2
Dd2G33A
Dd2G33V
EC50 summary - DOS
IDI-5994
IDI-5918
EC50 (nM)
MJM170
%Growth
log(inhibitor) (M)
EC50 summary
ARS
ATV
MJM170
EC50 (nM)

Fig. 6E

Fig. 6F

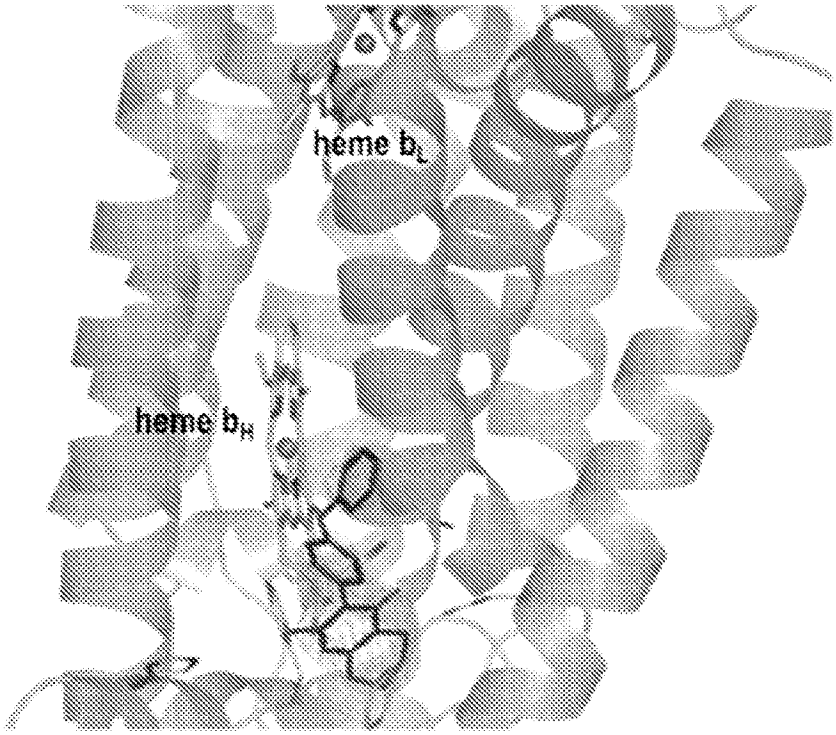


Fig. 6G

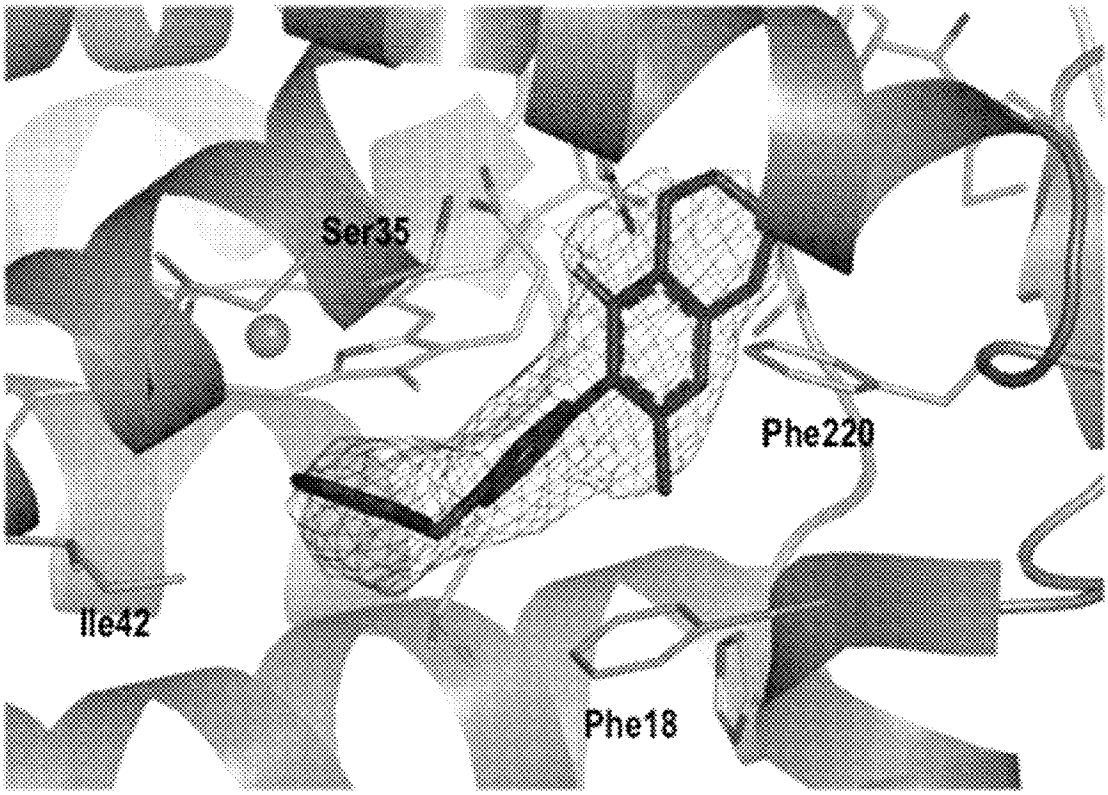
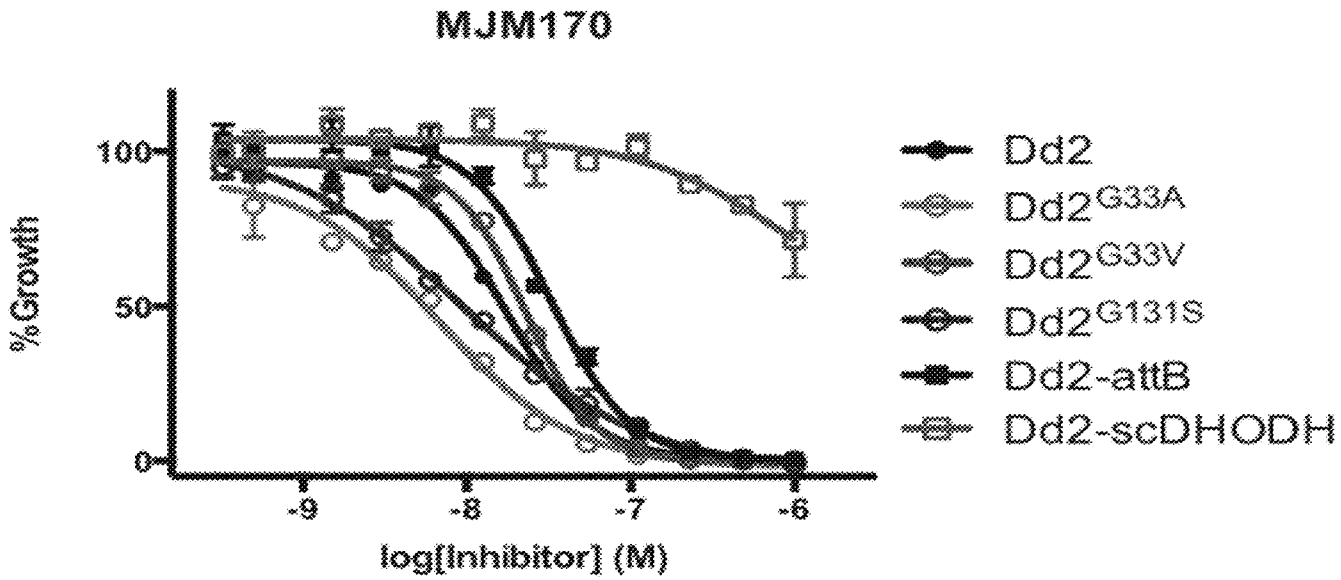
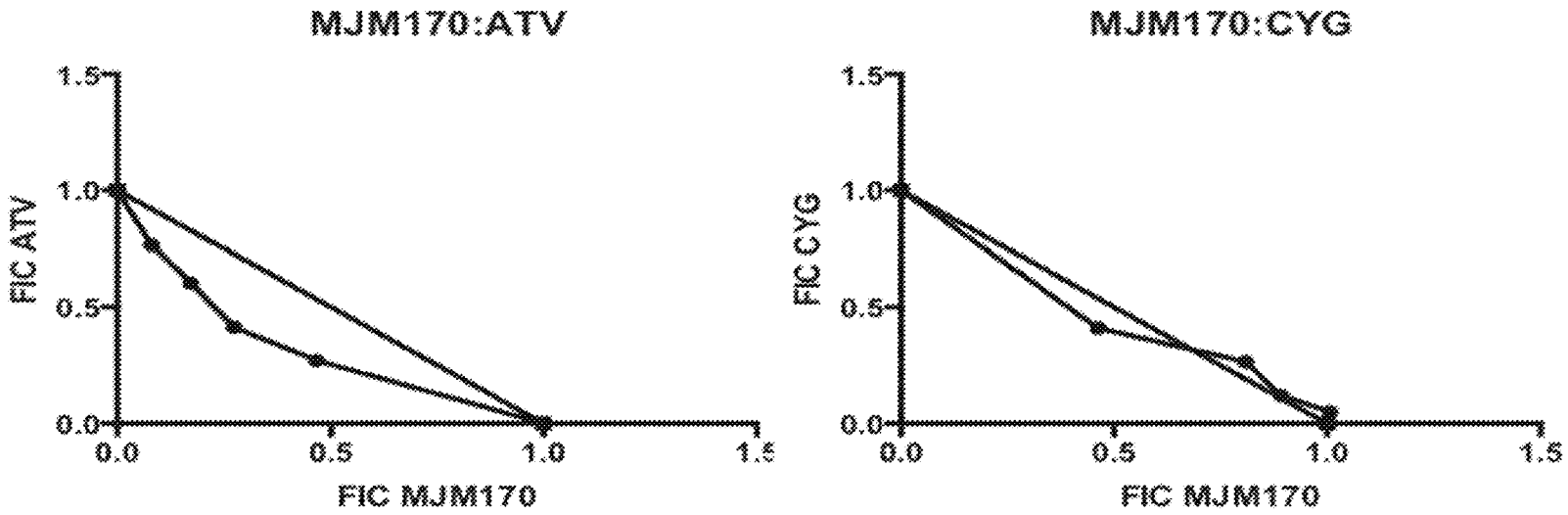


Fig. 7A



	Dd2	Dd2: G33A	Dd2: G33V	Dd2: G131S	Dd2-attB	Dd2-scDHODH
MJM170	29.5 ± 9.9	18.4 ± 9	14.3 ± 11	12.4 ± 3.8	35.5 ± 9.9	> 1,000
IDI-5918	9.24 ± 2.6	171 ± 73	> 1,000	5.85 ± 3	15.4 ± 3.2	> 1,000
BRD6323	35.6 ± 13	> 1,000	> 1,000	36.9 ± 13	81.8 ± 18	> 1,000
IDI-0020	49.0 ± 31	37.8 ± 24	21.7 ± 24	> 1,000	69.0 ± 56	> 1,000
Artesunate	2.95 ± 0.94	2.53 ± 0.69	2.08 ± 1.4	3.38 ± 0.48	4.40 ± 2.3	3.97 ± 2.3
Atovaquone	0.461 ± 0.14	0.365 ± 0.053	3.85 ± 7.1	0.479 ± 0.17	1.11 ± 0.27	> 50

Fig. 7B



Ratio MJM170:ATV or CYG	MJM170 + ATV			MJM170 + CYG		
	FIC MJM170	FIC ATV	Σ FIC	FIC MJM170	FIC CYG	Σ FIC
8:2	0.47 ± 0.08	0.35 ± 0.06	0.82	0.65 ± 0.2	0.41 ± 0.003	1.1
6:4	0.27 ± 0.03	0.52 ± 0.1	0.79	0.83 ± 0.04	0.2 ± 0.05	1.0
4:6	0.16 ± 0.02	0.72 ± 0.1	0.89	0.83 ± 0.05	0.089 ± 0.03	0.92
2:8	0.069 ± 0.01	0.82 ± 0.05	0.89	0.89 ± 0.1	0.036 ± 0.01	0.93

MJM170 + BRD6323			
Ratio MJM170:BRD6323	FIC MJM170	FIC BRD6323	Σ FIC
8:2	1.6 ± 0.4	0.39 ± 0.1	2
6:4	0.86 ± 0.3	0.55 ± 0.2	1.4
4:6	0.60 ± 0.3	0.91 ± 0.4	1.5
2:8	0.30 ± 0.09	1.2 ± 0.4	1.5

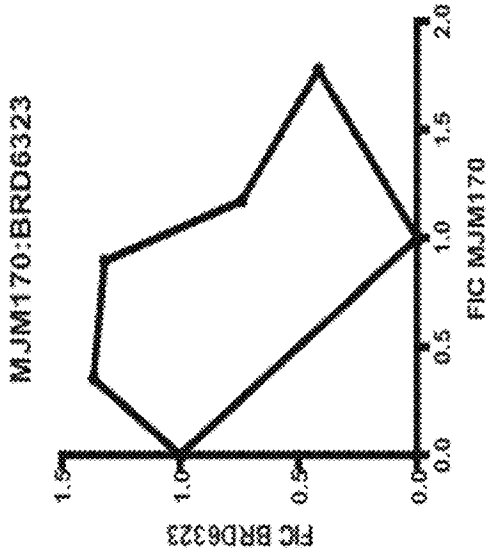


Fig. 7C

Fig. 8

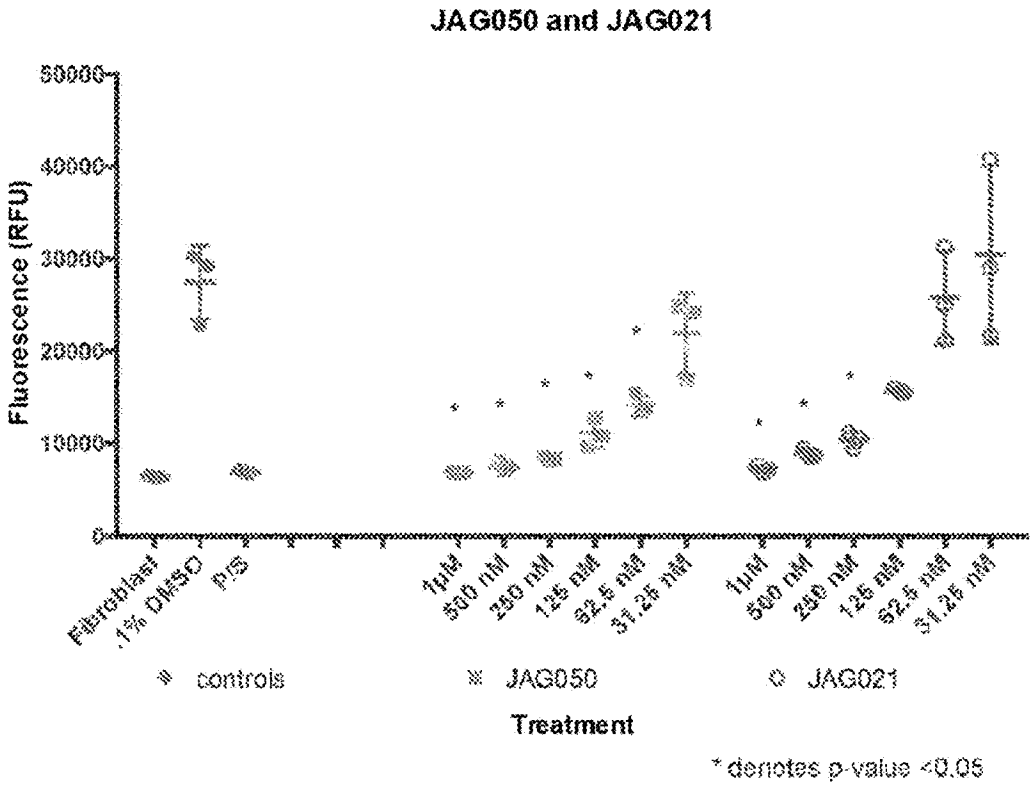

JAG050 and JAG021
Fluorescence (RFU)
50000
40000
30000
20000
10000
0
Fibroblast
1% DMSO
PBS
1μM
500 nM
250 nM
125 nM
62.5 nM
31.25 nM
1uM
500 nM
250 nM
125 nM
62.5 nM
31.25 nM
controls
JAG050
JAG021
Treatment
* denotes p-value <0.05

Fig. 9

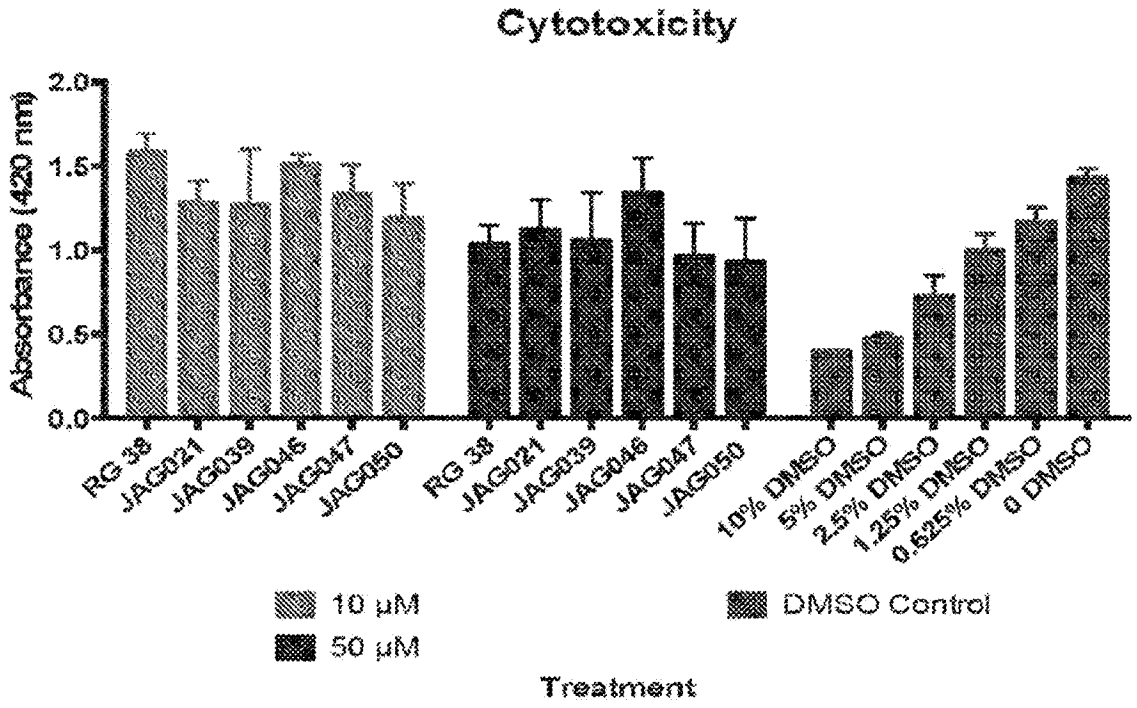

Cytotoxicity
Absorbance (420 nm)
2.0
1.5
1.0
0.5
0.0
RG 38
JAG021
JAG039
JAG046
JAG047
JAG050
RG 38
JAG021
JAG039
JAG046
JAG047
JAG050
10% DMSO
5% DMSO
2.5% DMSO
1.25% DMSO
0.625% DMSO
0 DMSO
10 µM
50 µM
DMSO Control
Treatment

Fig. 10A

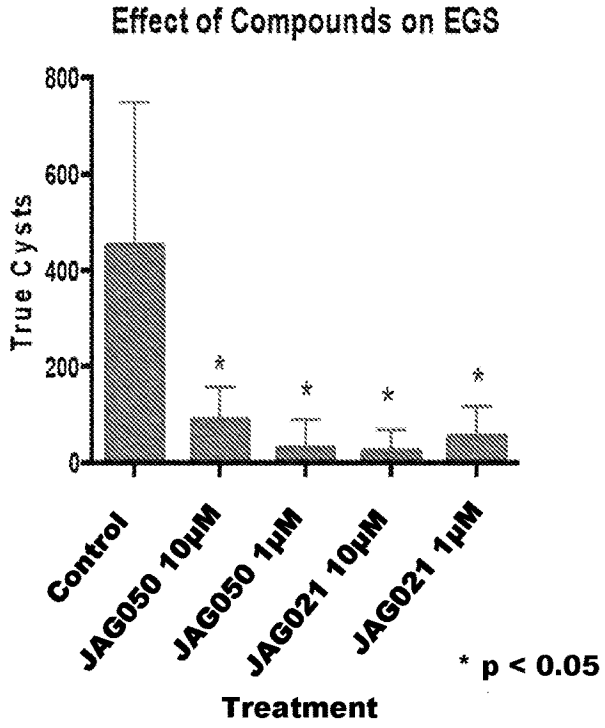


Fig. 10B

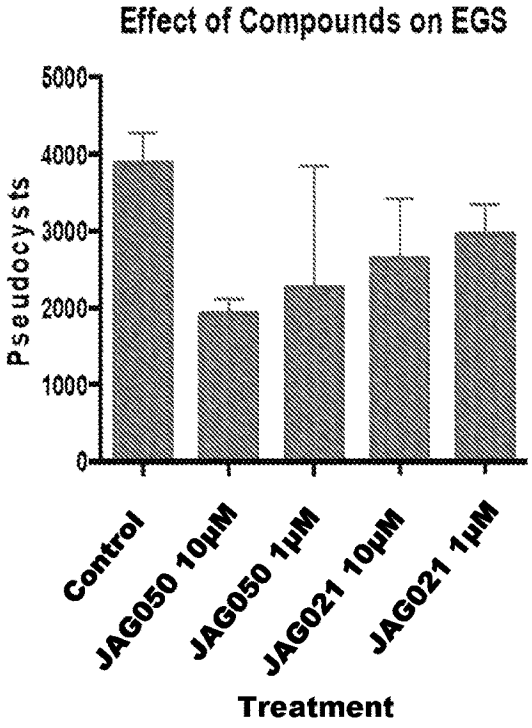


Fig. 10C

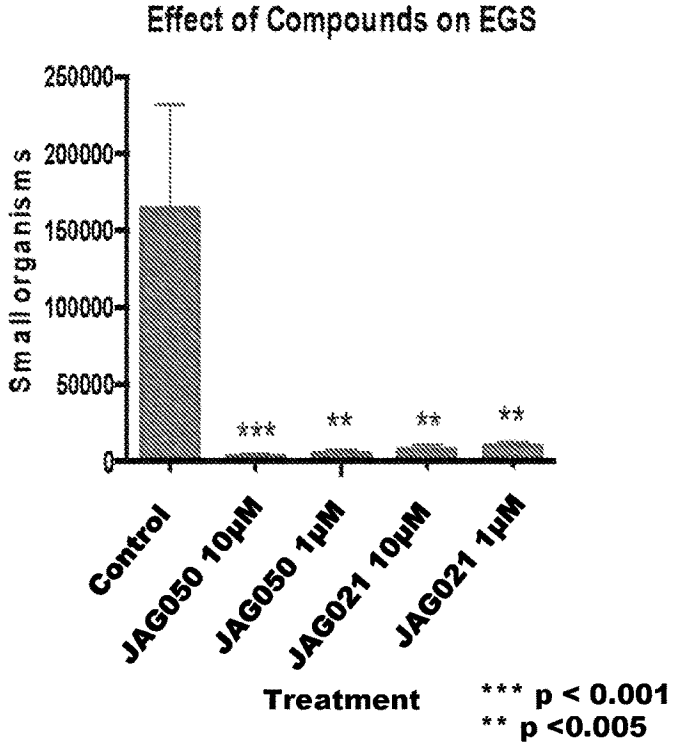


Fig. 11

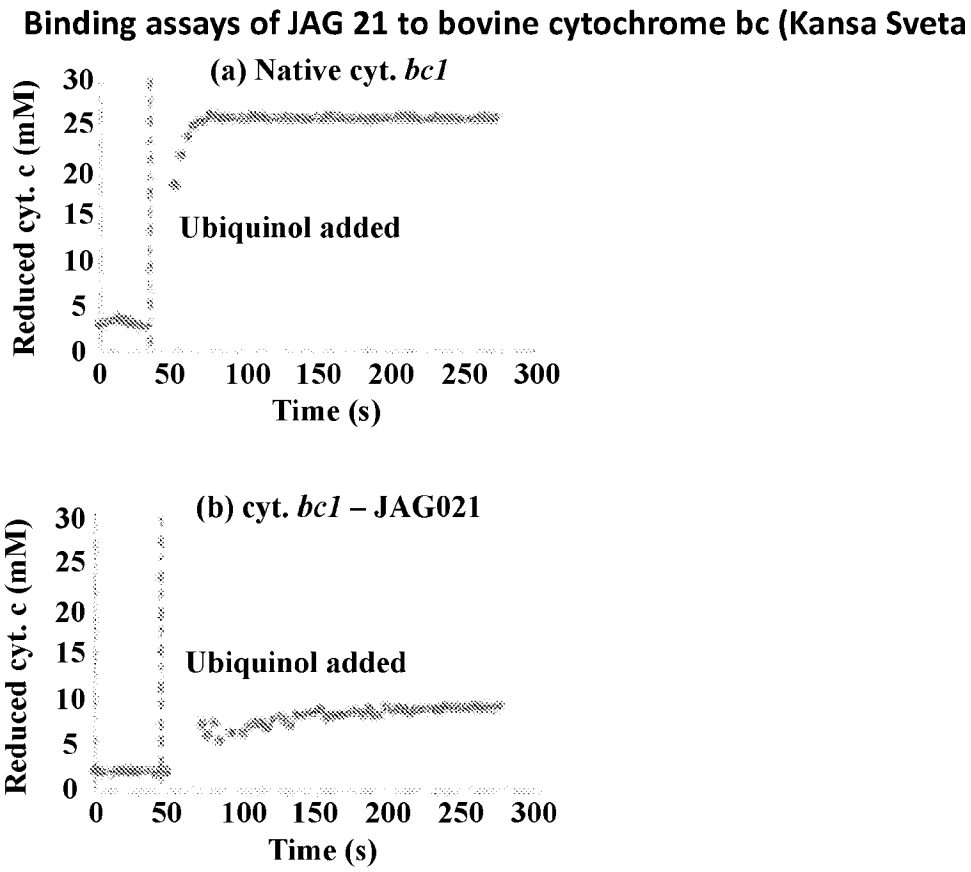
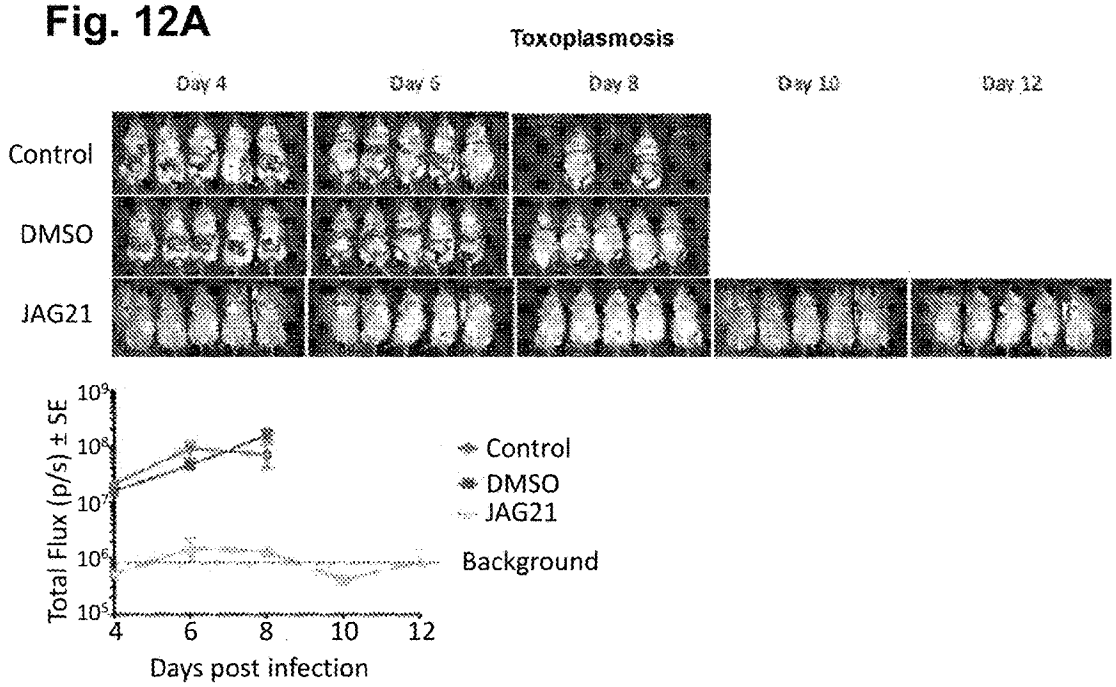

Binding assays of JAG 21 to bovine cytochrome bc (Kansa Sveta
(a) Native cyt. bc1
Reduced cyt. c (mM)
30
25
20
15
10
5
0
Ubiquinol added
0
50
100
150
200
250
300
Time (s)
(b) cyt. bc1 – JAG021
Reduced cyt. c (mM)
30
25
20
15
10
5
0
Ubiquinol added
0
50
100
150
200
250
300
Time (s)

Fig. 12A



Malaria

Fig. 12B

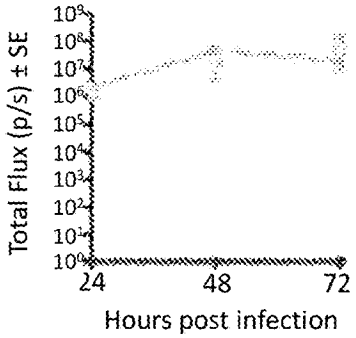


Fig. 12C

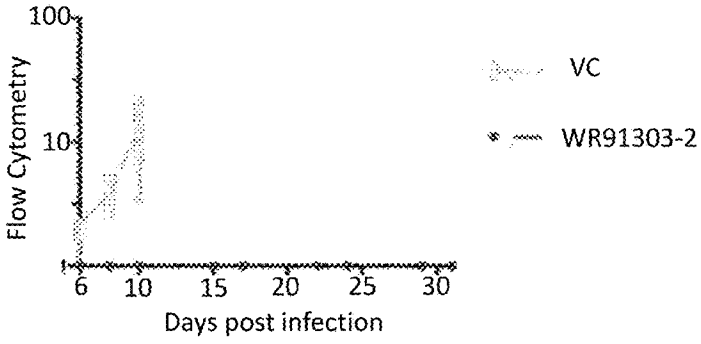


Fig. 12D

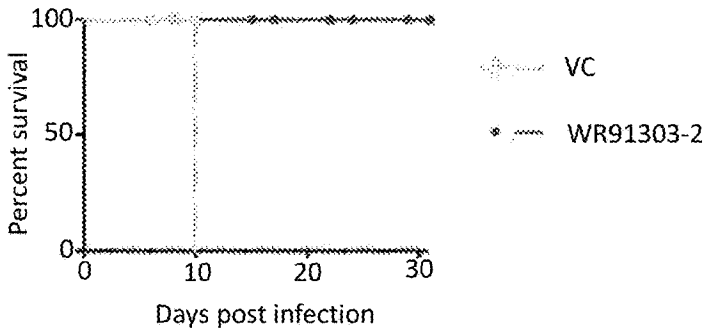
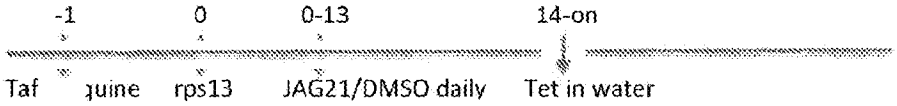


Fig. 13

Goal: To test if JAG21 and/or Tafenoquine be able to kill the dormant stage of T. gondii: rps13

Experiment design:



4 groups: 5/group	D-1: Inject Tafenoquine	D0 Inject rps13	D0-D13 daily Inject JAG21	D0-D13 daily Inject DMSO
Control		x		x
Tafenoquine	x	x		x
JAG21		x	x	
T+J	x	x	x	

4 groups: 5/group	D14 Give tet water	
Control	x	Monitor the mice
Tafenoquine	x	Take out the spleen once the
JAG21	x	mouse is really sick for histopath
T+J	x	

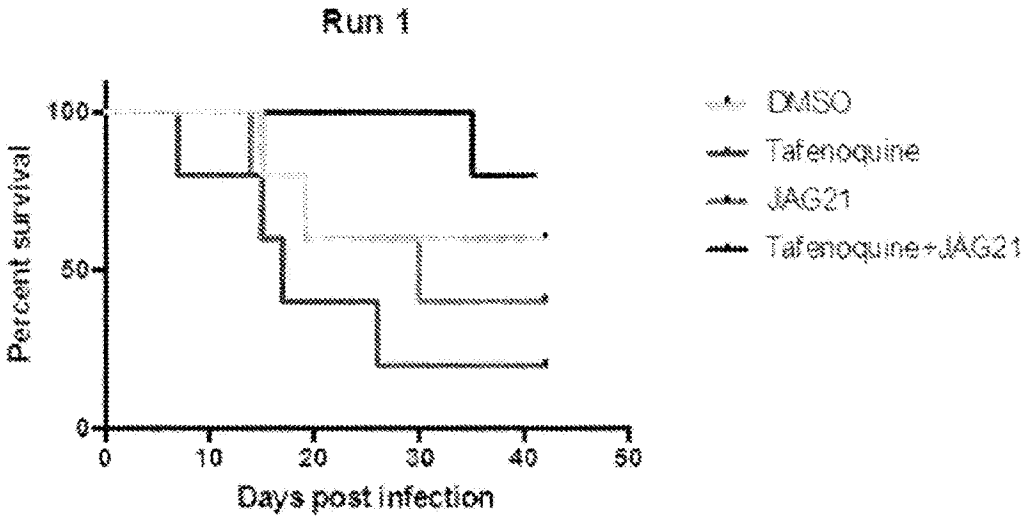


Fig. 14A

Pt	Ethnicity	New Seizure	Macular Disease	Increase Dye Test	Elevated CSF Protein
1a	Caucasian	Yes (myoclonic)	Yes	Yes	Yes
1b	Caucasian	No	No	ND	ND
2a	Filipino/Caucasian	Yes (myoclonic)	Yes	Yes	Yes
2b	Filipino/Caucasian	No	No	ND	ND
3a	Hispanic	Yes (hypsarrhythmia)	Yes	ND	ND
3b	Hispanic	No	No	ND	ND

Fig. 14B

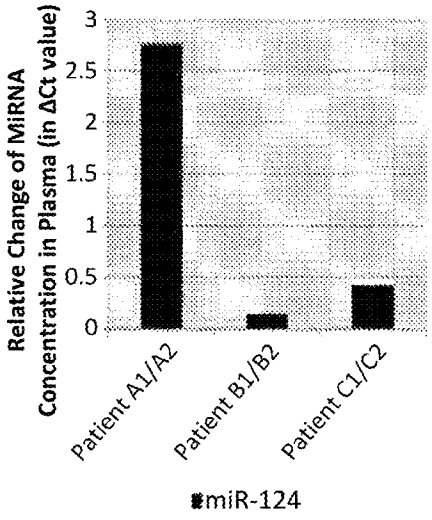


Fig. 14C

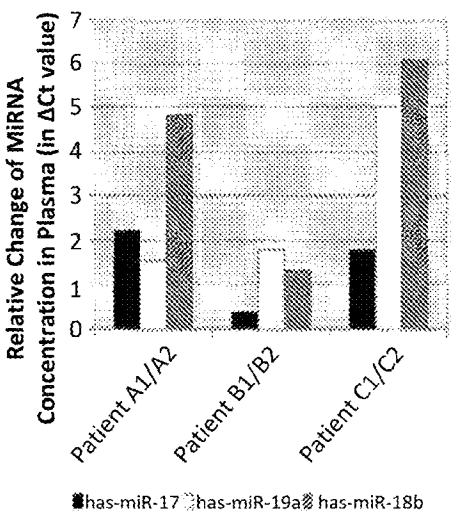
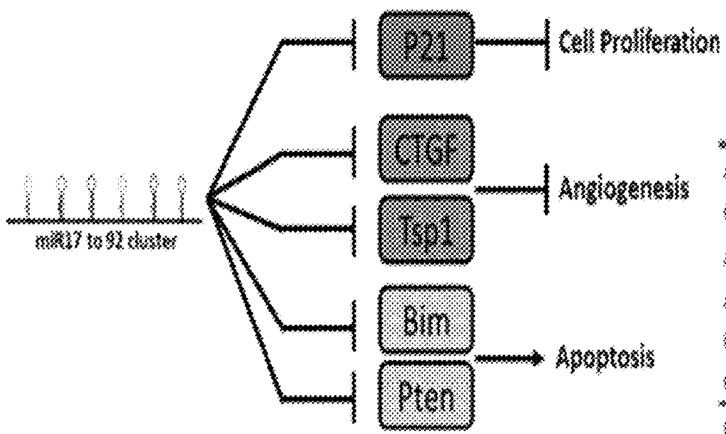


Fig. 14D



Protein Name	Protein Description	Organ	Endogenous Peptide abundances (Ratio to 100fmol of Spikded in Heavy peptide)					
			Patient A1	Patient A2	Patient B1	Patient B2	Patient C1	Patient C2
APCS	Serum Amyloid P-Component	Liver	22.034	10.772	15.123	9.468	32.030	15.008
GC	Vitamin D-Binding Protein Precursor	Liver	26.299	15.782	31.825	14.419	32.795	17.138
APCS	Serum Amyloid P-Component	Liver	12.999	7.469	12.680	7.361	20.519	11.146
APCS	Serum Amyloid P-Component	Liver	9.534	4.477	8.229	4.988	14.184	7.609
OXT	Oxytocin-Neurophysin 1	Brain	3.626	1.325	7.946	1.667	5.928	2.067
CLU	Clusterin	Brain	12.588	3.859	5.743	4.813	5.445	4.363
C4BPB	Component 4 Binding Protein, Beta	Liver	0.417	0.541	0.825	0.834	0.602	0.867
APOA1	Apolipoprotein A-I	Liver	0.759	1.574	0.888	1.347	0.423	0.786
PGLYRP2	N-Acetylmuramoyl-L-Alanine Amidase	Brain	3.980	5.163	4.645	4.684	2.960	4.290
SERPINA4	Kallistatin	Liver	6.307	8.867	4.402	4.548	2.530	4.920
APOA4	Apolipoprotein A-Iv	Liver	8.769	15.558	11.175	25.712	3.799	9.300
HPX	Hemopexin	Liver	157.685	167.509	124.487	155.807	88.474	131.117

COMPOUNDS AND METHODS FOR TREATING, DETECTING, AND IDENTIFYING COMPOUNDS TO TREAT APICOMPLEXAN PARASITIC DISEASES

CROSS REFERENCE

This application is a divisional application of U.S. patent application Ser. No. 16/063,877, filed Jun. 19, 2018, which is a U.S. national phase application of International Patent Application no. PCT/US2016/067795, filed on Dec. 20, 2016, which claims the benefit of U.S. Provisional Patent Application No. 62/270,264, filed Dec. 21, 2015, and U.S. Provisional Application No. 62/306,385, filed Mar. 10, 2016, each incorporated by reference herein in their entirety.

STATEMENT OF GOVERNMENT RIGHTS

This invention was made with government support under National Institutes of Health (NIH) contract number HHNS272200900007C, NIH, National Institute of Allergy and Infectious Diseases of the National Institutes of Health (NIAID) award numbers R01AI071319 (NIAID) and R01AI027530 (NIAID); NIAID contract Number HHNS272200900007C; NIAID award number U19AI110819: NIAID award numbers U01 AI077887 (NIAID) and U01AI082180 (NIAID); National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK) Grant #5T35DK062719-28: Defense Threat Reduction Agency award number 13-C-0055, and Department of Defense award numbers W911NF-09-D0001 and W911SR-07-C0101. The government has certain rights in the invention.

BACKGROUND

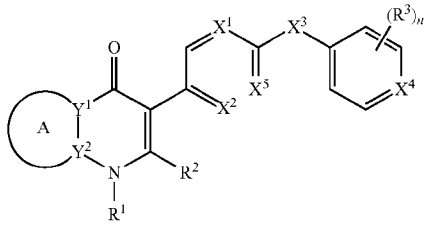
Apicomplexan parasitic infections, such as *Toxoplasma gondii* infections, can cause systemic symptoms, damage and destroy tissues, especially eye and brain and cause fatalities. Primary infections may be asymptomatic, or cause fever, headache, malaise, lymphadenopathy, and rarely meningoencephalitis, myocarditis, or pericarditis. Retinochoroiditis and retinal scars develop in up to 30% of infected persons, and epilepsy may occur. In immune-compromised and congenitally infected persons, active infection frequently is harmful. Recrudescence arises from incurable, dormant cysts throughout life. Current treatments against active *T. gondii* tachyzoites can have side effects such as hypersensitivity, kidney stones, and bone marrow suppression, limiting their use. Latent bradyzoites are not significantly affected by any medicines. Atovaquone partially, and transiently, limits cyst burden in mice, but resistance develops with clinical use. Thus, *T. gondii* infection is incurable with recrudescence from latent parasites posing a continual threat. Estimates of costs for available, suboptimal medicines to treat active, primary ocular, gestational and congenital infections, in just the U.S. and Brazil, exceed $5 billion per year.

Improved medicines are needed urgently. Molecular targets shared by *T. gondii* and Plasmodia make re-purposing compounds a productive strategy.

SUMMARY OF THE INVENTION

In one aspect, the invention provides compounds of the structure of Formula (I), pharmaceutical compositions thereof, and methods for their use in treating apicomplexan parasite related disorders):

(I)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof,

wherein

ring A combines with Y^1 and Y_2 to form a C_{3-7}cycloalkenyl or heteroaryl ring,

wherein the C_{3-7}cycloalkenyl or heteroaryl is optionally substituted by halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR, cyano or phenyl;

Y^1 is C or N;

Y^2 is C or N;

X^1 is $C(R^{x1})$ or N,

wherein R^{x1} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^2 is $C(R^{x2})$ or N,

wherein R^{x2} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^3 is O, N(R), S or C_{1-3}alkyl;

X^4 is C or N;

X^5 is C or N;

R^1 is hydrogen or C_{1-3}alkyl;

R2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR or —C(O)OR;

n is 0, 1, 2, 3 or 4;

each R^3 is independently halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5;

or two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane; and

each R is independently hydrogen or C_{1-3}alkyl.

In another aspect, the invention provides cell lines infected with an apicomplexan parasite, wherein the apicomplexan parasite genome comprises a gene encoding an Apetela 2 IV-4 protein with an M=>I modification at residue 570 ("AP2 IV-4 M570I") compared to its orthologous gene on the reference *T. gondii* ME49 strain (gene ID: TGME49_318470), non-human animal models comprising cell lines of the invention, and methods for use of each in identifying compounds for treating an apicomplexan parasitic infection.

In another aspect, the invention provides methods for treating an apicomplexan parasite infection (such as a *T. gondii* infection), comprising administering to a subject in need thereof an amount effective to treat the infection of an inhibitor (of up-regulated genes) or an activator (of down-regulated genes) of 1 or more up-regulated genes as discussed herein.

In a further aspect, the invention provides methods for identifying test compounds for apicomplexan parasite therapy, comprising identifying test compounds that reduce expression (for up-regulated genes), or increase expression (for down-regulated genes) of 1 or more apicomplexan parasite genes as discussed herein.

In one aspect, the invention provides a plurality of isolated probes that in total selectively bind to at least 2, 3, 4,

5, 6, 7, 8, 9, 10, 15, 20, 25, 30, 35, 40, 45, 100, 150, 200, 250, 500, or all of the markers as discussed herein, complements thereof, or their expression products, or functional equivalents thereof wherein at least 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, or all of the probes in total are selective for markers that are upregulated in the EGS strain of *T. gondii* after infection of human fibroblasts, neuronal stem cells or monocytic lineage cells.

In another aspect, the invention provides a plurality of isolated probes that in total selectively bind to at least 2, 3, 4, 5, 6, 7, 8, 9, 10, 15, 20, 25, 30, 35, 40, 45, 100, 150, 200, 250, 500, or all of the markers as discussed herein, complements thereof, or their expression products, or functional equivalents thereof, wherein at least 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, or all of the probes in total are selective for markers that are upregulated in human fibroblasts, neuronal stem cells or monocytic lineage cells after infection with *T. gondii*. including but not limited to infection with the EGS strain of *T. gondii*.

In another aspect, the invention provides methods for monitoring *T. gondii* infection in a subject, comprising monitoring levels in a blood sample from the subject of one or more markers selected from the group consisting of clusterin, oxytocin, PGLYRP2 (N-acetylmuramoyl-L-alanine amidase), Apolipoprotein A1 (apoA1), miR-17-92, and miR-124, wherein a change in levels of the one or more circulating markers compared to control correlates with *T. gondii* infection in the subject.

In another aspect, the invention provides methods for treating a *T. gondii* infection, comprising administering to a subject with a *T. gondii* infection an amount effective to treat the infection of ApoA1.

DESCRIPTION OF THE FIGURES

FIG. **1**A-**1**D. EGS morphology and effect on host cell transcriptomes FIG. **1***a*. EGS in human MM6 cells and NSC form cysts. Left NSC with EGS. Right MM6 with EGS. Note green dolichos cyst walls and BAG1 (red) in NSC. DAPI stained nuclei(blue). FIG. **1**B, FIG. **1**C. Effects of EGS infection on MM6 and NSC transcriptomes: EGS transcripts in MM6 compared with NSC shows overlap of, as well as unique patterns of, transcripts. Differentially expressed genes in MM6 and NSC cells infected with EGS parasite were identified based on criteria of 1% FDR and absolute fold-change ≥2. Number of DEGs in each cell line are presented with bar graph (FIG. **1**B) and Venn diagram are used to show general comparison of DEGs identified between the two cell lines (FIG. **1**B). There is both communality, overlap in genes modulated and independence in others between cell types indicating cell type also influences cell type. Red and green colors were used to represent up- and down-regulated genes, cell line used is indicated on bottom (FIG. **1**B-FIG. **1**C). Functional enrichment analysis was performed for gene ontology (GO) biological process and KEGG pathways. P-values derived from analysis were −log 10 transformed and presented as a heat map. Pink and blue colors indicate GO terms or KEGG pathways enriched by up- and down-regulated genes, respectively. Enriched pathways or biological processes are listed on right of panels and cell lines are indicated on top. FIG. **1**D. Host cell miR-seq analysis reveals that EGS regulates host cell miRNAs critical in pathogenesis and latency. An especially interesting down-modulated miRNA is hsa-miR-708-5p which is expressed particularly in brain and retina cells causing apoptosis[65]. When *T. gondii* downmodulates this as an encysted bradyzoite in neuronal cells, it would prevent hosts from initiating apoptosis to eliminate chronically infected neurons. f. Parasite genetics and human host cell type have a profound influence on *T. gondii* gene expression. MDS plot comparing *T. gondii* gene expression profiles from MM6 and NSC cells infected with EGS, GT1, ME49 and VEG strains for 18 hours and HFF cell cultures infected with EGS strain for 2, 18 and 48 hours.

FIG. **2**A-**2**B. Differential Gene Expression (DGE) analyses and effects of inhibition of cytochrome bc1. FIG. **2**A. DGE analysis of bradyzoite- and tachyzoite-specific markers during EGS infections of HFF cultures at 2, 18 and 48 hours (top panel), MM6 cells at 18 hours (middle panel) or NSC cultures at 18 hours (bottom panel) versus infections of same host cells with canonical strains GT1, ME49 or VEG at 18 hours (averaged across the three canonical strains for HFF infections). Genes reported as being over- or under-expressed during bradyzoite differentiation is indicated with red or green arrows respectively. "*", q-value ≤0.05; Log FC, logarithm of the fold change in gene expression. CST1, SAG-related sequence SRS44^{S114}; LDH2, lactate dehydrogenase 2^{S115}; LDH1, lactate dehydrogenase 1^{S115}; ENO2, enolase 2; ENO1, enolase 1^{S116}; SAG1, SAG-related sequence SRS29B; BAG1, bradyzoite antigen BAG1^{S115}. FIG. **2**B. Effect of known cytochrome b inhibitors on EGS. Morpholino conjugated to a Vivoporter (called PPMO) designed to knock down cytochrome b compared with off target control has a significant effect in reducing replication of YFP RH strain tachyzoites at 5 and 10 μM ($p<0.05$) but only a very small effect on size and number of EGS cysts in HFF. As a poorly soluble inhibitor of cytochrome b, ELQ271 was reported to partially reduce cyst numbers in mice[27] and is shown herein also to reduce the EGS cysts in vitro at 10 μM in this novel model. This demonstrates the utility of this novel in vitro model by indicating that inhibition of cytochrome b Qi is associated with reduction of cysts in vivo in a mouse model, even when there are serious limitations caused by insolubility of this inhibitory compound. This poor solubility significantly limits ELQ271 as a candidate for progression to a medicine. Increasing selectivity for the parasite enzyme with our new scaffold is another critical challenge.

FIG. **3**A-**3**B. FIG. **3**A. Structures of the ELQ class (1-3) and the tetrahydroquinolone scaffold (4).[27,45,49,53]. Low solubility of the ELQs has been a serious concern going into preclinical evaluation for treatment of malaria.[27] FIG. **3**B. *Saccharomyces cerevisiae* cytochrome bc_1 X-ray structure (PDB ID: 1KB9)[5] The complex contains 11 subunits and 3 respiratory subunits (cytochrome b, cytochrome c1 and Rieske protein). The cytochrome b subunit provides both quinone binding sites (Q_o and Q_i) highlighted as grey and pink surfaces respectively.

FIG. **4**A-**4**E. ELQ inhibitors provide a new scaffold and approach yielding compounds that are potent inhibitors of tachyzoites and cysts in vitro. Study of Inhibitors in vitro is summarized in Table 2 and led to selection of MJM170 as a promising novel scaffold for both tachyzoites and bradyzoites. FIG. **4**A. MJM170 markedly reduces RH YFP tachyzoites in tissue culture robustly at low nanoM levels. (Standard curve left and effect on RH YFP, right panel). FIG. **4**B, FIG. **4**C. MJM170 markedly reduces EGS bradyzoites in cysts in vitro. Inhibition of cytochrome b Qi eliminates cysts in HFF infected with EGS. Without inhibitory compound in HFF (note, oval cyst with green border staining dolichos) and adjacent panel with inhibitory MJM170 compound (note absence of cysts with small amount of amorphous residual dolichos). MJM170 eliminated tachyzoites followed to 10 days of culture and bradyzoites in cysts in

vitro. Summary comparison of each of the compounds tested in vitro and their ADMET is in Table 2. Note improvement in solubility, properties amenable for compounds to cross blood brain barrier with new scaffold. FIG. **4**D. EGS transfected with stage specific reporters for fluors, red tachyzoite SAG1, Green bradyzoite LDH2.

FIG. **5**A-**5**D. MJM170 is also effective against RH and Prugneaud tachyzoites and Me49 bradyzoites, in vivo with translucent zebrafish providing a novel model with potential for scalable in vivo assays in which tachyzoites with fluorescent reporters and bradyzoites in cysts can be visualized efficiently. FIG. **5**A: 25 mg/kg daily MJM170 administered intraperitoneally eliminates active infection due to RH tachyzoites stably transfected with YFP in mice (RFU control vs rx with MJM 170, $p<0.004$). For the standard curve in the inset, RFU increase with increasing concentrations of fluorescent tachyzoites ($R^2=0.99$). FIG. **5**B. MJM 170 25 mg/kg daily reduces Type 2 parasites. FIG. **5**C. MJM 170 reduces cysts in mice infected 2.5 months earlier and treated for 17 days with 12.5 mg/kg daily then without compound for 3 days:cyst count of wet prep of brain homogenate. FIG. **5**D. Zebra fish can be used to visualize fluorescent tachyzoites and cysts in more chronic infections.

FIG. **6**A-**6**G. MJM170 targets apicomplexan cytochrome bc_1 Qi: modelling, yeast surrogate assays, target validation, co-crystallography and nanoM inhibition of *P. falciparum* and *T. gondii* FIG. **6**A. Modeling: MJM170 (yellow) modelled within cytochrome b Q_i site (grey) highlighting residues (green) involved in binding. FIG. **6**B. Mutations for yeast, *P. falciparum*, predicted for *T. gondii* and bovine enzyme. Relevant mutations are indicated by colored dots in Q_i domains on the bottom of the image of mitochondrion membrane for *S. cerevesiae* and *P. falciparum*, and where those amino acids are in *T. gondii*, human and bovine enzymes. Red dot marks G33A/V in Q_i domain of *P. falciparum*. FIG. **6**C. Cytochrome b mutants and sequence accession numbers. FIG. **6**D. MJM 170 inhibits wild-type but not mutant yeast. Compounds MJM 170 and ELQ 271 with wild type and mutant yeast validate predictions that M221 K/Q would create a steric clash and resistance. FIG. **6**E. MJM170 is a potent low nM inhibitor of *Plasmodium falciparum*. In Table 2, wild type *P. falciparum* also are tested and is inhibited at <50 nM by this scaffold. D6 is a drug sensitive strain from Sierra Leone, C235 is a multi-drug resistant strain from Thailand, W2 is a chloroquine resistant strain from Thailand, and C2B has resistance to a variety of drugs including atovaquone. Mutant G33V did not confirm prediction of a steric clash. FIG. **6**F-**6**G. MJM170 binds within Q_i site of bovine cytochrome bc_1 as shown by X-ray crystallography. FIG. **6**F. An omit Fo-Fc electron density map (green) at 5σ allows unambiguous positioning of MJM170 (magenta) within the Q_i site with the tetrahydroquinolone group near heme b_H (white) and diphenyl ether directed out of the channel. FIG. **6**G MJM 170 molecule is included into the structure, the 2Fo-Fc electron density map at 1σ (grey) allows placement of the planar head between heme b_H and Phe220 with the carbonyl group positioned in a polar region surrounded by Ser35 and Asp228.

FIG. **7**A-**7**C. MJM170 potently inhibits *P. falciparum* mitochondrial electron transport important for synthesis of pyrimidines, is modestly synergistic with atovaquone, additive with cycloguanil and antagonistic with Q_i inhibitor. FIG. **7**A. MJM170 is highly potent (Dd2, black curve, $EC_{50}=29.5$ nM) without cross-resistance in previously reported cytochrome b drug-resistant mutant parasite lines including ubiquinone reduction site mutants ($Dd2^{G33A}$ and $Dd2^{G33V}$, light blue and dark blue curves. respectively). Dose-response curve from representative assay. MJM170 cannot inhibit a parasite supplemented with a yeast cytosolic DHODH (scDHODH, green curve) demonstrating that its primary activity in *P. falciparum* is to inhibit electron transport necessary for pyrimidine biosynthesis. Inset Table. Dose-response phenotypes of a panel of *P. falciparum* cytochrome b mutant parasite lines. EC_{50} values were calculated using whole-cell SYBR Green assay and listed as mean±standard deviation of three biological replicates, each with triplicate measurements. FIG. **7**B., FIG. **7**C. Isobolograms with MJM170 plus atovaquone or cycloguanil or Q_i inhibitor BRD6323: FIG. **7**B. Combinations were with atovaquone (ATV) or cycloguanil (CYG) at multiple fixed volumetric ratios (10:0, 8:2, 6:4, 4:6, 2:8, and 0:10) in Dd2 parasites. Slight synergy observed with combinations of MJM170 and atovaquone while MJM170 and cycloguanil dosed in combination showed additive effect. Fractional inhibitory concentrations (FIC) for each drug were calculated and plotted. Shown is a representative isobologram for each combination of compounds. Table below lists FICs for each compound and ratio tested (values are mean from three independent assays±standard deviation). Synergy was defined as a combined FIC<1.0, addivity as FIC=1.0, and antagonism as FIC>1.0. FIG. **7**C. Isobologram Figure: MJM170 was tested in combination with previously reported reduction site inhibitor BRD6323 at multiple fixed volumetric ratios (10:0, 8:2, 6:4, 4:6, 2:8, and 0:10) in Dd2 parasites. Antagonism was observed with combinations of MJM170 and BRD6323, another bulky inhibitor of cytochrome bc, as opposed to synergy observed with oxidation site inhibitor atovaquone. Fractional inhibitory concentrations (FIC) for each drug were calculated and plotted. Representative isobologram of three independent assays is shown. Table below lists FICs for each compound and ratio tested (values are means from three independent assays±standard deviation). Definitions as in b.

FIG. **8**. Effects of compounds against RH—YFP. Graph is a representative example of an experiment testing two of the compounds against tachyzoites (RH—YFP). On the vertical axis is fluorescence in relative fluorescence units, where decrease in fluorescence compared to the DMSO control indicates parasite inhibition. On the horizontal axis are the different treatment conditions.

FIG. **9**. The results of a cytotoxicity assay. 10 μM solution of compound was compared to the DMSO control closest to 0.1% DMSO (0% DMSO) and the 50 μM solution was compared to the DMSO control closest to 0.5% DMSO (0.625% DMSO). The differences were not found to be statistically significant.

FIG. **10**A-**10**C. Effects of compounds against EGS. FIG. **10**A-FIG. **10**C) Graphs comparing the effects of JAG050 and JAG021 on EGS.

FIG. **11**. Binding assays show selectivity with binding to the bovine enzyme which is not as robust as has been seen with other cytbc inhibitors FIG. **12**A-**12**D. JAG21 is a mature lead that protects against *Toxoplasma gondii* tachyzoites and cures *Plasmodium bergheii* sprorozoites, blood and liver stages with oral administration of single dose 2.5 mg/kg and 3 doses protect at 0.5 mg/kg. Single dose causal prophylaxis in 5 C57BL/6 albino mice at 2.5 mpk dosed on day 0, 1 hour after intravenous administration of 10,000 *P. berghei* sporozoites. 3 dose causal prophylaxis treatment in 5 C57BL/6 albino mice at 0.6 mpk dosed on days −1, 0, and +1. A representative figure for higher dose (5 mg/kg) is shown, but all experiments with the amounts mentioned above had efficacy measured as cure measured as

survival, luminesence and parasitemia quantitated by flow cytometry are similar to these.

FIG. **13**. Tafenoquine and JAG21 are both needed to contain RPS13Δ. One additional mouse in the tafenoquine and JAG21 died outside the time on this graph others remained healthy.

FIG. **14**A-**14**D. Serum biomarkers from boys with active brain disease due to *Toxoplasma* reflect infection and neurodegeneration. FIG. **14**A. Tabular clinical summary: Three pairs of children, matched demographically; one in each pair had severe disease and one mild or no manifestations. One pair dizygotic, discordant twins. Each ill child had new myoclonic or hypsarrythmic seizures. Two children had T2 weighted abnormalities on brain MRIs similar to active inflammatory and parasitic disease in murine model[8] FIG. **14**B-FIG. **14**D. Protein and miR serum biomarkers: Panel of nanoproteomics and miR sequencing performed on serum obtained at time of new illness. MiRNA concentration measured and difference in concentration graphed. Abundance of peptides measured. Note: Presence of markers of neurodegeneration, inflammation, and protein misfolding include clusterin, diminished ApoJ, serum amyloid, and oxytocin in ill children compared with their healthy controls.

DETAILED DESCRIPTION OF THE INVENTION

All references cited are herein incorporated by reference in their entirety. Within this application, unless otherwise stated, the techniques utilized may be found in any of several well-known references such as: *Molecular Cloning: A Laboratory Manual* (Sambrook, et al., 1989, Cold Spring Harbor Laboratory Press), *Gene Expression Technology* (Methods in Enzymology, Vol. 185, edited by D. Goeddel, 1991. Academic Press, San Diego, CA), "Guide to Protein Purification" in *Methods in Enzymology* (M. P. Deutscher, ed., (1990) Academic Press, Inc.); *PCR Protocols: A Guide to Methods and Applications* (Innis, et al. 1990. Academic Press, San Diego, CA), *Culture of Animal Cells: A Manual of Basic Technique,* 2nd Ed. (R. I. Freshney. 1987. Liss, Inc. New York, NY), *Gene Transfer and Expression Protocols*, pp. 109-128, ed. E. J. Murray, The Humana Press Inc., Clifton, N.J.), and the Ambion 1998 Catalog (Ambion, Austin, TX).

As used herein, the singular forms "a", "an" and "the" include plural referents unless the context clearly dictates otherwise. "And" as used herein is interchangeably used with "or" unless expressly stated otherwise.

As used herein, the amino acid residues are abbreviated as follows: alanine (Ala; A), asparagine (Asn; N), aspartic acid (Asp; D), arginine (Arg; R), cysteine (Cys; C), glutamic acid (Glu; E), glutamine (Gln; Q), glycine (Gly; G), histidine (His; H), isoleucine (Ile; I), leucine (Leu; L), lysine (Lys; K), methionine (Met; M), phenylalanine (Phe; F), proline (Pro; P), serine (Ser; S), threonine (Thr; T), tryptophan (Trp; W), tyrosine (Tyr; Y), and valine (Val; V).

All embodiments of any aspect of the invention can be used in combination, unless the context clearly dictates otherwise.

In one aspect, the invention provides cell lines infected with an apicomplexan parasite, wherein the apicomplexan parasite genome comprises a gene encoding an Apetela 2 IV-4 protein with an M=>I modification at residue 570 ("AP2 IV-4 M570I") compared to its orthologous gene on the reference *T. gondii* ME49 strain (gene ID: TGME49_318470). As described in the examples that follow, Apetela 2 (AP2) IV-iv is known to be a bradyzoite gene expression repressor[56], and the AP2 IV-4 M570I mutant results in an apicomplexan parasite that remains as a bradyzoite in tissue cultures passaged extensively, capable of producing oocysts when administered to cats definitively proving its true bradyzoite phenotype.

As further described in the examples that follow, critical flaws and limitations of available methods and models for developing medicines to cure apicomplexan infections, such as *T. gondii* infections, include lack of in vitro culture systems for cysts and scalable, easy to use animal models for screening compounds. The cell lines of this aspect of the invention unexpectedly possess a true, dormant parasite phenotype in tissue culture and can be used, for example, to screen for drugs that can be used to treat apicomplexan parasitic infections, as well as a research tool for studying apicomplexan parasites in the dormant phenotype. The cell lines can be used, for example, as a model of bradyzoite infection. A generalized apicomplexan life cycle comprises a rapidly growing tachyzoite and slow-growing, latent bradyzoite that forms tissue cysts (i.e.: dormant phenotype). Such dormant parasites are present in the brains of 2 billion persons worldwide across their lifetimes and are incurable. Quite remarkably, the inventors have discovered that in human cells this encysted parasite turns on host cell pathways important for altering ribosomal function, miss-splicing of transcripts, oxidative pathways, and, those pathways found to be altered in Alzheimer's and Parkinson's diseases. Extensive details on the model are described in the examples that follow.

In one embodiment, the apicomplexan parasite genome comprises a gene encoding AP2 IV-4 M570I that further differs from its orthologous gene on the reference *T. gondii* ME49 strain (gene ID: TGME49_318470) in encoding the amino acid sequence GGNRPHYHVAKQEWRVRYYMNGKRKMRTYSAKFYGYETAIITMAEDFAHYVDKH E (SEQ ID NO: 1) beginning at residue 821. In a further embodiment, the gene encoding AP2 IV-4 M570I encodes the following amino acid sequence, or functional equivalents thereof:

(SEQ ID NO: 2)

MAAPAPSAEARPAKRRCFPLPRETPVSSEDETRKTLQHDTLGCLPRSS

SGQPELAAASAASQVGHLSSAALLQLVQTQSAGGVPQAVLRNLFSSIH

RNPKPLPANALAATPNSSLYASLTSLSSAAALPGAGPAYSQAPSPASA

DLLQSEQFGSAAKNPSPNEASPILALLGEAARAATTPRTVPALSAVCP

AASSGVSLPSASDTLALAQSSLSSSTGCASDVKASRPEEHPAFASGTA

NRQSLLQALLLSTAPLAFSGPSLSSASTTLPASSGAVSSRNAGAYQFE

RLLQAEAAKVKALLPNATSKSMSQSSVPQRDLTRKTSLFPDPRGLSAD

DASRRYNTRGANSGGAGLRRGTGVHATTEQSGALDAGERTRPFGAGED

ESAQGKPDSRGRQRPGALDASNILGLLAAFQPSQAPAIRDLSAPSHLS

AAATGALPLTASFTASALASSQCLPAGTPASSSASPPFSEVLSTTEES

STTKETDASASTLLAFLQKYSAVSGLGGASDFLGQLQGKSSLPPLSLA

EPSSALPSSFLGGSDGGTIDTRNGNGEKTTPPIHLFQSAFRIPSPSQQ

NLLDALLASSCTTATSRSDGSGNLGCPVVDERNAKLAGPAHPLPCSFP

QISSSSGEPGRKTGGRVHRQGTSQSGGRVRSGKNGGSAAPPRQSSSEN

VPSTPTVSSHEAPHRAGFPSQTPYELSASPSHQLDLLRLGAFLGGAGK

-continued

QDASVHSDETGTLSGEPSHRSCSLSRGLTQESVLQLSDTTSTSREGEP

NEPSQGCVNVAASLPAFGPQPSSGAAKAREGRRGAGGAGAAPPVPLRA

DVTL**GGNRPHYHVAKQEWRVRYYMNGKRKMRTYSAKFYGYETAHTMAE**

DFAHYVDKHEALPDSMMMTAMMLQAQANSAASSGQTVPLARGIRASSA

SAGAGGHVSKSATKGSVAASSEGSTSMGSDATRSQEGEAAELCPLAAG

LSRPLASMHSAAGNAVAQGRQESKEEAPGGQAWFGEPGKFRASSEAAL

CGSGSSAEGRDGHESEVLWATLGKVHDASQGKKIKPEKPLTVARGRLA

LGAEDKSQNLGVDLGDSGGAQGLPGVRQPRQMKNSEECSLRDSDKGMA

LSKRFGFLPSQTPSCDSMTLPFPGGFDALSLSSALSSCASLPVAHEGN

NFQKGHTGDIVALASQSGTQRPASVVLSRDANVSGSSPSHPTWQREGA

AVSGRADEFSSLSVTPSTVPLSSFTMEDIKGEEGDPSRRFALVGESMK

NVSAPEVQALFPTSSIANAELLPVDFLHSNSCSADKLESSIPRGLAGN

NPSMTATAVAATAVSHQIFDTITLFGEFLREFAKEKVNEFHEYGLEAS

PLTVEASPEVSLFGKATFGRCPVAGGSTPAGISKMSGETLSGLSASEL

SLVSARTNTTTGEEQFALARGLFPGDSEGDRDEKKPQLSQQELLVLSH

ALVNLTSSTYVLMHTLKASLSKSTEAVQLHQPLLEAASEAKATDEAKT

REEQESSECDHEYPPGSSLEATTGALPFRLSPALSASSKDLPSLSASA

SLESVTPFAGLPLEEGTLSASVGLASSDDEHDTSLLFKTEAAKKRSLF

STAADGDESRTYNDGLGQPMEEEIRSCVSTSCGEAVATTTLSAIGPGT

GASGALLDSESRESLGEKPGAALRAGAHTPAPSRAPTPSRTFSFTSSS

TATSAALLCDSNVVHEKLSAQGKDSEAGERKGDSEKEEEVEMWKEEDE

EVQRCTGSAETDSTEATRGEEAWRRGKQSEKKPSVITTALNLLETHRH

LALTISQLKRPVAQQLRFILPIAAPQLLPCILPPASFQGTGESGDGKA

EAEAKGSSSLGQVLETALGHGTRLAPSASAMVPPRKDEAASAVPEAKT

LTGLANAGVTREAASRTLEAEQVSRKRSREEVVDSETAGDEGDMENVP

ETRDGTTRPGSRQYDTSPSNDGTKPPATAKSRVIRDQAALERLLLAPF

QDTPTCSCTDRPCPCDRQQVADMIYLFYAVPARQQAESSKEGSTQRLQ

FAARDTNERKDARTGEETQGGETEAKEVIRDPEERGVCEGSSSQNAHT

QFDAETASSSMSSDPRADKESNAQDAHMADKTSFVSDLPQPSGEFAPS

LLSETSLDVAMADSRGTPSEIHGFFTRSDEQKRASFSSSSLLAAGHAV

ASFSSSLAGVVSGAGERRECAGPSLGDLSTIGLLSLSYPAMLAFILPL

QSLLHTVSGMILTLHKKLIHRFICAHLRLVLDDDMRRPAGGALKSRGA

HGDTEAAEAQVERRRREHEREETTNLAIGYREGNAEAANTFPLVDTVS

SLLSPGSLRQENSEVERRDNDEERLELITGIARESPKPSEKDSVSPFL

STAPCPGTEAESSDCSASSACSGTPTEGTEGGETGDIASFLSPSGEVK

QTIMLA

The mutations at residue 570 and beginning at residue 821 compared to the Apetela 2 IV-4 orthologous protein encoded by the reference *T. gondii* ME49 strain (gene ID: TGME49_318470) are presented in bold font and underlining in the above sequence. A "functional equivalent" is a gene encoding an Apetela 2 IV-4 protein that is at least 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identical to the amino acid sequence provided above, includes the noted mutations at residue 570 and beginning residues 821-874, and that does not represses bradyzoite gene expression in a tachyzoite in an apicomplexan parasite.

As used here, "apicomplexan parasites" are a phylum of the kingdom Protista (formerly a division of protozoa called *Sporozoa*); named for a complex of cell organelles (apical microtubule complex) at the apex of the sporozoite form that can penetrate host cells. It includes, but is not limited to, the medically important genera *Plasmodium, Toxoplasma, Cryptosporidium*, and *Isospora*. Thus, in one embodiment, the apicomplexan parasite is selected from the group consisting of *Plasmodium, Toxoplasma, Cryptosporidium*, and *Isospora*. In a specific embodiment, the apicomplexan is a wild type, mutant, or recombinant *Toxoplasma gondii* strain. In various further embodiments, the apicomplexan parasite is *Toxoplasma gondii* of clade B.

In another embodiment, the apicomplexan parasite is a GO arrested parasite, including but not limited to s *Toxoplasma gondii* strain RPS 13 delta (Hutson et al., PLoS One. Nov. 22, 2010; dx.doi.org/10.1371/journal.pone.0014057). This cell line can be used, for example, to identify companion compounds for THQ that eliminate the GO arrested stage of *T. gondii* along with the active and the slowly growing bradyzoite stage.

In another embodiment, the apicomplexan parasite is *Toxoplasma gondii* strain EGS (ATCC® Number:PRA-396™). As described in the examples that follow, the EGS strain was extensively characterized in vitro to show that true cysts develop, making the EGS strain especially useful for drug development.

In all embodiments of this aspect of the invention, the cell line can be any suitable cell line capable of supporting apicomplexan parasitic infection, including but not limited to mammalian cells (mouse, rat, human, etc.), zebrafish cells, etc. In one specific embodiment, the cell line is a human cell line; exemplary human cell lines for use in this aspect of the invention include, but are not limited to fibroblasts, stem cells, neurons, monocytes, and ocular cells 9 including primary human cells). As described in the examples that follow, the apicomplexan parasites form cysts in the host cell lines that enlarge over time and then destroy host cell monolayers as single cell organisms. As such, the cell lines of the invention are extremely useful as in vitro models of apicomplexan parasite infection. Such models can be used, for example, to test for candidate compounds that inhibit cyst formation and/or destruction of host cell monolayers; such candidate compounds would be useful in treating apicomplexan parasite infection.

In another embodiment, the apicomplexan parasite genome is recombinantly engineered to express a reporter polypeptide, including but not limited to fluorescent or luminescent proteins. This embodiment permits ready visualization of the parasite and facilitates automated quantitative analysis. In one embodiment, the reporter polypeptide is operatively linked to a promoter that is activated in the bradyzoite stage or a promoter that is activated in the merozoite stage. Any suitable promoter that is activated in the bradyzoite stage or merozoite stage may be used. In one embodiment the promoter that is activated in the bradyzoite stage is the *T. gondii* BAG1 encoding gene promoter or a functional equivalent thereof. The BAG1 promoter sequence can be obtained as disclosed in Bohne et al., Molecular and Biochemical Parasitology 85 (1997) 89-98. In one embodiment, the BAG promoter comprises SEQ ID NO:3, or functional equivalent thereof, from the *T. gondii* VEG strain.

A "functional equivalent" of the BAG1 promoter is a promoter from any strain of *T. gondii* that promotes expression of BAG1 in the VEG strain, as well as an promoter nucleic acid sequence that is 50%, 60%, 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identical to SEQ ID NO:3 and drives expression of a BAG1 gene in a *T. gondii* strain.

As described in the examples that follow, the cell lines of the invention can be administered to/ingested by non-human animal models to provide an in vivo model of apicomplexan parasitic infection that develop the classic, gold standard bradyzoite phenotype of producing oocysts. This, in another aspect, the invention provides non-human animal models of apicomplexan parasitic infection, comprising a non-human animal that has ingested or otherwise comprises the cell line of any embodiment or combination of embodiments of the invention. In one embodiment, the non-human animal produces oocysts. Any suitable non-human animal model that produces oocysts can be used, including but not limited to cats.

In another aspect, the invention provides non-human animal models of apicomplexan parasitic infection, comprising a non-human animal that has ingested or otherwise comprises oocysts produced by the non-human animal model that has ingested or otherwise comprises the cell line of any embodiment or combination of embodiments of the invention. As described in detail in the examples that follow, oocysts given to non-human animal models such as mice created an illness and histopathology phenotypically characteristic for typical, virulent parasites causing dose related proliferation of the parasite (exemplified by *T. gondii*) with necrosis in terminal ileum, pneumonia at 9-10 days, with brain parasites by 17 days and dose-related mortality. Thus, the non-human animal models of this aspect of the invention are particularly useful in screening for drugs to treat the effects of apicomplexan parasite infection.

As will be understood by those of skill in the art, the oocysts do not need to be isolated prior to ingestion; they may be present, for example, in tissue (including but not limited to brain tissue) taken from the non-human animals that have ingested or otherwise comprise the cell lines of the invention, which then produce oocysts. Any suitable non-human animal model can be used, including but not limited to mice, rats, cats, zebrafish, non-human primates, cattle, sheep, and pigs. In one specific embodiment, the non-human animal is a mouse.

In another aspect the present invention provides methods of identifying compounds for treating an apicomplexan parasitic infection, comprising contacting one or more test compounds to the cell line of any embodiment or combination of embodiments of the invention, wherein those positive test compounds that reduce bradyzoite cyst amounts in the cell line are candidates to treat an apicomplexan parasitic infection. As disclosed above, the cell lines of the invention unexpectedly possess a true, dormant parasite phenotype in tissue culture and can be used to screen for drugs that can be used to treat apicomplexan parasitic infections. Any reduction in bradyzoite cyst amounts in the cell line indicates that the test compound may be useful for treating an apicomplexan parasitic infection. In various embodiments, positive test compounds are those that reduce bradyzoite cyst amounts by at least 5%, 10%, 15%, 20%, 25%, 50%, 75%, 90%, or more.

In another aspect the present invention provides methods of identifying compounds for treating an apicomplexan parasitic infection, comprising administering one or more test compounds to the animal model of any aspect or embodiment of the invention, wherein those positive test compounds that reduce one or more symptoms of the infection and/or reduce parasitic titer in the animal model are candidates to treat an apicomplexan parasitic infection. In one embodiment, those positive test compounds that reduce oocyst production and/or reduce bradyzoite cyst amounts in the animal model are candidates to treat an apicomplexan parasitic infection. Any reduction in oocysts production and/or bradyzoite cyst amounts indicate that the test compound may be useful for treating an apicomplexan parasitic infection. In various embodiments, positive test compounds are those that reduce oocysts production and/or bradyzoite cyst amounts by at least 5%, 10%, 15%, 20%, 25%, 50%, 75%, 90%, or more.

In one embodiment of any of the methods of identifying compounds for treating an apicomplexan parasitic infection of the invention, positive test compounds are candidates for treating *Toxoplasma gondii* or *Plasmodium falciparum*: infection, including drug resistant strains and or other plasmodial infections. The methods can be used to test any suitable type of candidate compound, including but not limited to polypeptides, antibodies, nucleic acids, organic compounds, etc. Treatment effects of the test compounds may be assessed relative to a suitable control, such as the cell lines or non-human animal models of the invention that are not treated with the test compound. It is well within the level of those of skill in the art to determine a suitable control in light of the teachings herein.

In one specific embodiment, the cell line, or non-human animal model that has ingested or otherwise comprises the cell line, comprises a G0 arrested parasite (such as RPS 13 delta) and is used to identify companion compounds for tetrahydroquinolones (THQ) that eliminate the G0 arrested stage of an apicomplexan parasite, such as *T. gondii*, along with the active and the slowly growing bradyzoite stage.

RPS 13 Δ is a genetically engineered conditional knockout parasite that has a unique transcriptome documenting its G0 state, G1 arrest in the absence of tetracycline but grows normally in the presence of tetracycline which removes the repressor from the promoter. The method may utilize, for example, a system that when there is no anhydrotetracycline to remove the engineered tetracycline responsive repressor from the 4 tetracycline response elements engineered in tandem in the promoter; these parasites persist in tissue culture for long times (months). This embodiment is based on the observation that this conditional knockout RPS13 delta parasite when it is in its arrested in G1 state is not susceptible to the effect of any inhibitors that effect processes essential to the tachyzoite or bradyzoite form including those tested in vitro so far. In this embodiment the conditional knockout RPS13 delta parasite is amenable to testing inhibitors of hypnozoite like organisms by culturing them without tetracycline in the presence of the compound and determining whether any parasites can be rescued by adding tetracycline to determine whether they are still capable of persisting and becoming tachyzoites that grow rapidly in the presence of tetracycline. Furthermore, the methods may comprise testing RPS13 delta in mice, and involves the observations that when RPS13 delta is administered to wild type mice without tetracycline the RPS13 delta parasite induces a protective immune response as a vaccine dependent on interferon gamma and has no adverse effect on the mice, nor can it be rescued with tetracycline or inhibitors of iNOS (intracellular nitrogen oxide synthase) such as LNAME (L-N^G-Nitroarginine methyl ester) which abrogate effects of interferon gamma after 7 days. For example, that in interferon gamma knock out mice or mice

treated with antibody to interferon, RPS13 delta is lethal for the mice, where the attenuated organism persists, can be observed, until mice succumb slowly. Another example may be a SCID mouse or a steroid treated mouse that is more susceptible due to immune compromise.

In another embodiment, RPS13 delta can be used to test compounds in vitro against this G0 truly dormant stage. and against compounds that can target the hypnozoite state but that must be metabolized in the liver to produce toxic electron containing compounds in vivo.

In all these embodiments, the methods can be used to determine if compounds have parasiticidal effects on hypnozoite forms and be used in conjunction with THQ compounds, such as tafenoquine, known in primate models and in humans as the only compound besides primaquine which has a lethal effect on the malaria hypnozoite in primates or in humans. However, tafenoquine is not active against actively proliferating organisms and requires a compound that is effective against more rapidly and slowly growing forms to produce radical cure of malaria.

In another embodiment, the THQ compound may be primaquine, a compound that has a similar effect when metabolized and is ineffective against *T. gondii* tachyzoites in tissue culture and can therefore be utilized in the same manner as tafenoquine to inhibit the hypnozoite form of apicomplexan parasites to produce radical cure.

Compounds identified using the methods of these various embodiment of the invention can be used together the THQ compound(s) to treat/cure the active tachyzoite, the slowly growing bradyzoite, and the G0 arrested hypnozoite-like phase of the apicomplexan infections the active tachyzoite, the slowly growing bradyzoite, and the G0 arrested hypnozoite-like phase of the apicomplexan infections

As described in detail in the examples that follow, the inventors carried out transcriptome analysis of both the apicomplexan parasite and the host cells after parasite infection of the host cells. The resulting transcriptomes provide a signature for apicomplexan parasites (such as *T. gondii* strains EGS), which helps in identification of targets for drug development, as well as a signature for infected host cells (such as human foreskin fibroblasts (HFF) human monocytic cells (ex: MM6), and human primary neuronal stem cells (NSC)), which helps in identification of targets for treating apicomplexan infection. As shown in the examples, EGS transcription was influenced by host cell type (FIGS. **1**A-**1**D). Transcriptomics using host mRNA and miR profiling of EGS cultures in MM6, and NSC cells for 18 hours demonstrated that this parasite modulates host transcripts involved in protein misfolding, neurodegeneration, endoplasmic reticulum stress, spliceosome alteration, ribosome biogenesis, cell cycle, epilepsy, and brain cancer among others (FIGS. **1**A-**1**D). The number of genes significantly up or down regulated in MM6 and NSC cells compared to uninfected controls are depicted in FIGS. **1**A-**1**D. Overexpressed genes differ from those of GT1, ME49 and VEG tachyzoite-infected human NSC cells, but modify the same or connected pathways. Hsa-miR-708-5p was the most affected miRNA (down-modulated) by EGS (FIG. **1**D. miR-708-5p is a regulator that promotes apoptosis in neuronal and retinal cells, which could maintain a niche for EGS-like encysted bradyzoites to persist.

The genes identified in the host transcriptome study are thus targets for anti-parasite therapy, as well as markers of apicomplexan parasite infection (such as *T. gondii* infection). In various embodiments, the invention may thus comprise methods for treating an apicomplexan parasite infection (such as a *T. gondii* infection), comprising administering to a subject in need thereof an amount effective to treat the infection of an inhibitor (of up-regulated genes) or an activator (of down-regulated genes) of 1 or more (i.e.: 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, or more) of the up-regulated genes listed discussed herein (protein encoding genes or miRNA). In various non-limiting embodiments, the inhibitor is selected from the group consisting of a target-specific inhibitory antibody, aptamer, siRNA, shRNA, antisense oligonucleotide, or small molecule.

In one embodiment, the target is miR-708-5p. In various further embodiments the targets are one or more of the genes listed in Table 1 below, or as discussed herein, which provide a more complete list of up-regulated or down-regulated genes involved in specific host cell pathways or indications. The table shows up-regulated or down-regulated genes involved in specific host cell pathways or indications ("KEGG pathway"), such as systemic lupus erythematosus, (SLE), Parkinson's disease, etc. In an exemplary embodiment, the methods comprise administering to the subject in need thereof an inhibitor (of up-regulated genes) or an activator (of down-regulated genes) of one or more (1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, or all 13) of genes HIST2H2AA3, HIST1H2AC, HIST1H2BC, ACTN4, SNRPD3, ELANE, C1R, HIST1H2BK, H2AFZ, SNRPB, H2AFX, CTSG, and HIST1H4H, to treat apicomplexan parasite infection-associated SLE. One of skill in the art will understand from the table that the methods may be used to treat apicomplexan parasite infection-associated Parkinson's, Huntington's disease, Alzheimer's disease, etc. using an inhibitor (of up-regulated genes) or an activator (of down-regulated genes) against 1 or more (i.e., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, or more, such as all) of the target genes listed for the specific indication. Similarly, one of skill in the art will understand from the table that the methods may be used to treat apicomplexan parasite infection-associated disorders relating to ribosomal assembly, spliceosome assembly, oxidative phosphorylation, etc. using an inhibitor (of up-regulated genes) or an activator (of down-regulated genes) against 1 or more (i.e., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, or more, such as all) of the target genes listed for the specific indication.

TABLE 1

KEGG id	KEGG pathway	P-value	Genes
hsa03010	Ribosome	4.6E-07	RPL35A, RPL36A, RPL35, RPS9, RPL27, RPS27L, RPL38, RPS25, RPS27. RPL30, RPL23, RPS29, RPL21, RPS20, RPS21, UBA52, RPL36AL.
hsa05322	Systemic lupus erythematosus	0.00082	HIST2H2AA3, HIST1H2AC, HIST1H2BC, ACTN4, SNRPD3, ELANE, C1R, HIST1H2BK, H2AFZ, SNRPB, H2AFX, CTSG, HIST1H4H
hsa04621	NOD-like receptor signalling pathway	0.01458	IL6, CCL2, XIAP, NFKBIB, NFKBIA, TNFAIP3, BIRC3, CCL5

TABLE 1-continued

KEGG id	KEGG pathway	P-value	Genes
hsa03040	Spliceosome	4.1E−06	SNRPA1, CCDC12, MAGOH, SNRPD3, LSM6, LSM7, SNRPB2, SNRPD2, PPIH, SNRPB, PQBP1, SYF2, LSM5, U2AF1, MAGOHB, THOC2, SNRPF, SNRPE, SNRPG
hsa00190	Oxidative phosphorylation	6.4E−16	ATP5E, NDUFB4, ATP6V0E1, NDUFB6, NDUFB9, COX7C, ATP6V1G1, ATP5G1, UQCRQ, NDUFB1, NDUFB2, NDUFS5, NDUFS4, ATP5L, NDUFS3, COX17, ATP5H, ATP5J, NDUFA4, NDUFA5, NDUFA2, NDUFA3, COX7A1, NDUFA6, COX8A, NDUFC2, NDUFC1, NDUFA1, ATP6V1F, NDUFV2, COX6A1, UQCRB
hsa05012	Parkinson's disease	2.4E−14	ATP5E, NDUFB4, NDUFB6, NDUFB9, COX7C, ATP5G1, UQCRQ, NDUFB1, NDUFB2, NDUFS5, NDUFS4, HTRA2, NDUFS3, ATP5H, ATP5J, NDUFA4, NDUFA5, NDUFA2, NDUFA3, COX7A1, NDUFA6, COX8A, NDUFC2, NDUFC1, UBE2L3, NDUFA1, VDAC3, NDUFV2, COX6A1, UQCRB
hsa05016	Huntington's disease	2.2E−13	ATP5E, NDUFB4, POLR2F, NDUFB6, POLR2K, NDUFB9, POLR2J, COX7C, ATP5G1, UQCRQ, NDUFB1, NDUFB2, NDUFS5, NDUFS4, TGM2, CREB3L1, NDUFS3, ATP5H, ATP5J, NDUFA4, NDUFA5, NDUFA2, NDUFA3, COX7A1, NDUFA6, COX8A, NDUFC2, NDUFC1, NDUFA1, VDAC3, SOD2, NDUFV2, COX6A1, UQCRB
hsa05010	Alzheimer's disease	9E−11	ATP5E, NDUFB4, NDUFB6, NDUFB9, COX7C, ATP5G1, UQCRQ, NDUFB1, NDUFB2, NDUFS5, NDUFS4, PPP3CA, NDUFS3, ATP5H, ATP5J, NDUFA4, NDUFA5, NDUFA2, NDUFA3, COX7A1, NDUFA6, COX8A, NDUFC2, NDUFC1, ITPR3, NDUFA1, NDUFV2, COX6A1, UQCRB
hsa04623	Cytosolic DNA-sensing pathway	0.0077	MAVS, IL6, POLR3K, NFKBIB, IRF7, NFKBIA, POLR1C, CCL5

In one embodiment, the invention provides methods for treating an apicomplexan parasite infection, comprising treating a subject with an apicomplexan parasite infection an amount effective to inhibit activity or expression from the apicomplexan parasite of one or more proteins as discussed herein.

In another embodiment, the invention provides methods for identifying a compound to treat an apicomplexan infection, comprising identifying a compound that inhibits activity or expression of one or more proteins as discussed herein from an apicoimplexan parasite present in an infected host cell.

Certain proteins as discussed herein are believed to be particularly important for apicomplexan parasite bradyzoite development and/or survival in the host. Thus, targeting expression and/or activity of these proteins from the apicomplexan parasite will be effective to inhibit bradyzoite development and/or survival in the host.

EGS transcripts in HHF, MM6, and NSC cells were enriched for genes transcribed in bradyzoites, including known bradyzoite transcripts, certain Apetela 2s and cytochrome b and other cytochromes. Among transcripts with the most increased fold change in EGS across all three cell lines were: cytochrome b; cytochrome c oxidase subunit III subfamily protein; apocytochrome b; cytochrome b, putative; and cytochrome b (N-terminal)/b6/petB subfamily protein. Other over-expressed genes include bradyzoite transcription factor AP2IX-9 and plant-like heat-shock protein BAG1 (FIG. **2**A). The up- or down-regulated genes identified in the parasite transcriptome study are thus targets against which to identify drugs for anti-apicomplexan parasite (such as *T. gondii*) therapy, by identifying test compounds that reduce expression of over-expressed genes, or promote expression of down-regulated genes. In various embodiments, positive test compounds are those that reduce expression (for up-regulated genes), or decrease expression (for down-regulated genes) of 1 or more (i.e.: 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 50, 100, or more) of the up-regulated apicomplexan parasite genes as discussed herein after host cell infection by at least 5%, 10%, 15%, 20%, 25%, 50%, 75%, 90%, or more. The drug screening assays may employ the cell lines and non-human animal models of the present invention. In one embodiment, the methods comprise identifying test compounds that reduce expression from the apicomplexan parasite after cell infection of 1 or more of cytochrome b; cytochrome c oxidase subunit III subfamily protein; apocytochrome b, cytochrome b, putative, cytochrome b (N-terminal)/b6/petB subfamily protein, bradyzoite transcription factor AP2IX-9 and plant-like heat-shock protein BAG1.

In another aspect, the invention provides compositions comprising a plurality of isolated probes that in total selectively bind to at least 2, 3, 4, 5, 6, 7, 8, 9, 10, 15, 20, 25, 30, 35, 40, 45, 100, 150, 200, 250, 500, or all of certain markers as discussed herein, complements thereof, or their expression products, or functional equivalents thereof wherein at least 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, or all of the probes in total are selective for markers that are upregulated in the EGS strain of *T. gondii* after infection of human fibroblasts, neuronal stem cells or monocytic lineage cells.

Functional equivalents are allelic variants of the recited marker from other *T. gondii* strains. In one embodiment, the

markers include two or more of apetela 2 transcription factors, cytochrome b, cytochrome oxidase, or functional equivalents thereof. The markers may further comprise one or more of enolase 1, lactate dehydrogenase 2, bradyzoite antigen 1 and cyst wall protein, or functional equivalents thereof. In another embodiment,

In a further aspect, the invention provides a plurality of isolated probes that in total selectively bind to at least 2, 3, 4, 5, 6, 7, 8. 9, 10, 15, 20, 25, 30, 35, 40, 45, 100, 150, 200, 250, 500, or all of certain markers as discussed herein, complements thereof, or their expression products, or functional equivalents thereof, wherein at least 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, or all of the probes in total are selective for markers that are upregulated in human fibroblasts, neuronal stem cells or monocytic lineage cells after infection with *T. gondii*, including but not limited to infection with the EGS strain of *T. gondii*.

In one embodiment of each of these aspects, the plurality of isolated probes comprises polynucleotide probes. In another embodiment, the plurality of isolated probes comprises antibody probes. In all of the above embodiments, the isolated probes can be labelled with a detectable label. Methods for detecting the label include, but are not limited to spectroscopic, photochemical, biochemical, immunochemical, physical or chemical techniques. Any suitable detectable label can be used.

The compositions can be stored frozen, in lyophilized form, or as a solution. In one embodiment, the compositions can be placed on a solid support, such as in a microarray or microplate format; this embodiment facilitates use of the compositions in various detection assays.

The compositions of the invention can be used, for example, to test patient samples for up-regulation or down-regulation of the one or more markers disclosed in the figures, to assist in diagnosing a subject as having an apicomplexan parasite infection (such as *T. gondii*) or to monitor treatment of a subject receiving therapy for an apicomplexan-associated disorder. In one embodiment, such methods comprise testing the patient samples for increased expression of at least 1, 2, 3, 4, 5, 6, or all 7 of apetela 2 transcription factors, cytochrome b, cytochrome oxidase, enolase 1, lactate dehydrogenase 2, bradyzoite antigen 1 and cyst wall protein, or functional equivalents thereof.

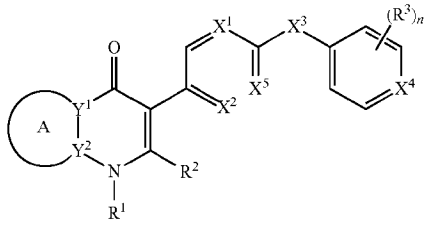
In one embodiment, the transcriptome provides the signature of cytochrome b as an important part of the bradyzoite transcriptional pathways and a signature that demonstrates effective inhibition of cytochrome b with abrogation of the signature when treatment is with an inhibitor of cytochrome b, which when used early after infection can confirm selectivity of compound. Cytochrome b functions for pyrimidine synthesis in *Plasmodium falciparum* so that it will be synergistic or additive in effect with inhibitors of DHODH.

The invention thus also provides pathway to improved inhibitors of cytochrome b through co-crystallography that defines the chemical space and pi stacking which facilitates design of improved medicines and their delivery into tachyzoites and bradyzoites using molecular transporters such as octaargine, or carbonate, and also improves their solubility and access to encysted bradyzoites.

In various embodiments, the methods for monitoring treatment of an apicomplexan parasitic infection (such as a *T. gondii* infection), comprising monitoring expression, protein in serum or plasma, and/or activity of 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, or all of certain markers as discussed herein (such as a human subject) being treated for an apicomplexan parasitic infection, wherein a decrease or increase in expression and/or presence and/or activity of the one or more markers indicates that the treatment is effective. In one exemplary embodiment, infection is in the subject's brain or other neurologic tissue.

In another aspect, the present disclosure provides compounds having the structure of Formula (I):

(I)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof,

wherein

ring A combines with Y^1 and Y_2 to form a C_{3-7}cycloalkenyl or heteroaryl ring,

wherein the C_{3-7}cycloalkenyl or heteroaryl is optionally substituted by halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR, cyano or phenyl;

Y^1 is C or N;

Y^2 is C or N;

X^1 is C(R^{x1}) or N,

wherein R^{x1} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^2 is C(R^{x2}) or N,

wherein R^{x2} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^3 is O, N(R), S or C_{1-3}alkyl;

X^4 is C or N;

X^5 is C or N;

R^1 is hydrogen or C_{1-3}alkyl;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR or —C(O)OR;

n is 0, 1, 2, 3 or 4;

each R^3 is independently halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5;

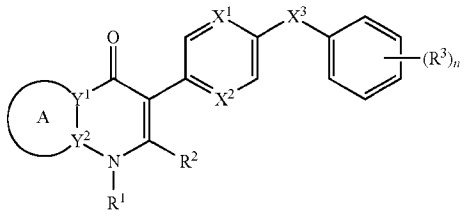
or two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane; and

each R is independently hydrogen or C_{1-3}alkyl.

The compounds of the invention have been demonstrated in the examples herein as useful, for example, in treating diseases associated with apicomplexan parasite infection.

In some embodiments, the compounds are of Formula (Ia):

(Ia)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof,

wherein

ring A combines with Y^1 and Y_2 to form a C_{3-7}cycloalkenyl or heteroaryl ring,

wherein the C_{3-7}cycloalkenyl or heteroaryl is optionally substituted by halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR, cyano or phenyl;

Y^1 is C or N;

Y^2 is C or N;

X^1 is $C(R^{x1})$ or N,

wherein R^{x1} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^2 is $C(R^{x2})$ or N,

wherein R^{x2} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^3 is O, N(R), S or C_{1-3}alkyl;

R^1 is hydrogen or C_{1-3}alkyl;

R^2 is hydrogen, C_{1-3}alkyl or —C(O)OR;

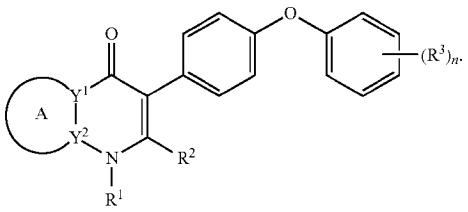
n is 0, 1, 2, 3 or 4;

each R^3 is independently halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5; and

each R is independently hydrogen or C_{1-3}alkyl.

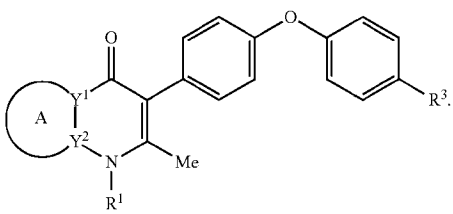
In some embodiments, the compounds are of Formula (Ib):

(Ib)



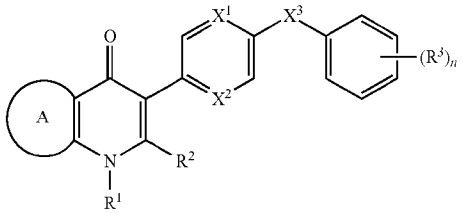
In some embodiments, the compounds are of Formula (Ic):

(Ic)



In some embodiments, the compounds are of Formula (II):

(II)

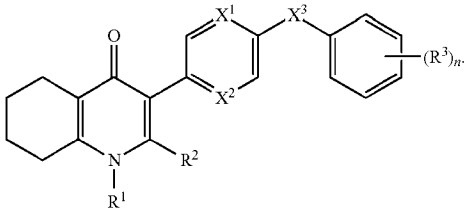


wherein

ring A combines with the carbon atoms with which it is attached to form a C_{3-7}cycloalkenyl.

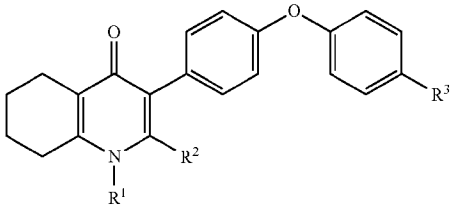
In some embodiments, the compounds are of Formula (IIa):

(IIa)



In some embodiments, the compounds are of Formula (IIa-1):

(IIa-1)

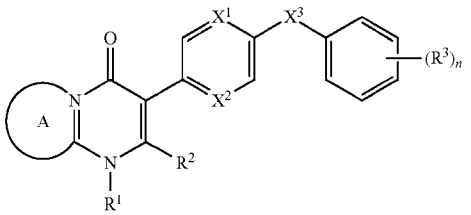


wherein

R^3 is hydrogen, halogen, C_{1-3}alkyl or C_{1-3}haloalkyl.

In some embodiments, the compounds are of Formula (III):

(III)

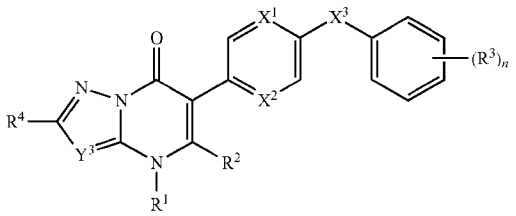


wherein

ring A combines with the nitrogen atom and carbon atom with which it is attached to form a heteroaryl ring.

In some embodiments, the compounds are of Formula (IIIa):

(IIIa)



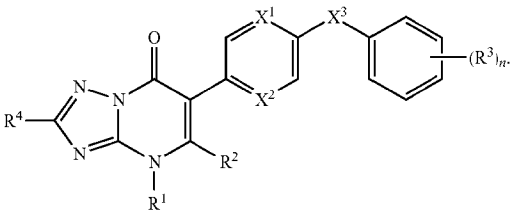
wherein

Y^3 is $C(R^5)$ or N; and

R^4 and R^5 are independently hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR, cyano or phenyl.

In some embodiments, the compounds are of Formula (IIIb):

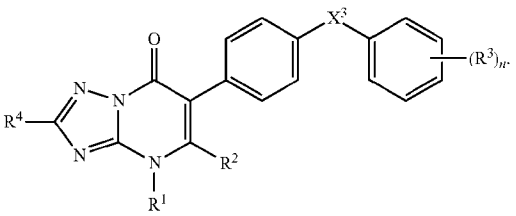
(IIIb)



In some embodiments, the compounds are of R^4 is hydrogen or C_{1-3}alkyl.

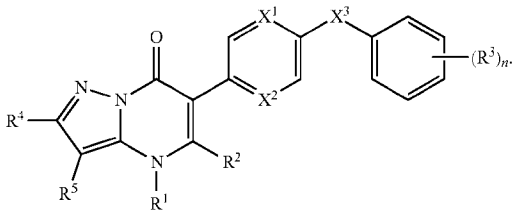
In some embodiments, the compounds are of Formula (IIIb-1):

(IIIb-1)



In some embodiments, the compounds are of Formula (IIIc):

(IIIc)



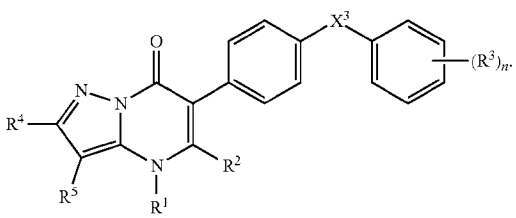
In some embodiments,

R^4 is hydrogen or C_{1-3}alkyl or phenyl; and

R^5 is hydrogen or cyano.

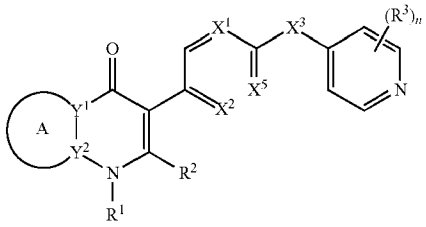
In some embodiments, the compounds are of Formula (IIIc-1):

(IIIc-1)



In some embodiments, the compounds are of Formula (IV):

(IV)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof,

wherein

ring A combines with Y^1 and Y_2 to form a C_{3-7}cycloalkenyl or heteroaryl ring, wherein the C_{3-7}cycloalkenyl or heteroaryl is optionally substituted by halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR, cyano or phenyl;

Y^1 is C or N;

Y^2 is C or N;

X^1 is $C(R^{x1})$ or N,

wherein R^{x1} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^2 is $C(R^{x2})$ or N,

wherein R^{x2} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^3 is O, N(R), S or C_{1-3}alkyl;

X^5 is C or N;

R^1 is hydrogen or C_{1-3}alkyl;

R2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR or —C(O)OR;

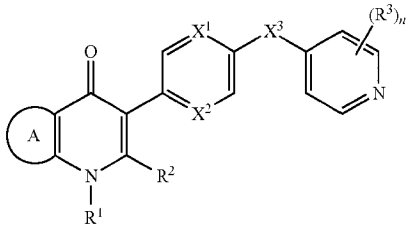
n is 0, 1, 2, 3 or 4;

each R^3 is independently halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5; and

each R is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the compounds are of Formula (IVa):

(IVa)

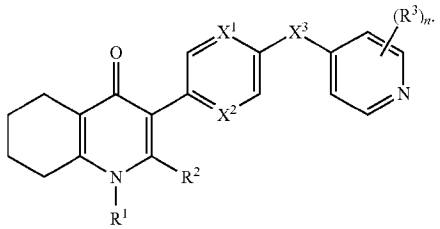


wherein

ring A combines with the carbon atoms with which it is attached to form a C_{3-7}cycloalkenyl.

In some embodiments, the compounds are of Formula (IVb):

(IVb)



In some embodiments, Y^1 is C. In other embodiments, Y^1 is N.

In some embodiments, Y^2 is C. In other embodiments, Y^2 is N.

In some embodiments, Y^1 is C. In other embodiments, Y^1 is N.

In some embodiments, X^1 is $C(R^{x1})$ wherein R^{x1} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl. In other embodiments, X^1 is N.

In some embodiments, X^1 is $C(R^{x1})$ wherein R^{x1} is selected from any of groups (1a)-(1x):

(1a) hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;
(1b) halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;
(1c) hydrogen;
(1d) halogen or C_{1-3}haloalkyl;
(1e) halogen or C_{1-3}alkyl;
(1f) C_{1-3}alkyl;
(1g) hydrogen, methyl or ethyl;
(1h) methyl or ethyl;
(1i) methyl;
(1j) ethyl;
(1k) propyl;
(1l) hydrogen, methyl or propyl;
(1m) methyl or propyl;
(1n) hydrogen, ethyl or propyl;
(1o) ethyl or propyl;
(1p) C_{1-3}haloalkyl;
(1q) C_{1-3}fluoroalkyl;
(1r) fluoromethyl
(1s) difluoromethyl
(1t) trifluoromethyl
(1u) fluoromethyl
(1v) fluoropropyl
(1w) —CH_2OH;
(1x) —CH_2OR;

In some embodiments, X^2 is $C(R^{x2})$ wherein R^{x2} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl. In other embodiments, X^2 is N.

In some embodiments, X^2 is $C(R^{x2})$ wherein R^{x2} is selected from any of groups (2a)-(2x):

(2a) hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;
(2b) halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;
(2c) hydrogen;
(2d) halogen or C_{1-3}haloalkyl;
(2e) halogen or C_{1-3}alkyl;
(2f) C_{1-3}alkyl;
(2g) hydrogen, methyl or ethyl;
(2h) methyl or ethyl;
(2i) methyl;
(2j) ethyl;
(2k) propyl;
(21) hydrogen, methyl or propyl;
(2m) methyl or propyl;
(2n) hydrogen, ethyl or propyl;
(2o) ethyl or propyl;
(2p) C_{1-3}haloalkyl;
(2q) C_{1-3}fluoroalkyl;
(2r) fluoromethyl
(2s) difluoromethyl
(2t) trifluoromethyl
(2u) fluoromethyl
(2v) fluoropropyl
(2w) —CH_2OH;
(2x) —CH_2OR;

In some embodiments, X^3 is selected from any of groups (3a)-(3p):

(3a) O, N(R), S or C_{1-3}alkyl;
(3b) O, N(R) or S;
(3c) O or N(R);
(3d) O;
(3e) N(R);
(3f) S or C_{1-3}alkyl;
(3g) O, N(R), or C_{1-3}alkyl;
(3h) O or C_{1-3}alkyl;
(3i) N(R), S or C_{1-3}alkyl;
(3j) N(R) or C_{1-3}alkyl;
(3k) O or C_{1-3}alkyl;
(31) C_{1-3}alkyl;
(3m) methylene
(3n) ethylene;
(3o) propylene;
(3p) NH.

In some embodiments, X^4 is C. In other embodiments, X^4 is N.

In some embodiments, X^5 is C. In other embodiments, X^5 is N.

In some embodiments, R^1 is selected from any of groups (4a)-(41):

(4a) hydrogen or C_{1-3}alkyl;
(4b) hydrogen;
(4c) C_{1-3}alkyl;
(4d) hydrogen, methyl or ethyl;

(4e) methyl or ethyl;
(4f) methyl;
(4g) ethyl;
(4h) propyl;
(4i) hydrogen, methyl or propyl;
(4j) methyl or propyl;
(4k) hydrogen, ethyl or propyl;
(4l) ethyl or propyl;

In some embodiments, R2 is selected from any of groups (5a)-(5gg):

(5a) hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR or —C(O)OR;
(5b) C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR or —C(O)OR;
(5c) hydrogen, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR or —C(O)OR;
(5d) hydrogen, C_{1-3}alkyl, —CH_2OH, —CH_2OR or —C(O)OR;
(5e) hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OR or —C(O)OR;
(5f) hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, or —C(O)OR;
(5g) hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, or —CH_2OR;
(5h) hydrogen or C_{1-3}alkyl;
(5i) hydrogen;
(5j) C_{1-3}alkyl;
(5k) hydrogen, methyl or ethyl;
(5l) methyl or ethyl;
(5m) methyl;
(5n) ethyl;
(5o) propyl;
(5p) hydrogen, methyl or propyl;
(5q) methyl or propyl;
(5r) hydrogen, ethyl or propyl;
(5s) ethyl or propyl;
(5t) C_{1-3}haloalkyl, —CH_2OH, —CH_2OR or —C(O)OR;
(5u) C_{1-3}haloalkyl;
(5v) C_{1-3}fluoroalkyl;
(5w) fluoromethyl
(5x) difluoromethyl
(5y) trifluoromethyl
(5z) fluoromethyl
(5aa) fluoropropyl
(5bb) —CH_2OH;
(5cc) —CH_2OR;
(5dd) —C(O)OR;
(5ee) —C(O)OH;
(5ff) —C(O)OMe;
(5gg) C(O)OEt

In some embodiments, n is selected from any of groups (6a)-(6k):

(6a) n is 1, 2, 3, or 4.
(6b) n is 0, 1, 2, or 3.
(6c) n is 0, 1, or 2.
(6d) n is 0 or 1.
(6e) n is 1 or 2.
(60 n is 2 or 3.
(6g) n is 1.
(6h) n is 2.
(6i) n is 3.
(6j) n is 4.
(6k) n is 0.

In some embodiments, R^3 is selected from any of groups (7a)-(7cc):

(7a) each R^3 is independently halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR, SF_5, or two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane;
(7b) each R^3 is independently C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5;
(7c) each R^3 is independently halogen, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5;
(7d) each R^3 is independently halogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5;
(7e) each R^3 is independently halogen, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5;
(7f) each R^3 is independently halogen, C_{1-3}alkyl, C_{1-3}alkoxy, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5;
(7g) each R^3 is independently C_{1-3}alkyl, C_{1-3}alkoxy, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5;
(7h) each R^3 is independently —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5;
(7i) each R^3 is independently halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5;
(7j) each R^3 is independently halogen, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —C(O)OR, or two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane;
(7k) each R^3 is independently halogen, C_1haloalkyl, —O-C_1haloalkyl, —C(O)OR, or two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane;
(7l) each R^3 is independently fluoro, chloro, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —C(O)OR, or two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane;
(7m) each R^3 is independently fluoro, chloro, trifluoromethyl, —O—C_{1-3}haloalkyl, —C(O)OR, or two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane;
(7n) each R^3 is independently fluoro, chloro, trifluoromethyl, —OCF_3, —C(O)OR, or two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane;
(7o) each R^3 is independently fluoro, chloro, trifluoromethyl, —OCF_3, —C(O)OH, or two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane;
(7p) each R^3 is independently trifluoromethyl, —OCF_3, —C(O)OH, or two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane;
(7q) each R^3 is independently fluoro, chloro, —OCF_3, —C(O)OH, or two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane;
(7r) each R^3 is independently fluoro, chloro, trifluoromethyl, —OCF_3, or two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane;
(7s) each R^3 is independently fluoro, chloro, trifluoromethyl or —OCF_3;
(7t) each R^3 is independently fluoro, chloro, trifluoromethyl, —OCF_3 or —C(O)OH;
(7u) each R^3 is independently fluoro, chloro, trifluoromethyl or —OCF_3;

(7v) each R^3 is independently fluoro, chloro or trifluoromethyl;
(7w) each R^3 is independently fluoro or chloro;
(7x) each R^3 is independently fluoro;
(7y) each R^3 is independently chloro;
(7z) each R^3 is trifluoromethyl;
(7aa) each R^3 is —OCF_3;
(7bb) each R^3 is —C(O)OH;
(7cc) two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane;

In some embodiments, R is selected from any of groups (8a)-(8l):

(8a) hydrogen or C_{1-3}alkyl;
(8b) hydrogen;
(8c) C_{1-3}alkyl;
(8d) hydrogen, methyl or ethyl;
(8e) methyl or ethyl;
(8f) methyl;
(8g) ethyl;
(8h) propyl;
(8i) hydrogen, methyl or propyl;
(8j) methyl or propyl;
(8k) hydrogen, ethyl or propyl;
(8l) ethyl or propyl;

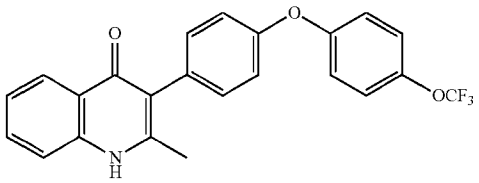
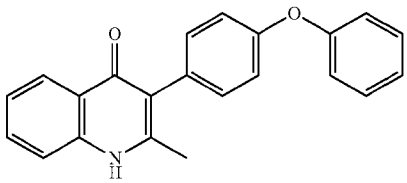
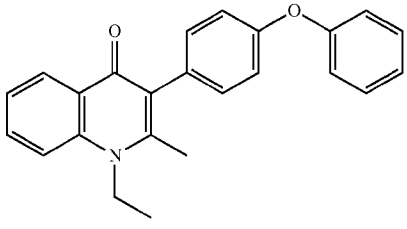
In some embodiments, the compound is of Formula (I), (Ia), (Ib) or (Ic), and X^1, X^2, X^3, R^1, R^2, R^3 and n are selected from any combination of groups (1a)-(8l).

In some embodiments, the compound is of Formula (II), (IIa) or (IIa-1), and X^1, X^2, X^3, R^1, R^2, R^3 and n are selected from any combination of groups (1a)-(8l).

In some embodiments, the compound is of Formula (III), (IIIa), (IIIb), (IIIb-1), (IIIc) or (IIIc-1), and X^1, X^2, X^3, R^1, R^2, R^3 and n are selected from any combination of groups (1a)-(8l).

In some embodiments, the compound is of Formula (IV), (IVa) or (IVb) and X^1, X^2, X^3, R^1, R^2, R^3 and n are selected from any combination of groups (1a)-(8l).

In some embodiments, the compound is:

No.	ID	Structure	Name
		ELQ-type systems	
PA1	MJM102/ MJM113 (ELQ-271)		2-methyl-3-(4-(4-(trifluoromethoxy)phenoxy)phenyl) quinolin-4(1H)-one
PA2	MJM 129		2-methyl-3-(4-phenoxyphenyl)quinolin-4(1H)-one
PA3	JM10		1-ethyl-2-methyl-3-(4-phenoxyphenyl)quinolin-4(1H)-one

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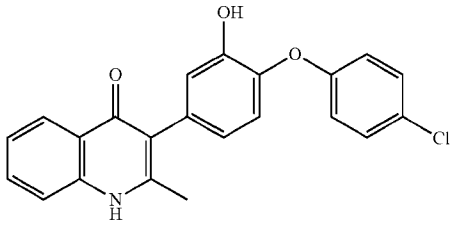
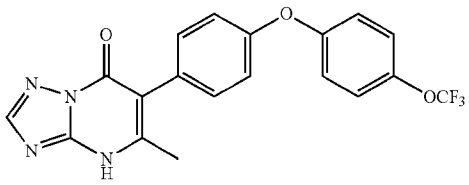
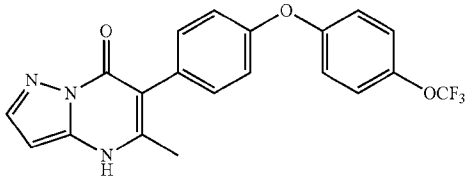
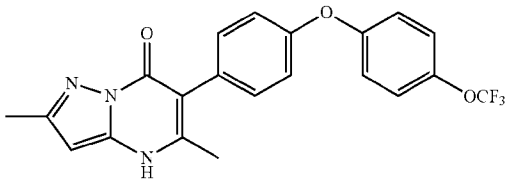
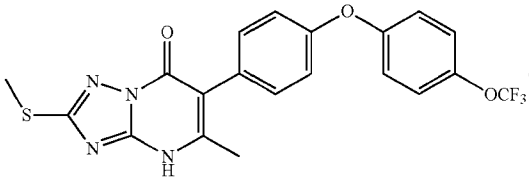
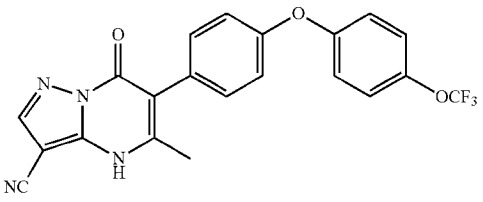
No.	ID	Structure	Name
PA4	RG38		3-(4-(4-chlorophenoxy)-3-hydroxyphenyl)-2-methylquinolin-4(1H)-one
		5,6-fused pyridone systems	
1	MJM136		5-methyl-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-[1,2,4]triazolo[1,5-a]pyrimidin-7(4H)-one
2	MJM141		5-methyl-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl) pyrazolo[1,5-a]pyrimidin-7(4H)-one
3	JAG006		2,5-dimethyl-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl) pyrazolo[1,5-a]pyrimidin-7(4H)-one
4	JAG013		5-methyl-2-(methylthio)-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-[1,2,4]triazolo[1,5-a]pyrimidin-7(4H)-one
5	JAG014		5-methyl-7-oxo-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-4,7-dihydropyrazolo[1,5-a]pyrimidine-3-carbonitrile

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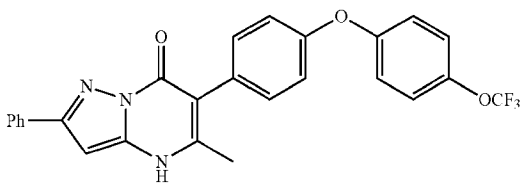
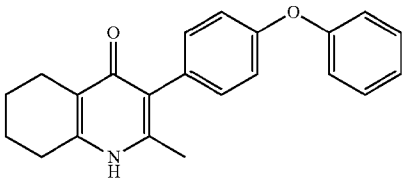
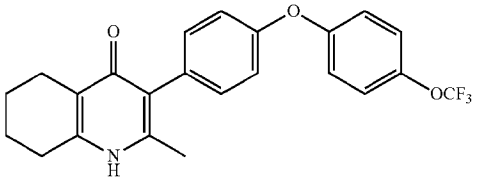
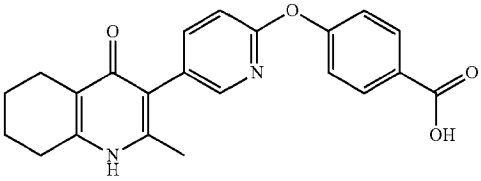
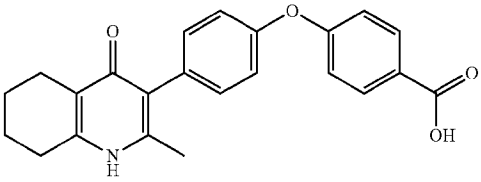
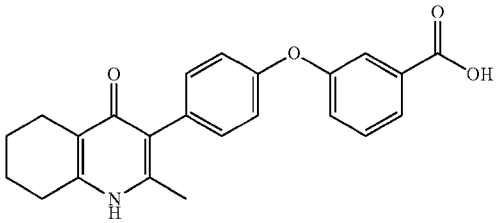
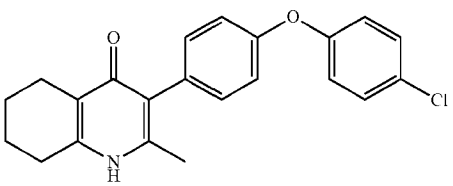
No.	ID	Structure	Name
6	JAG015		5-methyl-2-phenyl-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl) pyrazolo[1,5-a]pyrimidin-7(4H)-one
		Tetrahydroquinolones (THQ)	
7	MJM170		2-methyl-3-(4-phenoxyphenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one
8	JAG21		2-methyl-3-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one
9	JAG039		4-((5-(2-methyl-4-oxo-1,4,5,6,7,8-hexahydroquinolin-3-yl)pyridin-2-yl)oxy)benzoic acid
10	JAG046		4-(4-(2-methyl-4-oxo-1,4,5,6,7,8-hexahydroquinolin-3-yl)phenoxy)benzoic acid
11	JAG047		3-(4-(2-methyl-4-oxo-1,4,5,6,7,8-hexahydroquinolin-3-yl)phenoxy)benzoic acid
12	JAG50		3-(4-(4-chlorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one

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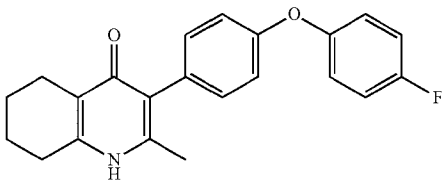
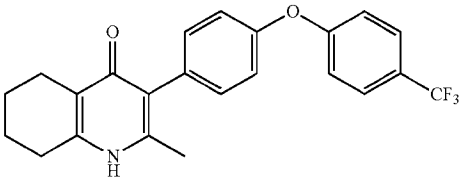
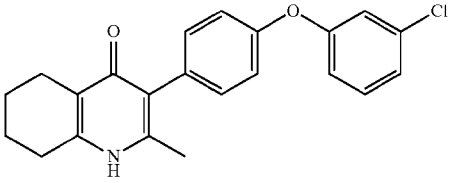
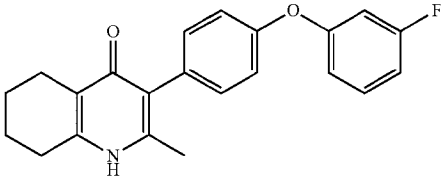
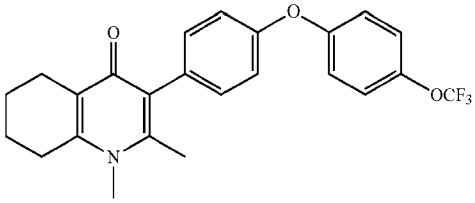
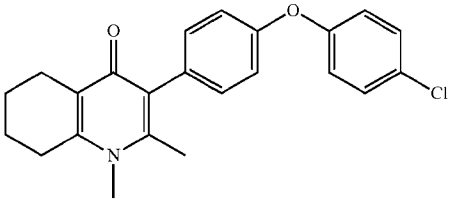
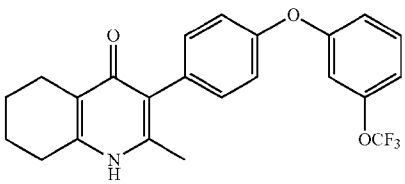
No.	ID	Structure	Name
13	JAG58		3-(4-(4-fluorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one
14	JAG63		2-methyl-3-(4-(4-(trifluoromethyl)phenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one
15	JAG062		3-(4-(3-chlorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one
16	JAG067		3-(4-(3-fluorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one
17	JAG023		1,2-dimethyl-3-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one
18	JAG077		3-(4-(4-chlorophenoxy)phenyl)-1,2-dimethyl-5,6,7,8-tetrahydroquinolin-4(1H)-one
19	AS006		2-methyl-3-(4-(3-(trifluoromethoxy)phenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one

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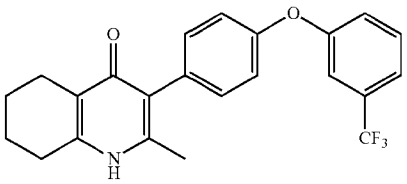
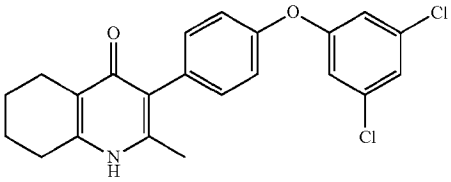
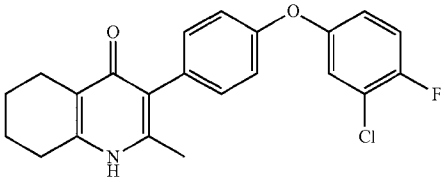
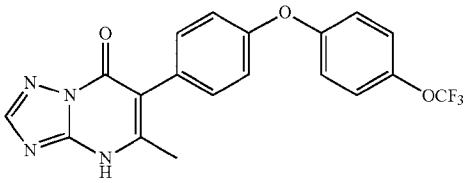
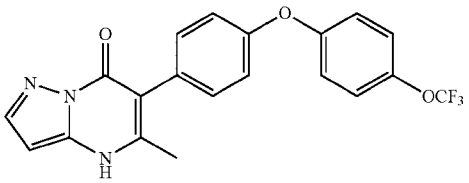
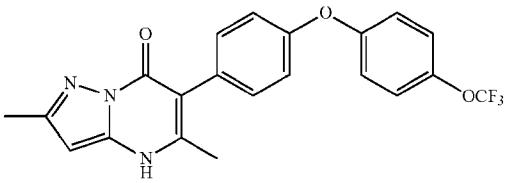
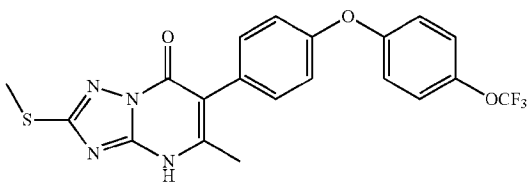
No.	ID	Structure	Name
20	AS0012		2-methyl-3-(4-(3-(trifluoromethyl)phenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one
21	AS021		3-(4-(3,5-dichlorophenoxy)phenyl)-2-methyl-5,6.7,8-tetrahydroquinolin-4(1H)-one
22	AS022		3-(4-(3-chloro-4-fluorophenoxy)phenyl)-2-methyl-5.6,7.8-tetrahydroquinolin-4(1H)-one

or a stereoisomer thereof or a pharmaceutically acceptable salt thereof.

In some embodiments, the compound is:

ID	Structure	Name
MJM136		5-methyl-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-[1,2,4]triazolo[1,5-a]pyrimidin-7(4H)-one
MJM141		5-methyl-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl)pyrazolo[1,5-a]pyrimidin-7(4H)-one
JAG006		2,5-dimethyl-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl)pyrazolo[1,5-a]pyrimidin-7(4H)-one
JAG013		5-methyl-2-(methylthio)-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-[1,2,4]triazolo[1,5-a]pyrimidin-7(4H)-one

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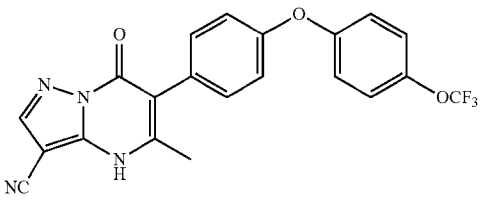
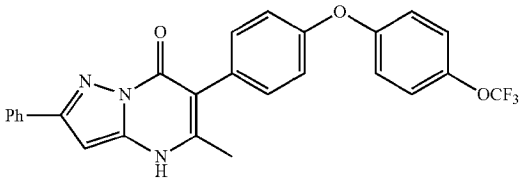
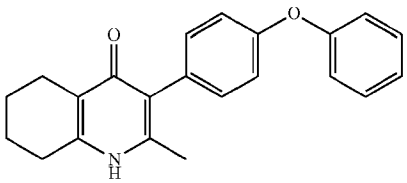
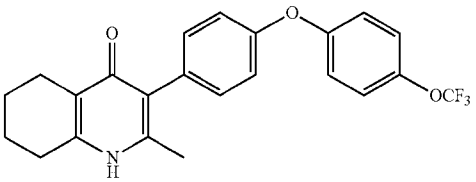
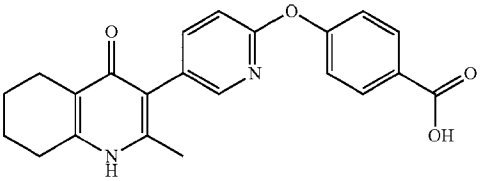
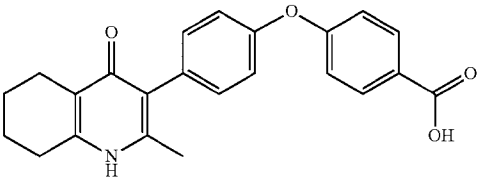
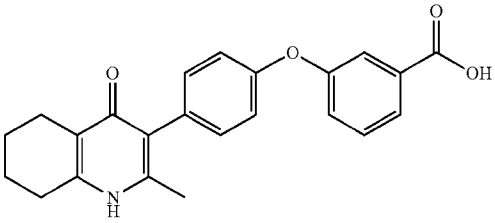
ID	Structure	Name
JAG014		5-methyl-7-oxo-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-4,7-dihydropyrazolo[1,5-a]pyrimidine-3-carbonitrile.
JAG015		5-methyl-2-phenyl-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl)pyrazolo[1,5-a]pyrimidin-7(4H)-one.
MJM170		2-methyl-3-(4-phenoxyphenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one
JAG21		2-methyl-3-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one
JAG039		methyl 3-((5-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)pyridin-2-yl)oxy)benzoate
JAG046		4-(4-(2-methyl-4-oxo-1,4,5,6,7,8-hexahydroquinolin-3-yl)phenoxy)benzoic acid
JAG047		3-(4-(2-methyl-4-oxo-1,4,5.6,7,8-hexahydroquinolin-3-yl)phenoxy)benzoic acid

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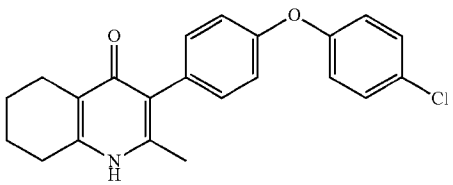
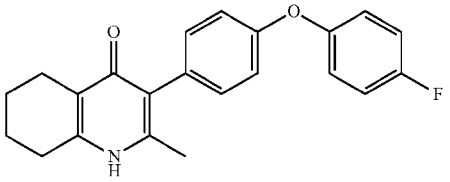
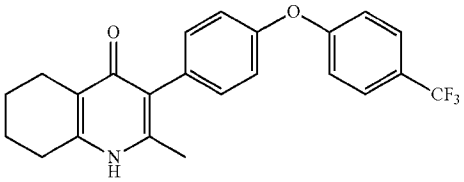
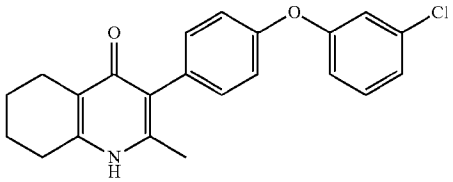
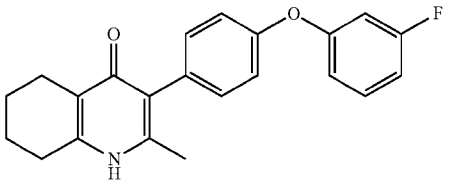
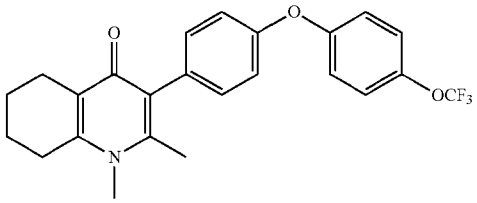
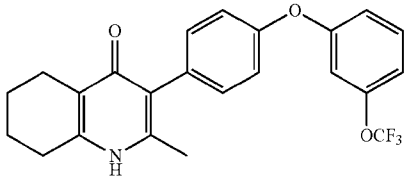
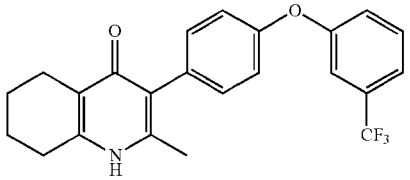
ID	Structure	Name
JAG50		3-(4-(4-chlorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one
JAG58		3-(4-(4-Fluorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one
JAG63		2-Methyl-3-(4-(4-(trifluoromethyl)phenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one
JAG062		3-(4-(3-Chlorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one
JAG069		3-(4-(3-Fluorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one
JAG023		1,2-dimethyl-3-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4-one
AS006/ JAG143		3-(4-(3,4-Dichlorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one
AS012/ JAG144		3-(4-(3,4-Dichlorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one

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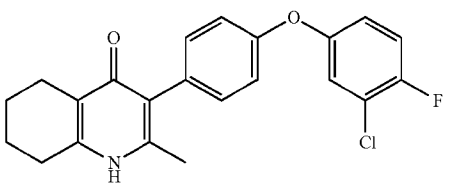
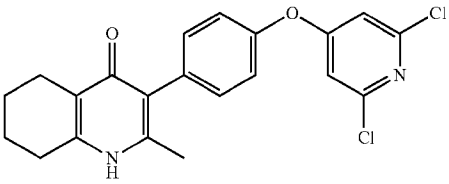
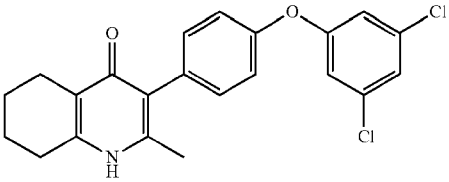
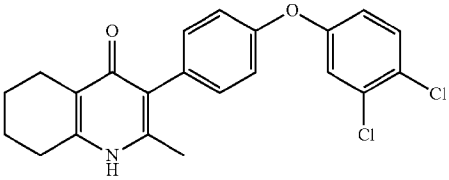
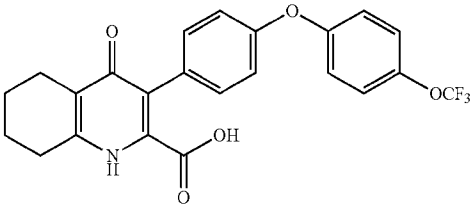
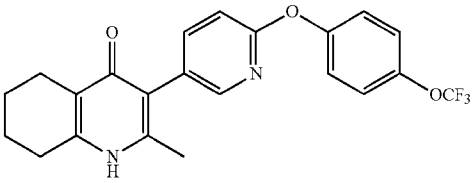
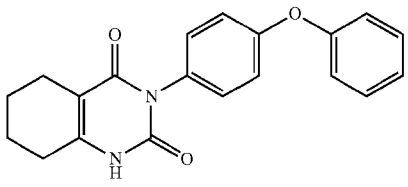
ID	Structure	Name
AS021/ JAG145		3-[4-(3-chloro-4-fluorophenoxy)phenyl]-2-methyl-5,6,7,8-tetrahydro-1H-quinolin-4-one
AS034/ JAG148		3-{4-[(2,6-dichloropyridin-4-yl)oxy]phenyl}-2-methyl-5,6,7,8-tetrahydro-1H-quinolin-4-one
AS022		3-[4-(3,5-dichlorophenoxy)phenyl]-2-methyl-5,6,7,8-tetrahydro-1H-quinolin-4(1H)-one
JAG084		3-(4-(3.4-Dichlorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one
JAG091		3(4-(4-Trifluoromethoxyphenoxy)phenyl)-2-(carboxylate)-5,6,7,8-tetrahydroquinolin-4(1H)-one
JAG092		3-(6-(4-Trifluoromethoxyphenoxy)pyrdin-3-yl)-2-methyl-5,6,7,8-tetrahydroquinolin-(4)-one
JAG095		3-(4-Phenonxyphenyl)-1.2,3,4.5,6,7,8-octahydroquinazoline-2,4,dione

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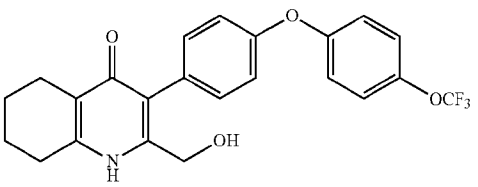
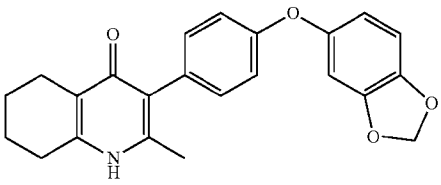
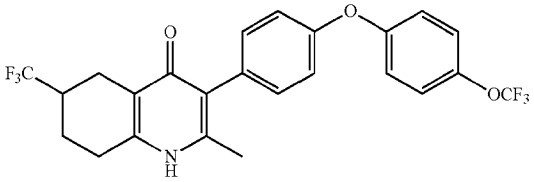
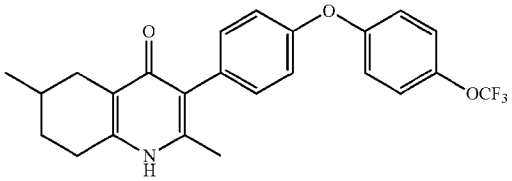
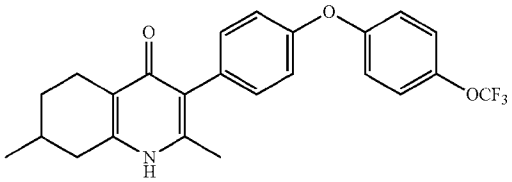
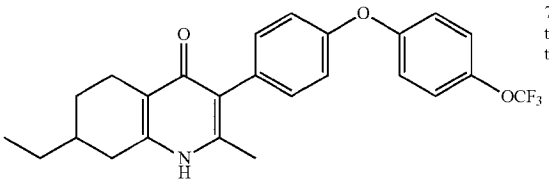
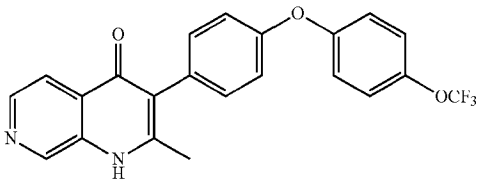
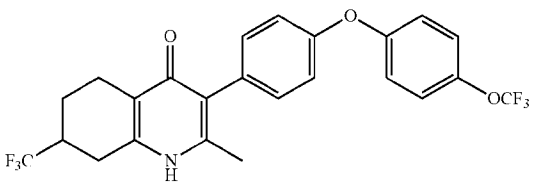
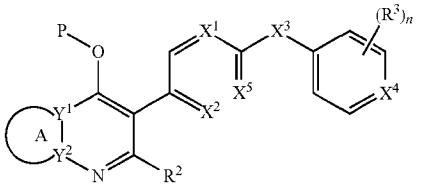
ID	Structure	Name
JAG099		3(4-(4-Trifluoromethoxyphenoxy)phenyl)-2-(methylhydroxy)-5,6,7,8-tetrahydroquinolin-4(1H)-one
AS032		3-[4-(2H-1,3-benzodioxol-5-yloxy)phenyl]-2-methyl-5,6,7,8-tetrahydro-1H-quinolin-4-one
JAG100		6-Ethyl-3-(4-(4-trifluoromethoxyphenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one
JAG106		3(4-(4-Trifluoromethoxyphenoxy)phenyl)-2,6-dimethyl-5,6,7,8-tetrahydroquinolin-4(1H)-one
JAG107		3(4-(4-Trifluoromethoxyphenoxy)phenyl)-2,7-dimethyl-5,6,7,8-tetrahydroquinolin-4(1H)-one
JAG121		7-Ethyl-2-methyl-3(4-(4-trifluoromethoxyphenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one
JA129		3(4-(4-trifluoromethoxyphenoxy)phenyl)-2-methyl-1,7-napthyrid-4(1H)-one
JAG162		7-Trifluromethyl-2-methyl-3(4-(4-trifluoromethoxyphenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one

or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof.

In another aspect, the invention provides prodrugs of a compound of Formula (I). The term "prodrug" is intended to represent covalently bonded carriers, which are capable of releasing the active ingredient when the prodrug is administered to a mammalian subject. Release of the active ingredient occurs in vivo. Prodrugs can be prepared by techniques known to one skilled in the art. These techniques generally modify appropriate functional groups in a given compound. These modified functional groups however regenerate original functional groups by routine manipulation or in vivo. Prodrugs of compounds of the invention include compounds wherein an amino, hydroxy, carboxylic or a similar group is modified. Examples of prodrugs include, but are not limited to esters (e.g., acetate, formate, and benzoate), carbamates (e.g., N,N-dimethylaminocarbonyl), amides (e.g., trifluoroacetylamino, acetylamino, and the like), and the like. A complete discussion of prodrugs is found in Huttunen, K. M. and Rautio J. *Current Topics in Medicinal Chemistry,* 2011, 11, 2265-2287 and Stella, V. J. et al. (2007). *Prodrugs: Challenges and Awards Part* 1. New York: Springer. The disclosure of both references is herein incorporated by reference in its entirety.

In some embodiments, the prodrug of a compound of Formula (I) has the structure of Formula (I-p):



(I-p)

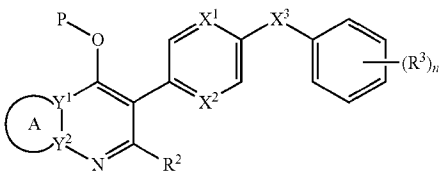
or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

ring A, Y^1, Y^2, X^1, X^2, X^3, X^4, X^5, R, R^3 and n are as described above;

R2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR, —C(O)OR or —CH_2OP; and

P is —C(O)OR', —C(O)R', —$C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (Ia-p):



(Ia-p)

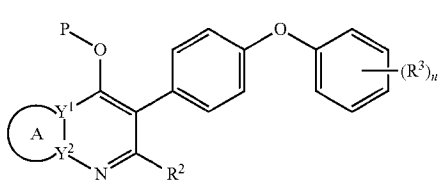
or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

ring A, Y^1, Y^2, X^1, X^2, X^3, R, R^2, R^3 and n are as described above;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR, —C(O)OR or —CH_2OP; and

P is —C(O)OR', —C(O)R', —$C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (Ib-p):



(Ib-p)

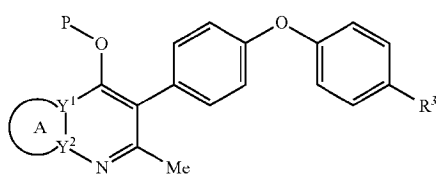
or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

ring A, Y^1, Y^2, R, R^2, R^3 and n are as described above;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR, —C(O)OR or —CH_2OP; and

P is —C(O)OR', —C(O)R', —$C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (Ic-p):



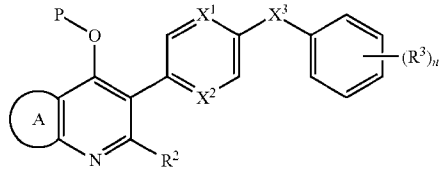
(Ic-p)

or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

ring A, Y^1, Y^2, R and R^3 are as described above; and

P is —C(O)OR', —C(O)R', —$C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (II-p):



(II-p)

or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

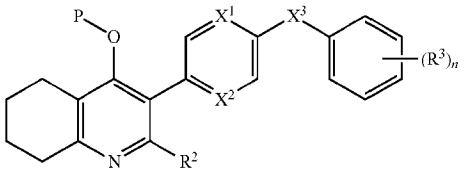
ring A, X^1, X^2, X^3, R, R^2, R^3 and n are as described above;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR, —C(O)OR or —CH_2OP; and

P is —C(O)OR', —C(O)R', —$C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (IIa-p):

(IIa-p)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

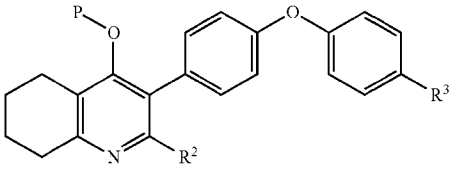
X^1, X^2, X^3, R, R^2, R^3 and n are as described above;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR, —C(O)OR or —CH_2OP; and

P is —C(O)OR', —C(O)R', —$C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (IIa-1-p):

(IIa-1-p)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

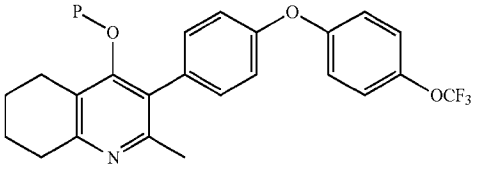
R, R^2, R^3 and n are as described above;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR, —C(O)OR or —CH_2OP; and

P is —C(O)OR', —C(O)R', —$C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (IIa-2-p):

(IIa-2-p)

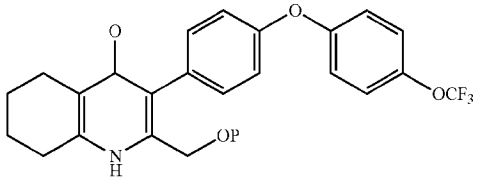


or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

P is —C(O)OR', —C(O)R', —$C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (IIa-3-p):

(IIa-3-p)

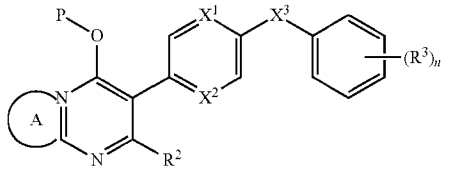


or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

P is —C(O)OR', —C(O)R', —$C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (III-p):

(III-p)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

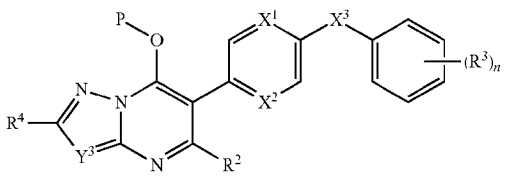
ring A, X^1, X^2, X^3, R, R^2, R^3 and n are as described above;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR, —C(O)OR or —CH_2OP; and

P is —C(O)OR', —C(O)R', —$C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (IIIa-p):

(IIIa-p)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

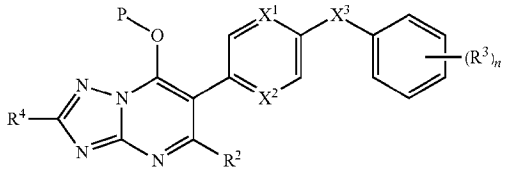
Y^3, X, X^2, X^3, R, R^2, R^3, R^4 and n are as described above;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR, —C(O)OR or —CH_2OP; and

P is —C(O)OR', —C(O)R', —$C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (IIIb-p):

(IIIb-p)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

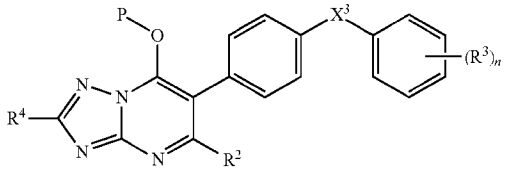
X^1, X^2, X^3, R, R^2, R^3, R^4 and n are as described above;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, CH_2OH, $—CH_2OR$, —C(O)OR or $—CH_2OP$; and

P is —C(O)OR', —C(O)R', $—C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (IIIb-1-p):

(IIIb-1-p)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

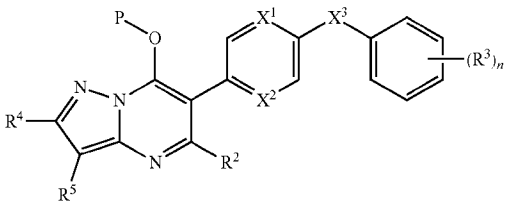
X^3, R, R^2, R^3, R^4 and n are as described above;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, $—CH_2OH$, $—CH_2OR$, —C(O)OR or $—CH_2OP$; and

P is —C(O)OR', —C(O)R', $—C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (IIIc-p):

(IIIc-p)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

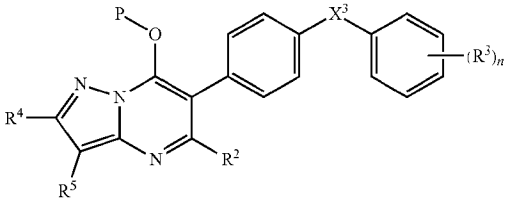
X^1, X^2, X^3, R, R^2, R^3, R^4, R^5 and n are as described above;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, $—CH_2OH$, $—CH_2OR$, —C(O)OR or $—CH_2OP$; and

P is —C(O)OR', —C(O)R', $—C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (IIIc-1-p):

(IIIc-1-p)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

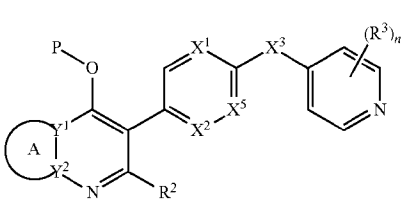
X^3, R, R^2, R^3, R^4, R^5 and n are as described above;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, $—CH_2OH$, $—CH_2OR$, —C(O)OR or $—CH_2OP$; and

P is —C(O)OR', —C(O)R', $—C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (IV-p):

(IV-p)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

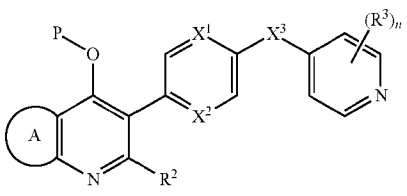
ring A, Y^1, Y^2, X^1, X^2, X^3, X^5, R, R^2, R^3 and n are as described above;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, $—CH_2OH$, $—CH_2OR$, —C(O)OR or $—CH_2OP$; and

P is —C(O)OR', —C(O)R', $—C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (IVa-p):

(IVa-p)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

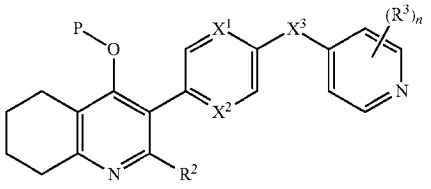
ring A, X^1, X^2, R, R^2, R^3 and n are as described above;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, $—CH_2OH$, $—CH_2OR$, —C(O)OR or $—CH_2OP$; and

P is —C(O)OR', —C(O)R', $—C(O)NR'_2$ or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the prodrug is a compound of Formula (IVb-p

(IVb-p)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof, wherein

X^1, X^2, X^3, R, R^2, R^3 and n are as described above;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR, —C(O)OR or —CH_2OP; and

P is —C(O)OR', —C(O)R', —C(O)NR'_2 or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl.

In some embodiments, the compound is of any of Formulae (I-p), (Ia-p), (Ib-p), (Ic-p), (II-p), (Ia-p), (IIa-1-p), (IIa-2-p), (IIa-3-p), (III-p), (IIIa-p), (IIIb-p), (IIIb-1-p), (IIIc-p), (IIIc-1-p), (IV-p), (Iva-p) or (IVb-p), and X^1, X^2, X^3, R^1, R^2, R^3 and n are selected from any combination of groups (1a)-(8l).

In some embodiments, the compound is of Formulae (I-p), (Ia-p), (Ib-p), (Ic-p), (II-p), (Ia-p), (IIa-1-p), (IIa-2-p), (IIa-3-p), (III-p), (IIIa-p), (IIIb-p), (IIIb-1-p), (IIIc-p), (IIIc-1-p), (IV-p), (Iva-p) or (IVb-p), X^1, X^2, X^3, R^1, R^2, R^3 and n are selected from any combination of groups (1a)-(8l), and P is —$C(O)_2R'$.

In some embodiments, the compound is of Formulae (I-p), (Ia-p), (Ib-p), (Ic-p), (II-p), (Ia-p), (IIa-1-p), (IIa-2-p), (IIa-3-p), (III-p), (IIIa-p), (IIIb-p), (IIIb-1-p), (IIIc-p), (IIIc-1-p), (IV-p), (Iva-p) or (IVb-p), X^1, X^2, X^3, R^1, R^2, R^3 and n are selected from any combination of groups (1a)-(8l), and P is —C(O)R'.

In some embodiments, the compound is of Formulae (I-p), (Ia-p), (Ib-p), (Ic-p), (II-p), (Ia-p), (IIa-1-p), (IIa-2-p), (IIa-3-p), (III-p), (IIIa-p), (IIIb-p), (IIIb-1-p), (IIIc-p), (IIIc-1-p), (IV-p), (Iva-p) or (IVb-p), X^1, X^2, X^3, R^1, R^2, R^3 and n are selected from any combination of groups (1a)-(8l), and P is —C(O)NR'_2.

In some embodiments, the compound is of Formulae (I-p), (Ia-p), (Ib-p), (Ic-p), (II-p), (Ia-p), (IIa-1-p), (IIa-2-p), (IIa-3-p), (III-p), (IIIa-p), (IIIb-p), (IIIb-1-p), (IIIc-p), (IIIc-1-p), (IV-p), (Iva-p) or (IVb-p), X^1, X^2, X^3, R^1, R^2, R^3 and n are selected from any combination of groups (1a)-(8l), and P is —OP(O)(OR')OR'.

In another aspect, the invention provides a pharmaceutical composition comprising a compound of Formula (I-p) and a pharmaceutically acceptable diluent, excipient, or carrier.

In some embodiments, the pharmaceutical composition is a combination comprising a compound of Formula (I), an 8-Aminoquinoline drug and a pharmaceutically acceptable diluent, excipient, or carrier.

In some embodiments, the pharmaceutical composition is a combination comprising a compound of Formula (I), tafenoquine and a pharmaceutically acceptable diluent, excipient, or carrier.

In some embodiments, the pharmaceutical composition is a combination comprising a compound of Formula (I-p), an 8-Aminoquinoline drug and a pharmaceutically acceptable diluent, excipient, or carrier.

In some embodiments, the pharmaceutical composition is a combination comprising a compound of Formula (I-p), tafenoquine and a pharmaceutically acceptable diluent, excipient, or carrier.

In another aspect, the invention provides a pharmaceutical composition comprising a compound of claim Formula (I) and a pharmaceutically acceptable diluent, excipient, or carrier. In another aspect, the invention provides a method for treating an apicomplexan parasitic infection, comprising administering to a subject (such as a human subject) in need thereof an amount effective to treat the infection of the compound of Formula (I) or a pharmaceutical composition comprising a compound of Formula (I). In some embodiments of the method, the infection comprises a *Toxoplasma gondii* infection and/or a *Plasmodium falciparum* infection. In some embodiments of the method, the infection comprises an infection in the subject's brain and/or the subject's eye. In some embodiments of the method, the compound is a prodrug of Formula (I-p).

In another aspect, the invention provides a method for treating an apicomplexan parasitic infection, comprising administering to a subject (such as a human subject) in need thereof an amount effective to treat the infection of a combination comprising a compound of Formula (I) and a 8-Aminoquinoline drug or a pharmaceutical composition comprising a compound of Formula (I) and a 8-Aminoquinoline drug. In some embodiments of the method, the infection comprises a *Toxoplasma gondii* infection and/or a *Plasmodium falciparum* infection. In some embodiments of the method, the infection comprises an infection in the subject's brain and/or the subject's eye. In some embodiments of the method, the compound is a prodrug of Formula (I-p). In some embodiments of the method, the 8-Aminoquinoline drug is tafenoquine.

In some embodiments of the method, the subject is immune compromised. In some embodiments of the method, the subject is immune compromised due to cancer/cancer treatment, autoimmune disease, and/or AIDS. In some embodiments of the method, the subject has malaria, and the treating comprises reducing severity of one or more symptoms of malaria, and/or reducing recurrence of symptoms of malaria. In some embodiments of the method, the subject has toxoplasmosis, and the treating comprises reducing severity of one or more symptoms of toxoplasmosis, and/or reducing recurrence of symptoms of toxoplasmosis. In some embodiments of the method, the treating comprises reducing parasitic load in the subject. In some embodiments of the method, the treating comprises reducing the bradyzoite form and/or the tachyzoite form of the parasite in the subject. In some embodiments of the method, the method further comprises administering to the subject one or more additional compounds in an amount effective to treat the infection. In some embodiments of the method, the one or more additional compounds are selected from the group consisting of pyrimethamine, sulfadiazine, cycloguanil, inhibitors of kcalcium kinases or dense granules or vacuolar atpases, atovoquone, and bulky cytochrome Qi inhibitors, itraconazole and other inhibitors of *T. gondii*.

In another aspect, the invention provides a method for monitoring treatment of an apicomplexan parasitic infection (including but not limited to any of the treatments of claims **23-33**), comprising monitoring expression, protein in serum or plasma, and/or activity of 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, or all of the markers listed in Table and figures in Example 8/FIGS. **3A-3B** in the Appendix in a subject (such as a human subject) being treated for an apicomplexan parasitic infection, wherein a decrease or increase in expression

and/or presence and/or activity of the one or more markers indicates that the treatment is effective.

In some embodiments of the method, the infection is a *T. gondii* infection. In some embodiments of the method, the infection is in the subject's brain or other neurologic tissue.

Definitions

Terms used herein may be preceded and/or followed by a single dash, "-", or a double dash, "=", to indicate the bond order of the bond between the named substituent and its parent moiety; a single dash indicates a single bond and a double dash indicates a double bond or a pair of single bonds in the case of a spiro-substituent. In the absence of a single or double dash it is understood that a single bond is formed between the substituent and its parent moiety; further, substituents are intended to be read "left to right" unless a dash indicates otherwise. For example, alkyl, alkyl-, and -alkyl indicate the same functionality.

Further, certain terms herein may be used as both monovalent and divalent linking radicals as would be familiar to those skilled in the art, and by their presentation linking between two other moieties. For example, an alkyl group can be both a monovalent radical or divalent radical; in the latter case, it would be apparent to one skilled in the art that an additional hydrogen atom is removed from a monovalent alkyl radical to provide a suitable divalent moiety.

The term "alkoxy" as used herein, means an alkyl group, as defined herein, appended to the parent molecular moiety through an oxygen atom. Representative examples of alkoxy include, but are not limited to, methoxy, ethoxy, propoxy, 2-propoxy, butoxy, tert-butoxy, pentyloxy, and hexyloxy.

The term "alkyl" as used herein, means a straight or branched chain hydrocarbon containing from 1 to 10 carbon atoms, unless otherwise specified. Representative examples of alkyl include, but are not limited to, methyl, ethyl, n-propyl, iso-propyl, n-butyl, sec-butyl, iso-butyl, tert-butyl, n-pentyl, isopentyl, neopentyl, n-hexyl, 3-methylhexyl, 2,2-dimethylpentyl, 2,3-dimethylpentyl, n-heptyl, n-octyl, n-nonyl, and n-decyl. When an "alkyl" group is a linking group between two other moieties, then it may also be a straight or branched chain; examples include, but are not limited to —CH_2—, —CH_2CH_2—, —$CH_2CH_2CHC(CH_3)$—, —$CH_2CH(CH_2CH_3)CH_2$—.

The terms "cyano" and "nitrile" as used herein, mean a —CN group. "Cycloalkenyl" as used herein refers to a monocyclic or a bicyclic cycloalkenyl ring system. Monocyclic ring systems are cyclic hydrocarbon groups containing from 3 to 8 carbon atoms, where such groups are unsaturated (i.e., containing at least one annular carbon-carbon double bond), but not aromatic. Examples of monocyclic ring systems include cyclopentenyl and cyclohexenyl. Bicyclic cycloalkenyl rings are bridged monocyclic rings or a fused bicyclic rings. Bridged monocyclic rings contain a monocyclic cycloalkenyl ring where two non-adjacent carbon atoms of the monocyclic ring are linked by an alkylene bridge of between one and three additional carbon atoms (i.e., a bridging group of the form —$(CH_2)_w$—, where w is 1, 2, or 3). Representative examples of bicyclic cycloalkenyls include, but are not limited to, norbornenyl and bicyclo[2.2.2]oct-2-enyl. Fused bicyclic cycloalkenyl ring systems contain a monocyclic cycloalkenyl ring fused to either a phenyl, a monocyclic cycloalkyl, a monocyclic cycloalkenyl, a monocyclic heterocyclyl, or a monocyclic heteroaryl. The bridged or fused bicyclic cycloalkenyl is attached to the parent molecular moiety through any carbon atom contained within the monocyclic cycloalkenyl ring. Cycloalkenyl groups are optionally substituted with one or two groups which are independently oxo or thia.

The term "halo" or "halogen" as used herein, means —Cl, Br, I or F.

The term "haloalkyl" as used herein, means at least one halogen, as defined herein, appended to the parent molecular moiety through an alkyl group, as defined herein. Representative examples of haloalkyl include, but are not limited to, chloromethyl, 2-fluoroethyl, trifluoromethyl, pentafluoroethyl, and 2-chloro-3-fluoropentyl.

The term "heteroaryl," as used herein, means a monocyclic heteroaryl or a bicyclic ring system containing at least one heteroaromatic ring. The monocyclic heteroaryl can be a 5 or 6 membered ring. The 5 membered ring consists of two double bonds and one, two, three or four nitrogen atoms and optionally one oxygen or sulfur atom. The 6 membered ring consists of three double bonds and one, two, three or four nitrogen atoms. The 5 or 6 membered heteroaryl is connected to the parent molecular moiety through any carbon atom or any nitrogen atom contained within the heteroaryl. Representative examples of monocyclic heteroaryl include, but are not limited to, furyl, imidazolyl, isoxazolyl, isothiazolyl, oxadiazolyl, oxazolyl, pyridinyl, pyridazinyl, pyrimidinyl, pyrazinyl, pyrazolyl, pyrrolyl, tetrazolyl, thiadiazolyl, thiazolyl, thienyl, triazolyl, and triazinyl. The bicyclic heteroaryl consists of a monocyclic heteroaryl fused to a phenyl, a monocyclic cycloalkyl, a monocyclic cycloalkenyl, a monocyclic heterocyclyl, or a monocyclic heteroaryl. The fused cycloalkyl or heterocyclyl portion of the bicyclic heteroaryl group is optionally substituted with one or two groups which are independently oxo or thia. When the bicyclic heteroaryl contains a fused cycloalkyl, cycloalkenyl, or heterocyclyl ring, then the bicyclic heteroaryl group is connected to the parent molecular moiety through any carbon or nitrogen atom contained within the monocyclic heteroaryl portion of the bicyclic ring system. When the bicyclic heteroaryl is a monocyclic heteroaryl fused to a phenyl ring or a monocyclic heteroaryl, then the bicyclic heteroaryl group is connected to the parent molecular moiety through any carbon atom or nitrogen atom within the bicyclic ring system. Representative examples of bicyclic heteroaryl include, but are not limited to, benzimidazolyl, benzofuranyl, benzothienyl, benzoxadiazolyl, benzoxathiadiazolyl, benzothiazolyl, cinnolinyl, 5,6-dihydroquinolin-2-yl, 5,6-dihydroisoquinolin-1-yl, furopyridinyl, indazolyl, indolyl, isoquinolinyl, naphthyridinyl, quinolinyl, purinyl, 5,6,7,8-tetrahydroquinolin-2-yl, 5,6,7,8-tetrahydroquinolin-3-yl, 5,6,7,8-tetrahydroquinolin-4-yl, 5,6,7,8-tetrahydroisoquinolin-1-yl, thienopyridinyl, 4,5,6,7-tetrahydrobenzo[c][1,2,5]oxadiazolyl, and 6,7-dihydrobenzo[c][1,2,5]oxadiazol-4(5H)-onyl. In certain embodiments, the fused bicyclic heteroaryl is a 5 or 6 membered monocyclic heteroaryl ring fused to either a phenyl ring, a 5 or 6 membered monocyclic cycloalkyl, a 5 or 6 membered monocyclic cycloalkenyl, a 5 or 6 membered monocyclic heterocyclyl, or a 5 or 6 membered monocyclic heteroaryl, wherein the fused cycloalkyl, cycloalkenyl, and heterocyclyl groups are optionally substituted with one or two groups which are independently oxo or thia.

The term "hydroxy" as used herein, means an —OH group.

The term "nitro" as used herein, means a —NO_2 group.

The term "oxo" as used herein means a =O group.

The term "saturated" as used herein means the referenced chemical structure does not contain any multiple carbon-carbon bonds. For example, a saturated cycloalkyl group as defined herein includes cyclohexyl, cyclopropyl, and the like.

The term "thia" as used herein means a =S group.

The term "unsaturated" as used herein means the referenced chemical structure contains at least one multiple carbon-carbon bond, but is not aromatic. For example, a unsaturated cycloalkyl group as defined herein includes cyclohexenyl, cyclopentenyl, cyclohexadienyl, and the like.

As used herein, the term "cell" is meant to refer to a cell that is in vitro, ex vivo or in vivo. In some embodiments, an ex vivo cell can be part of a tissue sample excised from an organism such as a mammal. In some embodiments, an in vitro cell can be a cell in a cell culture. In some embodiments, an in vivo cell is a cell living in an organism such as a mammal.

As used herein, the term "contacting" refers to the bringing together of indicated moieties in an in vitro system or an in vivo system. For example, "contacting" a parasite with a compound includes the administration of a compound described herein to an individual or patient, such as a human, infected with the parasite, as well as, for example, introducing a compound into a sample containing a cellular or purified preparation containing the parasite.

In another aspect, the invention provides methods for monitoring *T. gondii* infection in a subject, comprising monitoring levels in a blood sample from the subject of one or more markers selected from the group consisting of clusterin, oxytocin, PGLYRP2 (N-acetylmuramoyl-L-alanine amidase), Apolipoprotein A1 (apoA1), miR-17-92, and miR-124, wherein a change in levels of the one or more circulating markers compared to control correlates with *T. gondii* infection in the subject. The inventors have discovered these specific markers of active *T. gondii* infection, as described in the examples that follow.

The blood sample can be whole blood, serum, blood plasma, or any other suitable blood sample in which circulating IgG from a person with toxoplasmosis may be present. For example, the blood sample may be a plasma sample. As used herein, a "plasma sample" means blood plasma, the liquid component of blood, and is prepared, for example, by centrifugation of whole blood to remove blood cells. As used herein, a plasma sample also includes a blood serum sample, in which blood clotting factors have been removed.

Any suitable control can be used, including but not limited to a reference value obtained from one or more subjects that either do not have a *T. gondii* infection, or that are known to have a *T. gondii* infection, a previous blood sample obtained from the same subject, or any other suitable control. It is well within the level of those of skill in the art to determine an appropriate control for an intended use in light of the teachings herein. The change in level from control that correlates with *T. gondii* infection in the subject may be a difference of 10%, 25%, 50%, 100%, or more. In one embodiment, the difference is a statistically significant increase as judged by standard statistical analysis.

The level (e.g., quantity or amount) of a particular biomarker can be measured in the blood sample using a variety of methods known to those of skill in the art. Such methods include, but are not limited to, flow cytometry, ELISA using red blood cell, platelet, or white blood cell lysates (e.g., lymphocyte lysates), and radioimmunoassay.

In one embodiment, the method is used to monitor effect on the subject of a therapy for *T. gondii* infection. In this embodiment, the subject is receiving therapy for a *T. gondii* infection, and the methods permit attending medical personnel to assess efficacy of the therapy. In this embodiment, the blood sample test may, for example, be carried out periodically over time during the course of therapy. In another embodiment, the method is used to diagnose whether the subject is suffering from a *T. gondii* infection. In this embodiment, the subject is suspected of suffering from a *T. gondii* infection based on the presence of one or more symptoms, and the methods can be used to assist in providing a more definitive diagnostic, along with all other factors to be considered by an attending physician.

In various embodiments of these methods:

(a) an increase in level of one or more of clusterin, oxytocin, miR-17-92, or miR-124 compared to control correlates with active *T. gondii* infection; and/or

(b) a decrease in level of one or more of PGLYRP2 or ApoA1 compared to control correlates with active *T. gondii* infection.

In further embodiments of these methods:

(a) a decrease in level of one or more of clusterin, oxytocin, miR-17-92, or miR-124 compared to a level of the one or more markers in a serum sample obtained from the subject at an earlier time point correlates with a positive effect of the therapy in treating active *T. gondii* infection; and/or

(b) an increase in level of one or more of PGLYRP2 or ApoA1 compared to a level of the one or more markers in a serum sample obtained from the subject at an earlier time point correlates with a positive effect of the therapy in treating correlates with active *T. gondii* infection.

In one embodiment, the *T. gondii* infection involves neuronal damage and/or retinal damage in the subject. For example, the *T. gondii* infection may involve neuronal damage selected from the group consisting of neurodegeneration and/or seizures.

In another aspect, the invention provides methods for treating a *T. gondii* infection, comprising administering to a subject with a *T. gondii* infection an amount effective to treat the infection of ApoA1. As shown in the examples that follow, a reduction in apoA1 closely correlates with active *T. gondii* infection. The apoA1 may be administered as a protein therapeutic, or may be administered in an expression construct (such as a recombinant viral vector, etc.) that expresses apoA1 (i.e.: gene therapy).

In one embodiment, the subject to be treated has a decreased level of serum ApoA1 compared to control.

UniProtKB-P02647 (APOA1_HUMAN) (SEQ ID NO: 4)

MKAAVLTLAV LFLTGSQARH FWQQDEPPQS PWDRVKDLAT

VYVDVLKDSG RDYVSQFEGS ALGKQLNLKL LDNWDSVTST

FSKLREQLGP VTQEFWDNLE KETEGLRQEM SKDLEEVKAK

VQPYLDDFQK KWQEEMELYR QKVEPLRAEL QEGARQKLHE

LQEKLSPLGE EMRDRARAHV DALRTHLAPY SDELRQRLAA

RLEALKENGG ARLAEYHAKA TEHLSTLSEK AKPALEDLRQ

GLLPVLESFK VSFLSALEEY TKKLNTQ.

As used herein, the term "individual" or "patient," or "subject" used interchangeably, refers to any animal, including mammals, preferably mice, rats, other rodents, rabbits, dogs, cats, swine, cattle, sheep, horses, or primates, and most preferably humans.

As used herein, the phrase "amount effective", "therapeutically effective amount" or "effective to treat" refers to the amount of active compound or pharmaceutical agent that elicits the biological or medicinal response that is being

sought in a tissue, system, animal, individual or human by a researcher, veterinarian, medical doctor or other clinician.

In certain embodiments, a therapeutically effective amount can be an amount suitable for (1) preventing the disease; for example, preventing a disease, condition or disorder in an individual who may be predisposed to the disease, condition or disorder but does not yet experience or display the pathology or symptomatology of the disease;

(2) inhibiting the disease; for example, inhibiting a disease, condition or disorder in an individual who is experiencing or displaying the pathology or symptomatology of the disease, condition or disorder; or

(3) ameliorating the disease; for example, ameliorating a disease, condition or disorder in an individual who is experiencing or displaying the pathology or symptomatology of the disease, condition or disorder (i.e., reversing the pathology and/or symptomatology) such as decreasing the severity of disease.

As used here, the terms "treatment" and "treating" means (i) ameliorating the referenced disease state, for example, ameliorating a disease, condition or disorder in an individual who is experiencing or displaying the pathology or symptomatology of the disease, condition or disorder (i.e., reversing or improving the pathology and/or symptomatology) such as decreasing the severity of disease; or (ii) eliciting the referenced biological effect (e.g., reducing parasitic loador adverse effects the parasite is causing in the human it infects).

As used herein, the phrase "pharmaceutically acceptable salt" refers to both pharmaceutically acceptable acid and base addition salts and solvates. Such pharmaceutically acceptable salts include salts of acids such as hydrochloric, phosphoric, hydrobromic, sulfuric, sulfinic, formic, toluenesulfonic, methanesulfonic, nitric, benzoic, citric, tartaric, maleic, hydroiodic, alkanoic such as acetic, HOOC —$(CH_2)_n$—COOH where n is 0-4, and the like. Non-toxic pharmaceutical base addition salts include salts of bases such as sodium, potassium, calcium, ammonium, and the like. Those skilled in the art will recognize a wide variety of non-toxic pharmaceutically acceptable addition salts.

Example 1. New Paradigms for Understanding and Step Changes in Treating Active and Chronic, Persistent Apicomplexan Infections

Abstract

Toxoplasma gondii, the most common parasitic infection of human brain and eye, persists across lifetimes, can progressively damage sight, and is currently incurable. New, curative medicines are needed urgently. Herein, we develop novel models to facilitate drug development: EGS strain *T. gondii* forms cysts in vitro that induce oocysts in cats, the gold standard criterion for cysts. These cysts highly express cytochrome b. Using these models, we envisioned, and then created, novel 4-(1H)-quinolone scaffolds that target the cytochrome bc_1 complex Q_i site, of which, a substituted 5,6,7,8-tetrahydroquinolin-4-one inhibits active infection (IC_{50}, 30 nM) and cysts (IC_{50}, 4 μM) in vitro, and in vivo (25 mg/kg), and drug resistant *Plasmodium falciparum*: (IC50, <30 nM), with clinically relevant synergy. Mutant yeast and co-crystallographic studies demonstrate binding to the bc_1 complex Q_i site. Our results have direct impact on improving outcomes for those with toxoplasmosis, malaria, and ~2 billion persons chronically infected with encysted bradyzoites.

Toxoplasma gondii infections can cause systemic symptoms, damage and destroy tissues[1-1], especially eye and brain[1-10] and cause fatalities[S1-20] Primary infections may be asymptomatic, or cause fever, headache, malaise, lymphadenopathy, and rarely meningoencephalitis, myocarditis, or pericarditis[9,11,12]. Retinochoroiditis and retinal scars develop in up to 30% of infected persons[1,7,13] and epilepsy may occur[6,14] In immune-compromised and congenitally infected persons, active infection frequently is harmful[1-10]. Recrudescence arises from incurable, dormant cysts throughout life[6,7,9,10] In rodents, chronic infection alters fear, smell, reward pathways, neurotransmitters such as GABA and dopamine, and causes abnormal neurologic functions[15]. Although this parasite is present in the brains of 2-3 billion persons worldwide, consequences are unknown. Neurobehavioral abnormalities and differences in serum cytokines, chemokines, and growth factors were associated with seropositivity in humans[16, 17].

Current treatments against active *T. gondii* tachyzoites can have side effects such as hypersensitivity, kidney stones, and bone marrow suppression, limiting their use[10]. Latent bradyzoites are not significantly affected by any medicines[6]. Atovaquone partially, and transiently, limits cyst burden in mice[18], but resistance develops with clinical use[19-24]. Thus, *T. gondii* infection is incurable with recrudescence from latent parasites posing a continual threat. Estimates of costs for available, suboptimal medicines to treat active, primary ocular, gestational and congenital infections, in just the U.S. and Brazil, exceed $5 billion per year. Improved medicines are needed urgently. Molecular targets shared by *T. gondii* and Plasmodia make re-purposing compounds a productive strategy.

Critical flaws and limitations of available methods and models for developing medicines to cure *T. gondii* infections include lack of in vitro culture systems for cysts and scalable, easy to use animal models for screening compounds. To address these challenges, we characterized the EGS parasite, isolated in 1994 from amniotic fluid of a congenitally infected Brazilian fetus[24a], that form cyst-like structures in vitro[25]. In our characterization of EGS in vitro, herein, we discovered that true cysts develop, making EGS especially useful for drug development. EGS parasites can infect zebrafish, and we have characterized this, as well as a fluorescent tachyzoite and cyst assay in this new model[26]. Further, cytochrome bc_1 expression is markedly increased in encysted EGS bradyzoites suggesting cytochrome bc_1 might be a viable drug target for this life stage. This mitochondrial membrane bound protein complex cytochome bc_1, part of the electron transport chain responsible for generating ubiquinone for pyrimidine biosynthesis in *Plasmodium*, is the molecular target of the naphthoquinone, atovaquone[27-52]. Partial efficacy, rapid emergence of drug resistance in malaria and toxoplasmosis limit clinical usefulness of atovaquone. We present new 4-(1H)-quinolone scaffolds that target the Q_i site of cytochrome bc1 in apicomplexan parasites. Our lead 5,6,7,8-tetrahydroquinolin-4-one compound, MJM170, is highly effective against apicomplexan parasites and has substantially enhanced solubility compared with other reported quinolones due to its' new scaffold. Direct visualisation in the crystallographic structure opens the way to design a new generation of compounds for both parasites.

Results

Characterization of EGS Strain Develops Novel In Vitro Models to Test Compounds.

Genotyping and Phylogenetic Analysis of EGS: We isolated and sequenced genomic DNA from the EGS[25] (25) parasite, which formed cysts when grown in human foreskin fibroblasts (HFF) in culture. Phylogenetic analysis based on 796,168 SNPs across 62 *T. gondii* genomes revealed that

EGS is closely related to other Brazilian strains including TgCatBr1, TgCatBr18 and TgCatBr25 and ancient South American MAS. All these grouped to clade B, haplogroup 4 and 8. Full genome sequence analysis of EGS compared with canonical and geographically closely related parasite genomic sequences reveal a non-synonymous mutation and disordered c terminal sequence in Apetela 2 (AP2) IV-iv, a bradyzoite repressor. EGS differs from other isolates by non-synonymous SNPs in Apetela 2 IV-iv, M=>I (570) and a disordered area beginning at 821, GGNRPHYH-VAKQEWRVRYYMNGKRKMRTYSAKFY GYETAHT-MAEDFAHYVDKHE (SEQ ID NO: 1). AP2 IV-iv is a member of the plant-like transcription factor family unique to apicomplexan parasites. This AP2 represses tachyzoite to bradyyzoite conversion,[56] among other differences. Because AP2 IV-iv is a bradyzoite gene expression repressor[56], a mutation could create a parasite like EGS that remains as an encysted bradyzoite.

Phenotypes of EGS in Human cells in vitro, and in Cats and Mice:

EGS in human foreskin fibroblasts (HFF). In vitro, these EGS parasites form cysts that enlarge over ~48-96 hours and then destroy monolayers as single cell organisms. This created novel, useful in vitro models. Cyst walls are thick in electron micrographs (data not shown). Cyst-like structures' perimeters demonstrate dolichos, with bradyzoites within them staining with BAG1 and nuclei with Dapi. Kinetic analysis of EGS in HFF cultures, 2, 18, and 72 hours after infection, RNA-seq and MiR-seq results demonstrated varied expression signatures over time in culture with expression of bradyzoite markers by 18 hours and Apetela 2 signatures by 2 hours.

Cats fed EGS in HFF cultures or mouse brain produce oocysts. When HFF tissue cultures with these cyst structures were fed to cats, they developed the classic, gold standard bradyzoite phenotype of producing oocysts in two replicate experiments. All other *T. gondii* strains cultured for more than 30 passages, as EGS was since the 1990s, lose the ability to produce oocysts when fed to cats (JP Dubey, personal observations). This experiment established that these were true bradyzoites in cysts formed in vitro under standard culture conditions. Oocysts also formed 10 days after feeding cats mouse brains infected with EGS stably transfected with tachyzoite SAG1 promoter-driven mcherry, and bradyzoite BAG1 promoter-driven green fluorescent protein (GFP), and merozoite promoter-driven blue fluorescent protein, engineered to facilitate creation of automated, scalable in vitro and in vivo assays. In vitro, these promoters did not provide a fluorescence signal robust enough to detect differences between 2×10^5 and 650 parasites useful in scalable assays (data not shown).

EGS is virulent in Mice. When these EGS oocysts were fed to mice they produced disease indistinguishable from other virulent Brazilian strains. Oocysts given to mice per-orally created an illness and histopathology phenotypically characteristic for typical, virulent parasites causing dose related proliferation of *T. gondii* with necrosis in terminal ileum, pneumonia at 9-10 days, with brain parasites by 17 days and dose-related mortality.

EGS has a bradyzoite/cyst morphology and alters the transcriptomes of the biologically relevant human monocytic cell line MM6 and human primary neuronal stem cells (NSC). Human cells particularly relevant to human toxoplasmosis were infected with different strains of *T. gondii* to better characterize EGS parasites. Immunofluorescence staining of EGS-infected MM6 and NSC cultures revealed the development of cysts (FIG. **1**A) and accordingly, EGS gene expression resembled that of bradyzoites when compared to equivalent infections done with GT1, ME49 or VEG strains. Interestingly, EGS transcription was influenced by host cell type (FIGS. **1**A-**1**D). Transcriptomics using host mRNA and miR profiling of EGS cultures in MM6, and NSC cells for 18 hours demonstrated that this parasite modulates host transcripts involved in protein misfolding, neurodegeneration, endoplasmic reticulum stress, spliceosome alteration, ribosome biogenesis, cell cycle, epilepsy, and brain cancer among others (FIGS. **1**A-**1**D). The number of genes significantly up or down regulated in MM6 and NSC cells compared to uninfected controls are depicted in FIGS. **1**A-**1**D. Overexpressed genes differ from those of GT1, ME49 and VEG tachyzoite-infected human NSC cells (FIG. **2**A), but modify the same or connected pathways (McLeod et al, unpublished observations). Hsa-miR-708-5p was the most affected miRNA (down-modulated) by EGS (FIG. **1**D). miR-708-5p is a regulator that promotes apoptosis in neuronal and retinal cells, which could maintain a niche for EGS-like encysted bradyzoites to persist.

EGS transcripts demonstrate importance of cytochromes and key Apetela 2 transcription factors in this life cycle stage. EGS transcripts in HHF, MM6, and NSC cells were enriched for genes transcribed in bradyzoites, including known bradyzoite transcripts, certain Apetela 2s and cytochrome b and other cytochromes. Among transcripts with the most increased fold change in EGS across all three cell lines were: cytochrome b; cytochrome c oxidase subunit III subfamily protein; apocytochrome b; cytochrome b, putative; and cytochrome b (N-terminal)/b6/petB subfamily protein. Other over-expressed genes include bradyzoite transcription factor AP2IX-9 and plant-like heat-shock protein BAG1 (FIG. **2**A).

Identifying Novel and Efficacious Compounds Against *T. gondii* Cytochrome Bc_1,

Increased expression of cytochromes in EGS made it pertinent to synthesize and test an endochin-like quinolone (ELQ) 271, which was previously reported to inhibit *T. gondii* cytochrome bc_1 Q_i site and reduce, but not eliminate, brain cyst numbers in mice[27]. ELQ271 also inhibited EGS in vitro (FIG. **2**B bottom) demonstrating that our in vitro model correlates with previously reported partial activity of ELQ271 against bradyzoites in cysts in mouse brain. Vivoporter-PMOs inhibiting cytochrome bc_1 had a modest effect on tachyzoite replication and a small effect on size and number of EGS cysts (FIG. **2**B top). Minimal effect might be related to limited entry of vivoporter into cysts or mitochondria.

ELQs have been a focus for drug development for malaria (ELQ 300) and toxoplasmosis (ELQ 271 and 316) as they were reported to be potent and selective (versus human cytochrome bc_1) inhibitors of *P. falciparum* cytochrome bc_1 at nanomolar concentrations[27]. ELQs are part of the 4-(1H)-quinolone class of cytochrome bc_1 inhibitor[36,42,40,45,49,52,53,54,56-62] and (Dogget et al, 13th International Toxoplasmosis Meeting Abstract. Gettysburg Pennsylvania, June 2015), a scaffold that suffers from limited aqueous solubility. Another aspect of inhibitor design for this system is minimizing the inhibition of mammalian cytochrome bc_1, which shares ~40% sequence identity to the *T. gondii* ortholog within the Q substrate sites. Thus, we set out to design potent and selective inhibitors of *T. gondii* cytochrome bc_1 with improved solubility (FIGS. **3**A and **3**B) compared to known quinolone-based inhibitors. Noting the previous work of GSK on the preclinical development of Clopidol derivatives which led to terminating studies secondary to toxicity in the rat models, as another serious deficiency[63] and the incorporation of the diphenyl ether group onto the central 4-(1H)-quinolone core as reported by Riscoe et al.[27], we focused on the central core ring system. Doggett, Riscoe et al's[27] ELQ 271 (FIGS. **3**A and **3**B) was reported to be ineffective against yeast with a mutation in the

Q_i site. Nonetheless, it recently was shown that ELQs can bind both Q_i and Q_o depending on subtle chemical changes[61-3]. As a result of our initial efforts, a 5,6,7,8-tetrahydroquinolone (MJM170, 2) displayed promising results. (FIGS. **3**A and **3**B; Table 2). We chose ELQ271 for comparison because it had the greatest activity at the lowest dose (5 mg/kg) in the mouse model of Doggett despite the higher cytotoxicity toward human fibroblasts in the in vitro toxicity studies.[27]

MJM170 is a potent inhibitor of *T. gondii* tachyzoites (RH—YFP strain, IC_{50} 0.03 μM) and bradyzoites (EGS strain, IC_{50} 4 μM), equipotent to ELQ271 (Table 2, FIG. **4**A-**4**E). MJM170 showed 10-fold improved aqueous kinetic solubility (pH 7.4) over ELQ 271 (Table 2), 3-fold improved FaSSIF/FeSSIF (pH 6.5) solubility, with similar human microsomal stability profiles (146 vs 172 minutes). A different method from that in reference[49] was used. MJM170 has a significantly decreased mouse microsomal stability compared to ELQ 271 (20 vs>200 minutes). MJM170 was further evaluated with MDCK-MDR1 cells (a measure of blood-brain barrier permeability) and results suggest that MJM170 could cross the blood brain barrier and not suffer from P-glycoprotein efflux These data highlight the potential of the 5,6,7,8-tetrahydroquinolin-4-one scaffold for further hit-to-lead development.

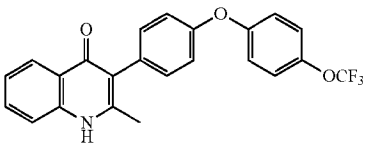
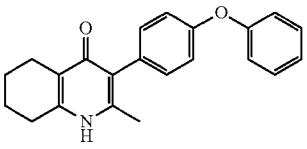
(data not shown). Translucent zebra fish can be infected with EGS, other *T. gondii* that make cysts, and RH YFP preparing for a novel model for scalable screening (FIG. **5**D).

Cytochrome bc_1 Q_i is the binding site of MJM170 which is potent against *Plasmodium falciparum*: and yeast. Tetrahydroquinolone binds to the Q_i site of cytochrome bc_1: Studies to determine whether cytochrome bc_1 Q_i is the molecular target of MJM170 initially included studies of resistance of yeast and *P. falciparum* with known cytochrome b Q_i mutations predicted to cause a steric clash with MJM170 (FIGS. **6**A-**6**G). Recently, we reported co-crystal structures of GSK's cytochrome bc_1 inhibitors bound to bovine cytochrome bc_1 at the Q_i site[52] demonstrating that these pyridone inhibitors and other structurally related inhibitors bind to an alternative site to atovaquone on cytochrome bc_1. This structure allowed us to model MJM170 within the Q_i site using the Maestro Suite from Schrödinger. This molecular modelling predicted steric clashes in mutant yeast and *P. falciparum* cytochrome bc_1 with MJM170 (FIG. **6**A).

Co-crystallization of MJM170 with bovine cytochrome bc_1 and modelling of the *T. gondii* enzyme confirm the target. Co-crystallization validates predictions made with modelling and confirmed using assays with *S. cerevesiae*

TABLE 2

Comparison of ELQ 271 and MJM170 in our biological assays: inhibition of apicomplexan parasites and ADME/Tox. ELQ 271 was synthesised in-house.

Compound	ELQ 271	MJM170
Structure		
Mol. Wt.	411.4	331.4
T. gondii Tachyzoite IC_{50} μM	0.03	0.03
T. gondii Bradyzoite IC_{50} μM	1	4
P. falciparum IC_{50} μM[a]	0.03 (D6) 0.09 (TM91C235) 0.10 (W2) 0.13 (C2B)	0.01 (D6) 0.03 (TM91C235) 0.03 (W2) 0.01 (C2B)
HFF Toxicity CC_{50} μM	20	20
Kinetic Solubility PBS pH 7.4 μM [b]	0.15	1.97
FaSSIF Solubility pH 6.5 μM [b]	3.4	9.8
Human microsomal stability $T_{1/2}$ mins [b]	171.9	146.3
Mouse microsomal stability $T_{1/2}$ mins [b]	>200	21.0
MDCK-MDR1 P_{app}A-B × 10^6 cm/s [b]	N.D.	32.1
MDCK-MDR1 Efflux Ratio [b]	N. D.	1.23

[a]The D6 strain (Sierra Leone) is drug sensitive, the TM91C235 (Thailand) is multi-drug resistant, the W2 strain (Thailand) is chloroquine resistant, and the C2B strain is multi-drug resistant with pronounced resistance to atovaquone.
[b] ADME carried out by ChemPartner Shanghai Ltd. N.D. not determined. Human and mouse microsomal stability differs as is known to occur for other compounds such as TMP/SMX.

MJM 170 is effective in vivo against tachyzoites, and modestly against bradyzoites in cysts of mice, and development of a scalable zebrafish assay. MJM170 was highly efficacious against RH (FIG. **5**A) and Prugniaud (FIG. **5**B) strain tachyzoites in mice at 25 mg/kg without toxicity for 5 days ($p<0.00$), and modestly reduced numbers of Me49 strain cysts established >2 months earlier when treated with 12.5-25 mg mg/kg for 17 days ($p<0.002$) (FIG. **5**C). In analysis of parallel histopathology, there was a similar trend

mutants (FIG. **6**D). There was no steric clash for *P. falciparum*—model based upon this crystal structure, consistent with in vitro assays (FIGS. **6**E-**6**F). MJM170 was co-crystallized with bovine cytochrome bc_1 and the resulting good quality electron density maps allowed for unambiguous placement of MJM170 within the Q_i site (FIG. **6**F). The planar region of the quinolone group is held between heme b_H and Phe220 and the additional ring further extends into the hydrophobic cavity at the apex of the binding site

towards Pro24 and Ile27. The carbonyl group of the compound is surrounded by Ser35, Asp228 and the carbonyl of Trp31, while its amine moiety lies between His201 and Ser205. The diphenyl ether group extends outwards towards the hydrophobic residues Ile39 and Ile42 and forms a stacking interaction with Phe18 (FIG. **6**G).

Surrogate assays demonstrate efficacy of compounds, providing target validation and added value as MJM170 is effective against wild type but not M221Q(F) mutant yeast. Mutants of *S. cerevesiae* were used to further confirm the molecular target of MJM170 (FIG. **6**D), and documented that the Q_i domain in cytochrome b is essential for its efficacy. This approach provided insight into binding of compounds to the enzyme. Crystallographic structure of bovine cytochrome bc_1 with GSK932121[52] indicates that certain amino acids are critical in tetrahydroquinolone binding and explains why there is inhibition by certain compounds. Previous studies reported that no cross-resistance is observed between ELQs and atovaquone in *P. falciparum*. This is rationalised as atovaquone binds the Q_o site on cytochrome bc_1. A yeast M221Q substitution within the Q_i site displayed resistance to ELQ inhibition further confirming that this to be the target site[27]. MJM170 and ELQ271 were effective against *S. cerevisiae* wild type parental AD1-9 strain at 1 mM, 100, 5 and 1 μM when grown on non-fermentable, glycerol medium forcing reliance on ATP production for respiration. Yeast strains with point mutations in the cytochrome b gene that substitute methionine by glutamine (M221Q) or phenylalanine (M221F) at position 221 in the Q_i site predicted to yield a steric clash upon inhibitor binding were resistant to MJM170 (FIG. **6**D).

Tetrahydroquinolones are potent against wild type *P. falciparum*, and *P. falciparum* G33A/V and other drug resistant mutants but not DHODH mutant. MJM170 is highly effective against *P. falciparum* (Table 2) including multiple strains resistant to available antimicrobials and a cytochrome bc_1 Q_i mutant (FIGS. **7**A-**7**C). Resistance against transgenic *P. falciparum* yeast DHODH mutant strain indicates MJM170 affects mitochondria suggesting that the mode of action against *P. falciparum* is through inhibition of electron transport (FIG. **7**A).

Potentially clinically useful combinations with tetrahydroquinolone demonstrated in synergy studies. To determine whether there might be clinically relevant synergies and additive effects, combinations of MJM170 with other clinically available and useful compounds also were tested. Earlier, we had found cycloguanil and related biguanide tnazines[64] were active against *T. gondii* tachyzoites and *P. falciparum* making it relevant to test them in combination with MJM170. We observed modest synergy in vitro for atovaquone, additive effect with cycloguanil, and antagonism with BRD6323, a Q_i inhibitor for *P. falciparum* (FIG. **7**B). Combining atovaquone with proguanil (active component cycloguanil) as Malarone^R for malaria provides an approach to reduce selection of drug resistant *plasmodium* mutants.

Discussion

The results presented here offer a molecular understanding and therapeutic strategies for one of the most common parasitic infections of human brain and eye, and that persists across lifetimes in around 2 billion people worldwide. We have developed new models to facilitate discovery of curative treatments for toxoplasmosis. We have characterized the Brazilian *T. gondii* isolate called EGS that was known to be morphologically similar to encysted bradyzoites in tissue culture. We further validate the cystic nature of these EGS infected cultures, since they are able to induce the intestinal life cycle when fed to cats ultimately resulting in oocyst secretion. This is the first such description of this phenotype and provides definitive proof that this unique parasite has a true cyst phenotype when maintained in vitro. Our data also provide a number of other major conceptual advances on EGS by demonstrating the following: (i) Genome sequencing of this EGS isolate demonstrates that EGS has a typical Brazilian virulent genotype and phylogeny, (ii) EGS is a haplogroup 4 *T. gondii*. Consistent with a genotype that is known to be pathogenic and virulent for mice, we demonstrate that EGS oocyst induced infection is similar to that of other virulent Brazilian parasites. For example, mice fed EGS oocysts demonstrated ileal parasites causing necrosis, as well as pneumonitis, encephalitis and systemic infection leading to death. This indicates that the ability to form cysts in culture does not alter the pathogenicity of EGS in mice. However, potentially relevant to its in vitro bradyzoite phenotype, full genome sequencing revealed that it has nonsynonymous single nucleotide repeat sequence differences from other Brazilian and canonical U.S. and European parasites which do not share its in vitro bradyzoite phenotype. EGS has a non-synonymous mutation in a bradyzoite repressor, Apetela 2 (AP2) IV-iv, plant like transcription factor. AP2s interact with HATs and HDACs to modulate transcriptional signatures in apicomplexan parasites[55]. This Apetela 2, plant-like transcription factor gene, AP2IV-4, represses bradyzoite genes during the tachyzoite cell cycle, thereby preventing commitment to the bradyzoite developmental pathway[56]. If the observed substitution or disordered N terminus results in a defective or non-functional molecule, this could provide an explanation for the observed bradyzoite phenotype of EGS parasites. This is consistent with our findings that EGS in HFF forms cysts by 24 hours, characterized by BAG1, and *Dolichos* staining at 24, 48, and 96 hours after infection. These cysts gradually enlarge until 48-96 hrs in culture, when single *T. gondii* begin to destroy HFF monolayers. These are the cultures of bradyzoites in cysts that when fed to cats ~48 hours after infection form oocysts which are virulent in mice, providing definitive proof of an in vitro bradyzoite phenotype for the EGS strain of *T. gondii*.

The transcriptomic studies with this EGS isolate have provided critical insights into host cell mechanisms that are a prominent part of the ability of the encysted parasite to persist in this untreatable life cycle stage, and biologic consequences of such persistent infection. RNAseq and miR seq of EGS infected human host cells included human fibroblasts, monocytic and neuronal stem cells with this encysted EGS strain parasite. These provide an understanding of the types of perturbations of biologically relevant host cells this bradyzoite life cycle stage can cause, providing insights into unique aspects of pathogenesis of this infection with untreatable cysts and its consequences. We found that EGS modifies critical host cell pathways. For example we find in vitro modulations of host cell pathways in human, primary neuronal stem cells are the same as those associated with modulation of host cell replication as seen with malignancies, and in neurodegenerative diseases. Further, it is noteworthy that the level of a microRNA that specifies apoptosis in eye and brain cells is markedly down modulated by this EGS bradyzoite which would inhibit host protective apoptotic mechanisms allowing parasites to persist in brain and eye without a critical protective mechanism. EGS, as an encysted bradyzoite, clearly alters biologic processes including cell cycle, cell death, alternative splicing, protein syn-

thesis, protein folding and ubiquitination and down regulates hsa-miR-708-5p that specifies apoptosis in neuronal and retinal cells[65].

RNA and MiR sequencing and transcriptomic analyses of the EGS parasites also identified molecular targets that are critical for the bradyzoite life cycle stage in the parasite as well. These molecular targets include cytochrome b, as critically increased in dormant, encysted parasites. Cytochrome b was increased along with known cyst constituents like enolase 1, Cyst wall protein, Lactate dehydrogenase 2. bradyzoite antigen 1, Apetela 2 plant like transcription factors not present in animals, such as AP2 IX-ix, and cytochrome oxidase. Our work provides a new means to identify stage specific molecular targets, and emphasizes that cytochrome bc 1 complex is a critical target. The transcriptome of EGS parasites in HFF over time are similar to those of in vivo bradyzoites in terms of known critical genes modified. Finally, EGS presents a much-needed assay for identifying novel molecular targets present in bradyzoites in vitro. EGS was also useful to evaluate the effect of inhibitors on encysted bradyzoites in vitro.

Recent crystallographic studies with the bovine cytochrome bc_1 complex allowed us to rationally design a novel compound to target the Q_i site of cytochrome b. Our novel compound was designed to address issues with poor solubility of existing quinolone/pyridone Q_i inhibitors. One of these compounds MJM170, a substituted 5,6,7,8-tetrahydroquinolin-4-one inhibits active infection (IC_{50} 30 nM) and cysts (IC_{50} 4 μM) in vitro, and in vivo (25 mg/kg). It is predicted to cross the blood brain barrier with no efflux as demonstrated in an in vitro MDR1-MDCK permeability assay (Table 2), indicating this class of compounds have promise for treatment of central nervous system infections. When we tested MJM170 against wild type and multi-drug resistant *P. falciparum*, we found it was also potent (IC_{50}<30 nM against all strains). In combination studies, MJM170 was identified as additive with cycloguanil and modestly synergistic with atovoquone. Studies of yeast and malaria mutants, as surrogate assays, and co-crystallography studies with bovine cytochrome bc_1 confirm the mechanism of action/target for MJM170. The co-crystal structure of MJM170 in complex with bovine cytochrome bc_1 reveals a clear binding mode within the Q_i site. Using homology models of the apicomplexan Qi sites, there are clear differences between the binding sites of the apicomplexan and mammalian orthologs which can be used to fine-tune the selectivity of our scaffold towards apicomplexan bc_1. The larger binding pocket of the apicomplexan versus the mammalian bc_1 may provide a way forward to increase selectivity. Our work provides a conceptual and a practical step change forward that provides a foundation for further testing and improvements to efficacy, toxicity, solubility, oral absorption, large animal toxicology that will be needed to reach the clinic. Our work reported herein not only provides new and important insights into the biology of *T. gondii*, especially the bradyzoite life cycle stage and the remarkable effects of this parasite on its human host's cells, but also provides critical molecular targets and new methods to identify others. Armed with this information, a novel scaffold with intrinsically higher solubility than the equivalent quinolone has been designed with holds promise towards developing a much-needed curative medicine for those with toxoplasmosis, malaria, and ~2 billion persons chronically infected with presently incurable, encysted bradyzoites which persist and can recrudesce lifelong.

Methods

All methods were carried out in accordance with approved guidelines set at the University of Leeds by the Education & Training Resources office and all experimental protocols were approved by the IRB committees; University of Chicago Institutional Animal Care and Use Committee (IACUC) and all experimental protocols were approved by the IRB committee; United States Department of Agriculture IACUC and all experimental protocols were approved by the IRB committees; J Craig Venter Institute Research ethics committee; University of Liverpool UK Office for Research Integrity (UKRIO) and all experimental protocols were approved by the IRB committees; Harvard School of Public Health HMS IACUC and all experimental protocols were approved by the IRB committees; The Broad Institute IACUC and all experimental protocols were approved by the IRB committees; Walter Reed Army Institute of Research Division of Human Subjects Protection (DHSP) and all experimental protocols were approved by the IRB committees; Oregon State University IACUC and all experimental protocols were approved by the IRB committees; Institute for Systems Biology ethics committee; Albert Einstein College of Medicine IACUC and all experimental protocols were approved by the IRB committees; Strathclyde University Ethics Committee (UEC) and all experimental protocols were approved by the IRB committees; Institute for Integrative Biology of the Cell IACUC and all experimental protocols were approved by the IRB committees, and the Centre national de la recherche scientifique IACUC and all experimental protocols were approved by the IRB committees.

Cells and Parasites for Work with *T. gondii*

Cells: The cells utilized for *T. gondii* assays included human foreskin fibroblasts (HFF), Human MonoMac 6 cells (MM6), and Neuronal Stem cells (NSC) from a temporal lobe biopsy.

Toxoplasma gondii. The strains of *T. gondii* utilized in this work were: RH—YFP Tachyzoites of the RH—YFP strain were passaged in human foreskin fibroblasts (HFF cells); EGS-Bradyzoite assays use the EGS strain, isolated from amniotic fluid of human with congenital toxoplasmosis; Other strains used are: Me49; Prugniaud; Beverly; Veg; GT1. All other than EGS are *T. gondii* tachyzoites. These parasites are passaged in HFF.

Isolation of DNA and RNA. EGS single celled organisms were grown in Human Foreskin Fibroblasts, filtered free of host cells. gDNA was isolated and processed for sequencing as described. For isolation of RNA RIN scores were >8.

Gene Sequencing, Genomics, RNA and MiR Sequencing, Systems Analysis, Metabolomics

Genome sequencing of *T. gondii* EGS strain. A single Illumina paired-end barcoded library was prepared from tachyzoite gDNA with Illumina TrueSeq library preparation kit. The library was then sequenced using 100 bp pared-end reads in one ninth of a lane of an Illumina HiSeq 2000 machine to generate ~2 Gbp of genome sequence.

Single nucleotide polymorphism (SNP) identification and annotation. Illumina genome sequencing reads from EGS or downloaded from GenBank SRA database for GT1 (SRR516419), VEG (SRR516406) and TgCatBr1 (SRR350737) were aligned to the *T. gondii* ME49 reference genome assembly (ABPA02000000, ToxoDB release 13.0) with Bowtie2 and realigned around gaps using the GATK toolkit. SNP calls were done simultaneously across all four strains with samtools utility mpileup, requiring a minimum SNP coverage of 5 reads and an alternative allele frequency of 0.8 or higher, given the haploid nature of these genomes. Thereafter, SnpEff and a gff3 file containing the annotation

of *T. gondii* ME49 downloaded from ToxoDB v13.0 were used to classify the different types of mutations identified in each strain. Allelic variants that were different between EGS and the rest of the strain were considered EGS-specific.

Phylogenetic network analysis. A total of 790,168 single nucleotide polymorphisms spanning the entire *T. gondii* genome from 62 different strains representing all major haplogroups were downloaded from ToxoDB, combined with SNP data from the same sites from the EGS strain and directly incorporated as a FASTA file into SplitsTree v4.13.1 to generate unrooted phylogenetic networks using a neighbor-net method.

Differential gene expression (DGE) analysis. Total RNA extracted from human cell cultures infected (or not) with a number of *T. gondii* strains for 2 h, 18 h or 48 h was treated with miRNeasy Mini Kit columns (Qiagen) following manufacturer instructions to separate mRNA and miRNA fractions. Afterwards, Illumina barcoded sequencing libraries were constructed with TruSeq RNA Sample Preparation Kits v2 (Illumina) for mRNA and miRNA TruSeq Small RNA Library Preparation Kit (Illumina) for miRNA. Libraries were sequenced as 100 bp single reads with Illumina HiSeq 2000 apparatus in pulls of 6 or 9 samples per lane for mRNA (yield ~3 Gbp per sample) and miRNA (yield ~2 Gbp per sample) libraries respectively. For protein coding genes, reads were mapped to the human (release GRCh38) and *T. gondii* ME49 strain (ToxoDB release 13.0) reference genome assemblies and annotations with CLC Genomic Workbench software (CLC Bio-Qiagen, Aarhus, Denmark) and raw read counts per gene were then analyzed with the R package EdgeR using a generalized linear model likelihood ratio test to identify genes that are differentially expressed among samples.

For miRNA DGE analysis, reads were depleted of adaptor and primer sequences and mapped to the human reference genome assembly (GRCh38) and the miRNA annotation from miRBase v21 (see the mirbase web site) with CLC Genomic Workbench software. Identification of human miRNA genes that are differentially expressed across treatments was carried out with EdgeR from raw read counts per miRNA gene using a generalized linear model likelihood ratio test.

For both mRNA and miRNA DGE analyses p-values were adjusted for multiple hypotheses testing using the False Discovery Rate method. MDS plots and heat maps were generated with the plotMDS tool from EdgeR and the R tool heatmap. Differentially expressed genes (DEGs) in MM6 and NSC cell lines infected with EGS parasites were identified under the criteria of 1% FDR and absolute log 2-fold-change >1.5 (i.e. fold-change >2 and <0.5 for up- and down-regulated genes, respectively).

Functional enrichment analysis GO enrichment analyses were performed for up- or down-regulated genes, by using the Database for Annotation, Visualization and Integrated Discovery (DAVID) v6.7. GO slim enrichment analysis was performed for genes carrying potential change-of-function mutations in EGS that were absent in strains ME49, VAND or TgCatBr1. GO slim database was downloaded from QuickGO provided by EMBL-EBI. Using taxonomy id "508771" for the ME49 strain, relevant GO slim terms were retrieved. GO slim enrichment analysis was performed with Fisher's exact test based on the GO slim terms.

Assay for oocyst development in cats. Oocysts were collected from feces of *Toxoplasma*-free cats 3-14 days after feeding infected cell cultures or infected mouse brains. Oocysts were separated from feces by sugar floatation, sporulated in 2% sulfuric acid by aeration at room temperature for 1 week. After removing sulfuric acid oocysts were inoculated orally in to Swiss Webster albino mice. All tissues of mice that died or euthanized were studied histologically after staining with hematoxylin and eosin and by BAG1 antibodies to *T. gondii* as described. (Dubey J P, Ferreira L R, Martins J, McLeod R. Oral oocyst-induced mouse model of toxoplasmosis: effect of infection with *Toxoplasma gondii* strains of different genotypes, dose, and mouse strains (transgenic, out-bred, inbred) on pathogenesis and mortality. Parasitology 139:1-13, Epub 2011. PMID: 22078010; also referred t herein as S39)

Chemical Synthesis. Final compounds had >95% purity determined by high performance liquid chromatography (HPLC) and 300 and/or 500 MHz NMR spectrometers. Liquid chromatography-mass spectrometry (LC-MS) and high resolution mass spectrometers (HRMS) analytical systems were used to determine integrity and purity of all intermediates and final compounds.

Synthesis of 2-methyl-5,6,7,8-tetrahydroquinolin-4-one (6) Platinum oxide (100 mg, 10 mol %) was added to a solution of 4-hydroxy-2-methylquinoline (5, 1.00 g, 6.28 mmol, 1.00 eq) in glacial acetic acid (10.0 ml). The heterogeneous mixture was catalytically hydrogenated under a balloon of hydrogen. After 22 hrs, TLC (10% MeOH-DCM) confirmed complete reaction. The mixture was filtered through celite under vacuum, washing thoroughly with EtOAc. The filtrate was concentrated and the resulting residue purified by column chromatography (10% MeOH-DCM) to give the desired product as a pale yellow oil (917 mg, 5.65 mmol, 89%); Rr 0.14 (10% MeOH-DCM); δ_H (300 MHz, $CDCl_3$) 1.74-1.76 (4H, m, CH_2), 2.29 (3H, s, Me), 2.49-2.52 (2H, m, CH_2), 2.67-2.70 (2H, m, CH_2), 6.16 (1H, s, Ar—H); δ_C (125 MHz, $CDCl_3$) 19.0 (Me), 21.8 (CH_2), 22.1 (CH_2), 27.1 (CH_2), 112.5 (CH), 122.4 (Cq), 146.4 (Cq), 147.0 (Cq), 178.3 (Cq); Spectroscopic data consistent with literature values (JMC, 1993, 36, 1245-54).

Synthesis of 2-methyl-3-iodo-5,6,7,8-tetrahydroquinolin-4-one (7) Butylamine (6.20 ml, 62.8 mmol, 10.0 eq) was added to a suspension of 2-methyl-5,6,7,8-tetrahydroquinolin-4-one (6, 1.02 g, 6.28 mmol, 1.00 eq) in DMF (10.0 ml). To this heterogeneous mixture was added 12 (1.60 g, 6.28 mmol, 1.00 eq) in a saturated solution of KI (6.00 ml). After 20 hrs stirring at R.T., a precipitate formed in the orange solution, Excess iodine was quenched with 0.1 M sodium thiosulfate solution. The precipitate was filtered by vacuum filtration, washed with distilled H_2O and dried (Na_2SO_4) to give the desired product as a colourless solid (1.76 g, 6.09 mmol, quantative yield); δ_H (300 MHz, DMSO-d_6) 1.61-1.70 (4H, m, CH_2), 2.29 (2H, t, J 6.0, CH_2), 2.43 (2H, s, CH_2), CH_3 under DMSO peak.

Synthesis of 2-methyl-3-iodo-4-ethoxy-5,6,7,8-tetrahydroquinoline (8) Potassium carbonate (1.53 g, 11.1 mmol, 2.00 eq) was added to a heterogeneous mixture of 2-methyl-3-iodo-5,6,7,8-tetrahydroquinolin-4-one (7, 1.60 g, 5.56 mmol, 1.00 eq) in DMF (15.0 ml), and the reaction heated to 50° C. for 30 mins. The R.B. flask was removed from the heating mantle and ethyl iodide was added dropwise. The reaction was then heated at 50° C. for 18 hrs. The reaction was cooled to R.T., quenched with water (40 ml). The resulting emulsion formed which was extracted with EtOAc (50 ml). EtOAc layer were washed with water (3×30 ml), brine (3×30 ml), dried (Na_2SO_4) and concentrated to give a pale yellow oil (1.09 g, 3.44 mmol, 61%); Rr 0.88 (1:1 Pet-EtOAc); HPLC (RT=1.67 mins); LCMS (Method A), (RT=1.6 min, m/z (ES) Found MH^+ 318.0); δ_H (500 MHz, $CDCl_3$) 1.49 (3H, t, J 7.0, ethoxy CH_3), 1.73-1.78 (2H, m, CH_2) 1.84-1.88 (2H, m, CH_2), 2.78-2.69 (5H, m, CH_2 &

CH_3), 2.84 (2H, t, J 6.5, CH_2), 3.97 (2H, q, J 7.0, OCH_2); δ_C (125 MHz, $CDCl_3$) 15.6 (CH_3), 22.3 (CH_2), 22.8 (CH_2), 23.6 (CH_2), 29.3 (CH_3), 32.0 (CH_2), 68.4 (OCH_2), 90.9 (Cq), 124.5 (Cq), 158.3 (Cq), 158.9 (Cq), 163.9 (Cq).

Synthesis of 2-methyl-3-(4-phenoxyphenyl)-4-ethoxy-5,6,7,8-tetrahydroquinoline (10) 2-Methyl-3-iodo-4-ethoxy-5,6,7,8-tetrahydroquinoline (8, 0.266 g, 0.839 mmol, 1.00 eq), $Pd(PPh_3)_4$ (0.048 mg, 0.0419 mmol, 5 mol %) and 4-phenoxyphenylboronic acid (9, 0.270 mg, 1.26 mmol, 1.50 eq) were charged to a R.B. flask under $N_2(g)$[49]. Degassed DMF (10.0 ml) was added to the flask followed by 2M K_2CO_3 (1.60 ml). The flask was heated to 85° C. under $N_2(g)$. After 15 mins, TLC (4:1 Pet-EtOAc) confirmed reaction was complete. The reaction was cooled and diluted with EtOAc (15 ml), filtered through celite and partitioned between EtOAc (10 ml) and H_2O (25 ml). Combined organics were washed with H_2O (3×30 ml), then brine (3×30 ml), dried (Na_2SO_4) and concentrated to give a red oil which was purified by column chromatography (3:1 Pet-EtOAc), to give the desired product as a pale yellow oil (0.235 mg, 0.655 mmol, 78%); Rr 0.31 (3:1 Pet-EtOAc); HPLC (RT=3.08 mins); δ_H (300 MHz, $CDCl_3$) 1.04 (3H, t, J 7.0, ethoxy CH_3), 1.76-1.93 (4H, m, 2×CH_2), 2.32 (3H, s, CH_3) 2.72 (2H, t, J 6.0, CH_2), 2.91 (2H, t, J 6.5, CH_2), 3.50 (2H, q, J 7.0, OCH_2), 7.05-7.16 (5H, m, Ar—H), 7.20-7.29 (2H, m, Ar—H), 7.31-7.43 (2H, m, Ar—H); δ_C (125 MHz, $CDCl_3$) 15.7 (CH_3), 22.5 (CH_2), 23.0 (CH_3), 23.3 (CH_2), 23.4 (CH_2), 32.7 (CH_2), 68.2 (OCH_2), 118.6 (CH), 118.9 (CH), 123.4 (CH), 126.8 (Cq), 129.8 (CH), 131.5 (CH), 154.9 (Cq), 156.5 (Cq), 157.1 (Cq), 157.3 (Cq); m/z (ES) (Found: MH^+, 360.1973. $C_{24}H_{26}NO_2$ requires MH, 360.1964).

Synthesis of 2-methyl-3-(4-phenoxyphenyl)-4-ethoxy-5,6,7,8-tetrahydroquinoline (MJM170, 4)[49] Aqueous hydrobromic acid (>48%) (1.00 ml) was added to a solution of 2-methyl-3-(4-phenoxyphenyl)-4-ethoxy-5,6,7,8-tetrahydroquinoline (10, 0.226 mg, 0.630 mmol, 1.00 eq) in glacial acetic acid (2 ml). The reaction was stirred at 90° C. for 5 days, monitoring by LMCS. The reaction was cooled to R.T. and the pH adjusted to pH5 with 2M NaOH. The precipitate was collected by vacuum filtration and recrystallized from MeOH:H_2O to give the desired product as an off-white solid (0.155 g, 0.467 mmol, 74%); HPLC (RT=2.56 mins); δ_H (500 MHz, DMSO-d_6) 1.66-1.72 (4H, m, 2×CH_2), 2.08 (3H, s, CH_3) 2.31 (2H, t, J 6.0, CH_2), 2.56 (2H, t, J 6.0, CH_2), 6.99 (2H, d, J 8.5, Ar—H), 7.06 (2H, d, J 7.5, Ar—H), 7.14-7.18 (3H, m, Ar—H), 7.40-7.43 (2H, m, Ar—H), 11.0 (1H, s, NH); δ_C (125 MHz, DMSO-d_6) 17.7 (CH_3), 21.5 (CH_2), 21.8 (CH_2), 21.9 (CH_2), 26.2 (CH_2), 117.8 (CH), 118.6 (CH), 121.2 (Cq), 123.3 (CH), 123.7 (Cq), 130.0 (CH), 131.4 (Cq), 132.3 (CH), 142.3 (Cq), 143.2 (Cq), 155.0 (Cq), 156.8 (Cq), 175.4 (Cq); nm/z (ES) (Found: MH^+, 332.1654. $C_{22}H_{22}NO_2$ requires MH, 332.1645).

ADME studies of inhibitors: Compounds that were highly effective in vitro (IC_{50} <1 μM) were tested for ADME profiling[S43-58] by Shanghai ChemPartner Ltd. Initial studies focused on aqueous kinetic solubility pH 7.4, microsomal metabolic stability (human and mouse) and Blood-Brain Barrier (BBB) permeability (performed with MDCK-MDR1 cells as described).

In Vitro Assays

Cytotoxicity Assay

Toxicity Analysis. Lack of toxicity for mammalian host cells was demonstrated first by visual inspection of monolayers following giemsa staining, in separate methods by incorporation of a mitochondrial cell death reagent called WST we used successfully for this purpose and in separate experiments.

Toxicity assays were conducted using WST-1 cell proliferation reagent (Roche). HFF were grown on a flat, clear-bottomed, black 96-well plate. Confluent HFF were treated with inhibitory compounds at concentrations equal to those being tested in challenge assays. Compounds were diluted in IMDM-C, and 20 μl were added to each designated well, with triplicates for each condition. A gradient of 2 fold-decreasing concentrations of DMSO in clear IMDM-C was used as a control. The plate was incubated for 72 hours at 37° C. 10 μl of WST-1 reagent (Roche) were added to each well and the cells were incubated for 30 to 60 minutes. Absorbance was read using a fluorometer at 420 nm. A higher degree of color change (and absorbance) indicated mitochondrial activity and cell viability.

In Vitro Cellular Assays for Effects on *T. gondii*

Vivo PMO: Vivo-PMO (Vivo porter linked to morpholinos) to knock down cytochrome b and an off-target PPMO (Vivo porter) were utilized at concentrations of 5 and 10 μM as previously described with both cultures of RH—YFP tachyzoites and EGS. Morpholino sequence for cytochrome b/c knockdown is 5' AGTGTTCTCGAAAC-CATGCTAACAC 3' (SEQ ID NO: 5), and for unrelated sequence, off target, is 5' CCTCTTACCTCAGTTACAAT-TTATA 3' (SEQ ID NO: 6).

Tetrahydroquinolone Compounds: Compounds synthesized at the University of Leeds were initially prepared in 10 mM Stock solutions made with 100% Dimethyl Sulfoxide (DMSO) [Sigma Aldrich], and working concentrations were made with IMDM-C (1×, [+]glutamine, [+] 25 mM HEPES, [−] Phenol red, 10% FBS) [Gibco, Denmark]).

Tachyzoite Assays:

Type 1 parasites. Human foreskin fibroblasts (HFF) were cultured on a flat, clear-bottomed, black 96-well plate to 90% to 100% confluence. IMDM (1×, [+] glutamine, [+] 25 mM HEPES, [+] Phenol red, 10% FBS [gibco, Denmark]) was removed from each well and replaced with IMDM-C (1×, [+] glutamine, [+] 25 mM HEPES, [−] Phenol red, 10% FBS)[Gibco, Denmark]). Type I RH parasites expressing Yellow Fluorescent Protein (RH—YFP) were lysed from host cells by double passage through a 27-gauge needle. Parasites were counted and diluted to 32,000/ml. in IMDM-C. Fibroblast cultures were infected with 3200 tachyzoites of the Type I RH—YFP strain and returned to incubator at 37° C. for 1-2 hours to allow for infection. Diluted solutions of the compounds were made using IMDM-C, and 20 μl were added to each designated well, with triplicates for each condition. Controls included pyrimethamine/sulfadiazine (current standard of treatment), DMSO only, fibroblast only, and an untreated YFP gradient with 2 fold dilutions of the parasite. Cells were incubated at 37° C. for 72 hours. The plates were read using a fluorimeter (Synergy H4 Hybrid Reader, BioTek) To ascertain the amount of yellow fluorescent protein, in relative fluorescence units (RFU), as a measure of parasite burden after treatment. Compounds were not considered effective or pursued for further analysis if there were no signs of inhibition at 1 μM. Data was collected using Gen5 software and analyzed with Excel.

Type II parasites. To test type II parasites, *T. gondii* ME49 and Prugneaud parasites expressing luciferase or GFP. We tested them in vitro and in vivo as we have described.

EGS strain Bradyzoite Assay. HFF cells were grown in IMDM (1×, [+] glutamine, [+] 25 mM HEPES, [+] Phenol red, 10% FBS, [Gibco, Denmark]) on removable, sterile glass disks in the bottom of a clear, flat-bottomed 24-well

plate. Cultures were infected with 3×10^4 parasites (EGS strain) per well, in 0.5 mL media and plate was returned to incubator at 37° C. overnight. The following day, the media was removed and clear IMDM and compounds were added to making various concentrations of the drug, to a total volume of 0.5 mL. Two wells were filled with media only, as a control. Plates were returned to the 37° C. incubator for 72 hours.

Efficacy was determined following fixation. Staining was used to determine the numbers of cysts in cultures without and with treatment with the test compounds. Cells were fixed using 4% paraformaldehyde and stained with Fluorescein-labeled *Dolichos biflorus* Agglutinin, DAPI, and anti-BAG1, and anti-SAG1. Disks were removed and mounted onto glass slides and visualized using microscopy (Nikon T17). Slides were also scanned using a CRi Pannoramic Scan Whole Slide Scanner and viewed using Panoramic Viewer Software.

When cysts that had dolichos in their cyst wall were eliminated or markedly reduced in size and number, a compound was considered efficacious against bradyzoites in cysts.

Statistical Analyses. Significance of differences were determined using Student's t-test. $P<0.05$ was considered significant. Every experiment was replicated at least twice. A Pearson test was used to confirm a correlation between increasing dose and increasing inhibition. An ANOVA and subsequent pair wise comparison with Dunnett correction was used to determine whether or not inhibition or toxicity at a given concentration was statistically significant. Stata/SE 12.1 was used for this analysis. This study was approved by the University of Chicago IRB, IBC, and IACUC.

In Vivo Analysis (Mice and Zebrafish):

Initial screening with tachyzoites using IVIS, fluorescence, and histopathology: Ability of compounds to abrogate tachyzoites multiplication was assessed using an in vivo imaging system (IVIS). To facilitate this we have *T. gondii* strains from each of the 3 major lineages expressing the luciferase gene. In these studies mice are injected intraperitoneally with tachyzoites and parasite proliferation followed up to 30 days post infection. Removal of brains at 30 days allows parasite quantitation by bioluminescence ex vivo using the IVIS. As an alternative method to improve screening efficiency and scalability it is possible for initial screening to use zebrafish with histopathology and visualization as shown in FIGS. **1**A-**1**D. Quantitation also was performed using QT PCR as described for mice or in translucent Casper zebrafish with parasites with fluors or luciferase to screen rapidly. Tachyzoites and bradyzoites in cysts were used for IP infection and compounds given intraperitoneally.

Type II parasites. To test type II parasites, we used *T. gondii* Me49 and Prugneaud parasites[S39].

Encephalitis: The ability of compounds to reduce cyst burden and prevent encephalitis induced by the Type II strain of *T. gondii* were tested. Encephalitis was assessed by histological analyses and parasite burdens evaluated by quantitation of cysts.

Oocyst induced disease: The oocyst challenge model is ideal for this study because oocysts can be diluted at one time and stored at 4° C. for 12 months without loss of infectivity titer. For treatment of chronic infection there were 5 to 10 mice per group treated 2 months after infection was established by compound in DMSO for parenteral administration administered once per day. Treatment was for 17 days.

Zebrafish Zebrafish were acclimatized to 37 degrees a degree a day and then infected with tachyzoites or cysts of RH YFP, Me49, Veg *T. gondii* as described. The use of RH YFP was performed for the first time herein in order to develop a rapidly scalable assay for drug development. This is the initial demonstration of cyst formation by 10 days in Zebrafish.

Tissue processing and histopathology: All organs including eyes and brains were fixed in 0.1M phosphate buffer (pH 7.4) containing 4% formaldehyde. Sections were cut from paraffin-embedded tissues and stained with Hematoxylin and Eosin (H&E) or immunoperoxidase stained. All sections were examined and assessed without knowledge of the group from which they originated[S39].

Testing of Cytochrome b Q_i Mutant Yeast

Target Validation with Mutant *S. cerevisiae* (Growth Inhibition):

Three *S. cerevisiae* strains were used: M221Q and M221F cytochrome b mutants and wild type. They share the same nuclear genetic background deriving from AD1-9 (kindly given by M. Ghislain, UCL, Belgium). AD1-9 harbors multiple deletions in the ABC transporter genes that render the strain more sensitive to drugs than standard yeast strainss[S65].

Cytochrome b mutant M221F was generated by mitochondrial transformation as described. M221Q was selected as suppressor from a respiratory deficient mutant. Analysis of revertants from respiratory deficient mutants within the center N of cytochrome b in *Saccharomyces cerevisiae*.

Protocol: Yeast strains were grown over 48 hours at 33° C. in liquid YPG medium [1% yeast extract, 2% (wt/vol) peptone, and 3% (vol/vol) glycerol). Cultures were diluted to an OD_{600} of 0.05 and grown for 2 hrs. Cultures were then combined with YPG containing 6% melted agar for a total volume of 15-20 mL and poured onto OmniTray single-well rectangular plates that measured 86 mm by 128 mm (ThermoScientific). Filter paper disks (7 mm diameter, 3 um thick) were placed onto the cooled agar plates. Compounds were dissolved in DMSO in diluted concentrations (1 mM, 500 μM, 100 μM, and 10 μM) and 10 microliters were applied to a disk. A single disk with DMSO on each plate was used as a control. Plates were incubated at 33° C. Images were obtained after 4 days using GelDoc XR Imaging System (BioRad) and Quantity One software. Drug effect was assessed by the presence and size of a zone of inhibition around the disks.

Testing of *P. falciparum*: D6 is a drug sensitive strain from Sierra Leone, C235 is a multi-drug resistant strain from Thailand, W2 is a chloroquine resistant strain from Thailand, and C2B has resistance to a variety of drugs including atovaquone.

Testing of *P. falciparum* Cytochrome b Qi and DHODH Mutants and Drug Combinations for *P. falciparum*

Parasite Strains and Culture Maintenance. We used the following parasite line from the MR4 repository of the American Type Culture Collection (ATCC): Dd2 (MRA-156). Mutant Dd2 parasites harboring a G33A or G33V substitution in cytochrome b were as reported. Dd2 parasites with a G131S mutation in cytochrome b and transgenic lines expressing a chromosomally integrated copy of the *S. cerevisiae* DHODH were utilized as previously described. Parasites were cultured by standard methods in RPMI media supplemented with 5% human O^+ serum and 0.25% AlbuMAX® II (Life Technologies 11021-045).

In Vitro Drug Sensitivity and EC_{50} Determinations

Drug susceptibility was measured using the SYBR Green method. Twelve point curves based on 2-fold dilutions of the test compound were carried out in triplicate each day and replicated on at least three different days. EC_{50} values were

calculated using a nonlinear regression curve fit in Prism 6.0 for Mac (GraphPad Software, Inc.).

Studies of compound, drug combinations in vitro. Isobologram experiments were performed in similar fashion utilizing the modified fixed ratio methodology. Briefly, MJM170 and either atovaquone or cycloguanil or BRD6323 were mixed at multiple fixed volumetric ratios (10:0, 8:2, 6:4, 4:6, 2:8, and 0:10) and then serially diluted in 12-point 2-fold dilutions and dispensed in triplicate to 384-well assay plates and replicated on three different days. EC_{50} values were calculated as above, and FICs were calculated for each drug combination as described[S76]. Synergy was defined as an FIC<1.0, additivity as FIC=1.0, and antagonism as FIC>1.0.

Molecular Modelling/Chemogenomics. X-ray structures of the cytochrome bc_1 complex are available from the Protein DataBank[S80]. An Homology model of the *T. gondii* cytochrome bc_1 complex was generated using the Phyre webserver. Molecular modelling and docking was performed on high performance Linux clusters at the University of Leeds, using specialist software: SPROUT[S82] & eHiTs[S83] (SymBioSis), Maestro & Glide[S84] (Schrodinger), AutoDock (Scripps Institute), ROCS/EON[S85] & VIDA[S86] (OpenEye) and the Marvin/JChem suites (ChemAxon).

X-ray crystallography: Cytochrome bc_1 was purified usig standard techniques. Crude bovine mitochondria were isolated from fresh cow heart and solubilised in DDM. The solution was clarified by ultracentrifugation at 200,000 g for 1 hour at 4° C. and the supernatant applied to a DEAE CL-6B sepharose column ca. 50 ml pre-equilibrated in 50 mM KPi (pH 7.5), 250 mM NaCl, 3 mM NaN_3, 0.1 g/L DDM, washed with two CV and eluted along a gradient from 250 mM to 500 mM NaCl. Cyt. bc_1 containing fractions were pooled and concentrated before loading on a Sepharose S300 column ca. 120 ml equilibrated with 20 mM KMOPS (pH 7.2), 100 mM NaCl, 0.5 mM EDTA, 0.1 g/L DDM at 0.5 ml/min. 10 mM MJM170 stock in DMSO was added to the eluted protein in a two-fold molar excess and allowed to incubate at 4° C. for 1 hour. Increasing amounts of PEG4000 were then added to precipitate cyt. bc_1 and separate remaining contaminants. The cyt. bc_1 was then resuspended before buffer exchange into a final buffer (25 mM KPi (pH 7.5), 3 mM NaN_3, 0.015% DDM) and concentrated to 40 mg/ml. 1.6% HECAMEG was added to the protein solution prior tp crystals growing by the hanging drop vapour diffusion method against a reservoir of 50 mM KPi (pH 6.8), 100 mM NaCl, 3 mM NaN_3, 9% PEG4000, 0.16% HECMAEG. Crystals were flash frozen in 23% glycerol in reservoir solution as a cryoprotectant. Multiple wedges of data were collected at 100K from different points on the same crystal at 124 Diamond Light Source using 0.9686 Å X-rays with a Pilatus3 6M detector.

Datasets were processed in iMosfim and combined using Blend to produce a complete merged dataset. Refinement was carried out with Refmac using Prosmart to generate secondary structure restraints to assist in the low-resolution refinement. The ligand MJM170 was produced using JLigand[S92] and modelled in the Q_i site of cyt. bc_1 using Coot. Cycles of alternating Refmac5 and manual modelling resulted in a completed model. Data collection and refinement statistics are summarised In table 1A. For 3715 residues 95.2% are Ramachandran favored, 4.6% allowed and 0.3% outliers.

Interpretation of Data and Statistical Analyses

(1) Sample size and number of experiments. There were 3 replicate samples per group for in vitro experiments. All experiments were performed with sufficient sample sizes to have an 80% power to detect differences at the 5% level of significance.

(2) Statistics. Groups included untreated or mock treated controls. Results were compared using students T test, Chi square analysis or Fisher's exact test as appropriate for the data set. When there were more than two groups. pairwise comparisons were made only when F-test for the ANOVA was significant at the 5% levels using protected least significant difference (LSD) test approach.

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Example 2. Potent anti-apicomplexan tetrahydroquinolone

Summary: Apicomplexan infections cause substantial morbidity and mortality. Herein, we created a next generation tetrahydroquinolone that we found to be an anti-apicomplexan, mature, lead compound. We utilized sphere-like 3D space and predicted flexibility conferred by eliminating double bonds in this lead compound. This was to optimize ADMET and create a compound, JAG21, that is potent against *Toxoplasma gondii* tachyzoites (IC 90<125 nM) and bradyzoites (IC 90 500 nM), and drug resistant *Plasmodium falciparum* in vitro (IC 90<50 nM), not toxic to human HepG cells (>17 μM). Further, we demonstrate metabolic stability with assays for human and mouse liver microsomal activity and logs improved aqueous solubility at pH 7.4. This compound displays a balanced set of physicochemical and pharmacologic properties, including clean hERG, CYP profile, and a long (days in humans), predicted half-life and predicted ability to cross blood brain barrier. This allowed progression towards in vivo studies. In vivo *Toxoplasma* tachyzoites were cleared from mice at a dose of 5 mg/kg/day (IP). JAG21 acted in conjunction with tafenoquine (3 mg/kg single dose) to protect against a G0 arrested parasite that could persist in interferon ϒ knockout mice similar to the effect of tafenoquine for malaria hypnozoites. There was cure with oral dosing, 0.625 mg/kg, 3 oral doses of JAG21, and cure with a single dose of 2.5 mg/kg, of *P. berghei* sporozoite, blood and liver stages in mice. There was no parasitemia and 100% survival at 30 days. This mature lead compound has improved solubility and diminished toxicity relative to other cytochrome b Qi inhibitors, without formulation as a pro-drug. Selectivity for apicomplexan enzyme relative to mammalian enzymes was demonstrated with co-crystallography, binding and enzyme assays. This compound has real promise as a mature, lead compound.

Malaria results in death of one child every eleven seconds and 1 million children a year, with drug resistance eliminating usefulness of successive generations of new medicines each decade. The related apicomplexan parasite, *Toxoplasma gondii*, is the most frequent parasitic infection of humans, in the world. It is the second most frequent, single cause of food born associated death in the United States; It is the most frequent infectious cause of destruction of the back of the human eye; It is a cause of death and illness from recrudescent disease from its latent form in those who are immune compromised or immunologically immature; It has been estimated that in a ten year period, there are 1.9 million new cases of this congenital infection globally, causing 12 million disability adjusted life years from damage to the fetal brain and eye. This is a neglected, rarely diagnosed, and thus often untreated or mistreated disease. There are approximately 2 billion people throughout the world who have this parasite in their brain lifelong. No medicine eliminates this chronic encysted form of the parasite which causes epilepsy and may contribute to neurodegenerative disease. Certainly, new and improved medicines are greatly needed for both these diseases, These two apicomplexan parasites, *Plasmodia* and *Toxopalsma*, often share molecular targets inhibited by the same inhibitory compounds.

Herein we identify a mature lead compound that is highly efficacious against *T. gondii* tachyzoites and bradyzoites in vitro, tachyzoites in vivo, likely to be active against cysts in vivo with experiments ongoing, all drug resistant forms of *Plasmodium falciparum, Plasmodium berghei* in mouse model in single or three doses at low amounts against the sporozoite, blood and liver stages of plasmodium when administered orally at 2.5 mg/kg and at 1.25 mg kg for 100% of mice with three doses. It was found to add to protection in conjunction with tafenoquine in immune compromised mice infected with a G0/tachyzoite form of *T. gondii* which resembles the malaria hypnozoite when treated with tafeno-

quine in conjunction with anti-blood stage parasite compounds. The data which follow present the creation and characterization of this broad spectrum anti-apicomplexan lead compound.

Materials and Methods

Toxoplasma gondii

Tachyzoites of the RH—YFP strain were passaged in human foreskin fibroblasts (HFF cells)(15). Bradyzoite assays use the EGS strain, isolated from a human with congenital toxoplasmosis (16,17). These parasites are also passaged in human foreskin fibroblasts. RPS13 delta was prepared and utilized as described (Hutson, McLeod et al 2010)

Tetrahydroquinolone (THQ) Compounds

The THQ compounds were synthesized at the University of Leeds as described in Example 3. 10 mM stock solutions were made with 100% Dimethyl Sulfoxide (DMSO) [Sigma Aldrich] and working concentrations were made with IMDM-C (1×, [+] glutamine, [+] 25 mM HEPES, [−] Phenol red, 10% FBS)[Gibco, Denmark]). Compounds are shown herein.

In Vitro Challenge Assay for *Toxoplasma* Tachyzoites

Protocol adapted from Fomovska, et. al. (18,19). Human foreskin fibroblasts (HFF) were cultured on a flat, clear-bottomed, black 96-well plate to 90% to 100% confluence. IMDM (1×, [+] glutamine, [+] 25 mM HEPES, [+] Phenol red, 10% FBS [gibco, Denmark]) was removed from each well and replaced with IMDM-C (1×, [+] glutamine, [+] 25 mM HEPES, [−] Phenol red, 10% FBS)[gibco, Denmark]). Type I RH parasites expressing Yellow Fluorescent Protein (RH—YFP) were lysed from host cells by double passage through a 27-gauge needle. Parasites were counted and diluted to 32,000/mL in IMDM-C. Fibroblast cultures were infected with 3200 tachyzoites of the Type I RH strain expressing Yellow Fluorescent Protein (RH—YFP) and returned to incubator at 37° C. for 1-2 hours to allow for infection (15). Various concentrations of the compounds were made using IMDM-C, and 20 μl were added to each designated well, with triplicates for each condition. Controls included pyrimethamine/sulfadiazine (current standard of treatment), 0.1% DMSO only, fibroblast only, and an untreated YFP gradient with 2 fold dilutions of the parasite. Cells were incubated at 37° C. for 72 hours. Plates were read using a fluorimeter (Synergy H4 Hybrid Reader, BioTek) to ascertain the amount of yellow fluorescent protein, in relative fluorescence units (RFU), as a measure of parasite burden after treatment. Data was collected using Gen5 software. IC_{50} was calculated by graphical analysis in Excel.

An initial screening assay of 10 μM, 1 μM, 100 nM, and 10 nM was performed. Compounds were not considered effective or pursued for further analysis if there were no signs of inhibition of tachyzoites at 1 μM. If compounds did appear to be effective at 1 μM, another experiment was conducted to assess effect at 1 μM, 500 nM, 250 nM, 125 nM, 62.5 nM, and 31.25 nM.

Cytotoxicity Assay

Toxicity assays were conducted using WST-1 cell proliferation reagent (Roche) as described in Fomovska, et. al. (18,19). HFF were grown on a flat, clear-bottomed, black 96-well plate. Confluent HFF were treated with inhibitory compounds at concentrations of 10 μM and 50 μM. Compounds were diluted in IMDM-C, and 20 μl were added to each designated well, with triplicates for each condition. A gradient of 2 fold-decreasing concentrations of DMSO from 10% to 0% in clear IMDM-C was used as a control. The plate was incubated for 72 hours at 37° C. 10 μl of WST-1 reagent (Roche) were added to each well and the cells were incubated for 30 to 60 minutes. Absorbance was read using a fluorimeter at 420 nm. A higher degree of color change (and absorbance) indicated mitochondrial activity and cell viability.

In Vitro Challenge Assay for Bradyzoites

HFF cells were grown in IMDM (1×, [+] glutamine, [+] 25 mM HEPES, [+] Phenol red, 10% FBS, [gibco, Denmark]) on removable, sterile glass disks in the bottom of a clear, flat-bottomed 24-well plate. Cultures were infected with 3×10^4 parasites (EGS strain) per well, in 0.5 mL media and plate was returned to incubator at 37° C. overnight. The following day, the media was removed and clear IMDM and compounds were added to making various concentrations of the drug, to a total volume of 0.5 mL. 2 wells were filled with media only, as a control. Plates were returned to the 37° C. incubator for 72 hours, and checked once every 24 hours. If tachyzoites were visible in the control before 72 hours, the cells were fixed and stained.

Cells were fixed using 4% paraformaldehyde and stained with Fluorescein-labeled *Dolichos biflorus* Agglutinin, DAPI, and BAG1. Disks were removed and mounted onto glass slides and visualized using microscopy (Nikon T17). Slides were scanned using a CRi Pannoramic Scan Whole Slide Scanner and viewed using Panoramic Viewer Software. Effects of the compounds were quantified by counting cysts in the controls and treated cells. Cysts and persisting organisms were counted in a representative field of view and then multiplied by a factor determined by the total area of the disk in order to estimate the number of cysts and organisms in each condition.

Assessment of Compound Degradation and Microbicidal Effect on *Toxoplasma*

HFF were cultured in a 96-well plate and infected with RH—YFP as described above on Day 0, 20 μL of compound was added to 9 wells for each compound and concentration (3 conditions, 3 wells per condition). In condition I, media was removed and replaced with fresh media on Day 3. In condition II, media was removed and replaced with fresh media and more compound on Day 3. In condition III, media was not replaced on Day 3, nor was the compound refreshed. On Day 6, media was removed and replaced with clean media in all wells. On Day 3, 6, and 9 plate was read in the fluorimeter and analyzed graphically in Prism (GraphPad Software).

Toxoplasma In Vivo

IVIS. Mice were infected intraperitoneally with 20×10^3 *Toxoplasma gondii* (Pru strain expressing luciferase) tachyzoites. Treatment commenced 2 hours later with JAG21 (5 mg/kg) which was dissolved in DMSO and administered intraperitoneally in a total volume of 0.05 ml. Mice were imaged every second day starting on day 4 post infection using a IVIS Spectrum (Caliper Life Sciences) for a 1 minute exposures, with medium binning, 20 minutes post injection with 150 mg/kg of D-luciferin potassium salt solution.

Brain cysts: Mice were infected intraperitoneally with 20×10^3. Treatment commenced 2 hours later with JAG21 (5 mg/kg) which was dissolved in DMSO and administered intraperitoneally in a total volume of 0.05 ml. After 30 days, treatment with JAG21 was begun each day for 14 days intraperitoneally. In experiments when tafenoquine was administered alone or with JAG21 in some groups 3 mg/kg tafenoquine was administered once on day −1. Cysts in brain were quantitated after concluding treatment

RPS13 Δ. This G0 arrested parasite persists in tissue culture for prolonged times in the absence of tetracycline. The design of this experiment is shown in FIG. **13**. The

parasite (x) was used to infect interferon gamma knockout mice. For the first y days no tetracycline was administered. After that time tetracycline was administered. Mice were observed and at the time they appeared ill or at the termination of the experiment they were euthanized and tissues fixed in formalin and stained with hematoxylin and eosin or immunoperoxidase stained and parasite burden was assessed.

Malaria Assays

Methods for enzyme assays[21-3]: Professor Giancarlo A. Biagini, Dr Richard S. Priestley, Department of Parasitology, Liverpool School of Tropical Medicine, Pembroke Place, Liverpool, L3 5QA, UK.

Materials

Plasmodium falciparum: 3D7 strain was obtained from the Liverpool School of Tropical Medicine. Protease cocktail inhibitor was obtained from Roche. Bradford protein assay dye reagent was obtained from Bio-Rad. All other reagents were obtained from Sigma-Aldrich. Decylubiquinol was produced as per Fisher et al. (Fisher et al. 2004)[21]. In brief, 25 mg of decylubiquinone were dissolved in 400 µl of nitrogen-saturated hexane. An equal volume of aqueous 1 M sodium dithionite was added, and the mixture vortexed until colorless. The organic phase containing the decylubiquinol was collected, the solvent was evaporated under N_2 and the decylubiquinol finally dissolved in 100 µl of 96% ethanol (acidified with 10 mM HCl). Concentrations of decylubiquinol was determined spectrophotometrically on a Cary 300 Bio UV/visible spectrophotometer (Varian, UK) from absolute spectra, using 8288-320=8.1 mM-1 cm^{-1}. Decylubiquinol was stored at −80° C. and used within two weeks.

Plasmodium falciparum: Culture and Extract Preparation

Plasmodium falciparum: strain 3D7 blood-stage cultures were maintained by the method of Trager and Jensen (Trager & Jensen 2005)[23]. Cultures contained a 2% suspension of O+ human erythrocytes in RPMI 1640 medium containing L-glutamine and sodium carbonate, and supplemented with 10% pooled human AB+ serum, 25 mM HEPES (pH 7.4) and 20 µM gentamicin sulphate. Cultures were grown under a gaseous headspace of 4% 02 and 3% CO_2 in N_2 at 37° C. Cultures were grown to a parasitaemia of 5% before use.

The protocol for the preparation of parasite extract was adapted from Fisher et al. (Fisher et al. 2009)[22]. Free parasites were prepared from infected erythrocytes pooled from five T75 flasks, by adding 5 volumes of 0.15% (w/v) saponin in phosphate-buffered saline (137 mM NaCl. 2.7 mM KCl, 1.76 mM K_2HPO_4. 8.0 mM Na_2HPO_4. 5.5 mM D-glucose, pH 7.4) for 5 min, followed by three washes by centrifugation in RPMI containing HEPES (25 mM), and a final resuspension in potassium phosphate buffer (50 mM K_2HPO_4, 50 mM KH_2PO_4, 2 mM EDTA, pH7.4) containing a protease inhibitor cocktail (Complete Mini; Roche). Parasite extract was then prepared by disruption with a sonicating probe for 5 s, followed by a 1 min rest period on ice to prevent the sample overheating. This process was performed three times. The parasite extract was used immediately. The protein concentration of the parasite extract was determined by Bradford protein assay (Bio-Rad).

$Pfbc_1$ Native Assay

Plasmodium falciparum: bc_1 complex cytochrome c reductase ($Pfbc_1$) activity was measured by monitoring cytochrome c reduction at 550 versus 542 nm using a Cary 300 Bio UV-Visible Spectrophotometer (Varian, UK), using a protocol adapted from Fisher et al. (Fisher et al. 2009)[21-23]. The assay was performed in potassium phosphate buffer in a quartz cuvette and in a final volume of 700 µL. Potassium cyanide (10 µM), oxidised cytochrome c (30 µM), parasite extract (100 µg protein) and compound/DMSO were added sequentially to the cuvette, with mixing between each addition. Test compounds were added to a final concentration of 1 µM. DMSO (0.1% v/v) and atovaquone (1 µM), a known malarial cytochrome bc_1 complex inhibitor, were used as negative and positive controls respectively. The reaction was initiated by the addition of 50 µM decylubiquinol and allowed to proceed for 3 min.

Data Analysis

Malaria

In vitro studies: D6 is a drug sensitive strain from Sierra Leone, C235 is a multi-drug resistant strain from Thailand, W2 is a chloroquine resistant strain from Thailand, and C2B has resistance to a variety of drugs including atovaquone. These assays were performed as described.

Compound Activity Against *Plasmodium falciparum:*

Compound activity against *P. falciparum*, a causative agent of malaria, was tested using the Malaria SYBR Green I—Based Fluorescence (MSF) Assay. This; microtiter plate drug sensitivity assay uses the presence of malarial DNA as a measure of parasitic proliferation in the presence of antimalarial drugs or experimental compounds based on modifications of previously described methods by Plouffe et al (20) and Johnson et al. As the intercalation of SYBR Green I dye and its resulting fluorescence is relative to parasite growth, a test compound that inhibits the growth of the parasite will result in a lower fluorescence.

Selected compounds were examined for activity against four strains of *P. falciparum*: D6 (CDC/Sierra Leone), a drug-sensitive strain readily killed by chloroquine, TM91-C235, a multi-drug resistant strain resistant to chloroquine, W2, a chloroquine resistant strain from Thailand, and C2B has resistance to a variety of drugs including atovaquone.

P. berghei Model Sporozoite, Blood Stage, and Liver Stage Model.

P. berghei sporozoites. The methods that follow are taken directly from[24,25]: From laboratory-reared female *Anopheles stephensi*, isolation, inoculation and viability check *Plasmodium berghei* sporozoites (luciferase expressing) were obtained and maintained at 18° C. for 17 to 22 days after feeding on malaria-infected Swiss CD-1/ICR mice. From malaria-infected mosquitoes, salivary glands were extracted and sporozoites obtained. Briefly, mosquitoes were separated into head/thorax and abdomen. Thoraxes and heads were triturated with a mortar and pestle and suspended in medium RPMI 1640 containing 1% C57BL/6 mouse serum (Rockland Co, Gilbertsville, PA, USA). 50-80 heads with glands total were placed into a 0.5 ml Osaki tube on top of glass wool with enough dissection media to cover the heads. Until all mosquitoes had been dissected, the Osaki tube was kept on ice. Sporozoites that were isolated from the same batch of mosquitoes were inoculated into C57BL/6, 2D knock-out and 2D knock-out/2D6 knock-in C57BL/6 mice on the same day to control for biological variability in sporozoite preparations. On day 0, each mouse was inoculated intravenously in the tail vein with approximately 10,000 sporozoites suspended in 0.1 ml volume. They were stained with a vital dye containing fluorescein diacetate (50 mg/ml in acetone) and ethidium bromide (20 µg/ml in phosphate buffered saline; Sigma Chemical Co, St. Louis, MO, USA) and counted in a haemocytometer to ensure that inoculated sporozoites were viable following the isolation procedure. Viability of the sporozoites ranged from 90 to 100%.

Animals

The mice used in these experiments were Swiss Webster females. The animals were acclimated for seven days (quarantine) on arrival. The animals were housed in a cage maintained in a room with 34-68% relative humidity, a temperature range of 64-79° F., and a 12-hr light/dark cycles. Water and food were provided during quarantine and throughout the study. The mice were fed a standard rodent maintenance diet. All animal studies were performed under protocols that are IACUC-approved. All animal care, handling, and use was performed in accordance with the current Guide for the Care and Use of Laboratory Animals (1996).

Test Compounds and Administration

At the time of preparation of the suspension solution, compounds tested in these experiments were dosed based on the body weight. The suspension solution of oral agents, using homogenizer (PRO Scientific Inc, Monroe, CT, USA) with 10 mm open-slotted generator to homogenize drug powder mixture at 20,000-22,000 rpm for 5 min in ice bath, were prepared in 0.5% (w/v) hydroxyethyl cellulose and 0.2% (0.5% HECT, v/v) Tween-80 in distilled water.

A three consecutive day-treatment regimen (−1, 0, 1 day) or a once-a-day, one dose on day 0 was used in assessments. Drug suspensions were transferred to a 20-ml bottle, drawn into a 1-ml syringe, and delivered to the designated recipient via intragastric feeder (18 gauge).

1 hour after intravenous administration of 10,000 *P. berghei* sporozoites, single dose causal prophylaxis in 5 C57BL/6 albino mice at 2.5 mpk dosed on day 0. In 5 C57BL/6 albino mice, 3 dose causal prophylaxis treatment at 0.6 mpk dosed on days −1, 0, and +1.

In Vivo Imaging System Spectrum

All of the in vivo imaging system (IVIS) methods utilized have been described previously [6]. Briefly TQ and NPC-1161B were administered orally on days −1, 0 and 1 with respect to sporozoite inoculation. All inoculated mice were tested using the Xenogen IVIS-200 Spectrum (Caliper Life Sciences, Hopkinton, MA, USA) IVIS instrument at 24, 48 and 72 hr post-sporozoite infection. Additionally, using a flow cytometry system (FC500 MPL, Beckman Coulter, Miami, FL, USA), blood-stage infections were measured. For the IVIS calibration in each test, positive and negative controls were used. D-Luciferin potassium salt, (Xenogen, California and Goldbio, St Louis, MO, USA), the luciferase substrate, was inoculated intraperitoneally into mice at a concentration of 200 mg/kg 15 min before luminescence analysis. Three min post-luciferin administration the mice were anesthetized using isoflurane. The mice, in the IVIS on the 37° C. platform, were then positioned ventral side up. Through nose cone delivery, the mice continued to receive isoflurane. The exposure time of the camera was 5 min for the 24, 48 and 72 hr time points with f-stop=1 and large binning setting. Using Living Image® 3.0 software, photons emitted from specific regions were quantified.

Parasitemia was measured after days of IVIS imaging. During a total of 30 days, mice were observed and parasitemia level determined using FACs analysis. (Pybus et al. Malaria Journal 2013, 12:212; Marcsisin et al. Malaria Journal 2014, 13:2).

Bovine Cytochrome bc1 Purification Protocol

Preparation of crude mitochondria: Whole bovine heart was collected directly from slaughter and transported on ice to the cold room. All work was carried out at 4° C. Fat and other tissues were removed leaving only lean muscle that was then cut into small cubes. The cubes were then transferred to a waring blender and homogenisation buffer (250 mM sucrose; 20 mM K_2HPO_4; 2 mM succinic acid; 0.5 mM EDTA) was added at a ration of 2.6 L buffer per 1 L of muscle tissue. The solution was then homogenised. The resulting homogenate was adjusted to pH 7.8 using 2 M Tris and PMSF was added to a concentration of 0.1 mM. The homogenate was then centrifuged in a Sorvall GS-3 rotor at 3000 rpm for 20 mins. The resulting supernatant was then transferred to a Sorvall GSA rotor and centrifuged at 12,000 rpm for 20 mins. The pellet was then re-suspended and washed in buffer 1 (50 mM KPi (pH 7.5); 0.1 mM PMSF) before centrifugation under the same conditions again. The pellet was collected and frozen at −80° C. for use later.

Solubilisation of membrane proteins: The frozen mitochondria were thawed and re-suspended in buffer 2 (50 mM KPi (pH 7.5); 150 mM NaCl; 3 mM NaN_3; 0.1 mM PMSF) and a sample taken for a BCA assay. The remaining sample was centrifuged at 42,000 rpm in a Beckman Ti70 rotor for 60 mins. The pellet was re-suspended in the same wash buffer to a volume of 70 ml with the addition 0.1 mg DDM per 1 mg of protein and centrifuged at 42,000 rpm in a Beckman Ti70 rotor for 60 mins. The pellet was then re-suspended in the same wash buffer to a final volume of 215 ml with the addition of 0.9 mg DDM per 1 mg of protein and centrifuged for a final time at 42,000 rpm in a Beckman Ti70 rotor for 60 mins. The supernatant was collected.

Purification of cytochrome bc_1: Whilst being purified, the presence of protein was determined using 280 nm absorbance and the presence of haem was determined using 415 nm soret band peak and 462 nm absorbance. The solubilised protein solution was first applied to a DEAE-Sepharose CL-6B column (ca. 50 ml) pre-equilibrated in buffer A (50 mM KPi (pH 7.5); 150 mM NaCl; 0.03% DDM; 3 mM NaN_3) washed with 2 CV buffer A and eluted along a gradient with buffer B (50 mM KPi (pH 7.5); 350 mM NaCl; 0.03% DDM; 3 mM NaN_3). The collected protein was pooled and diluted twofold with buffer C (50 mM KPi (pH 7.5); 0.03% DDM; 3 mM NaN_3) before application to a hydroxyapatite column (ca. 15 ml) pre-equilibrated with buffer C. The column was washed with 10 CV of buffer C before elution along a gradient with Buffer C* (1000 mM KPi (pH 7.5); 0.03% DDM; 3 mM NaN_3). Fractions containing cytochrome bc_1, as identified by 415 nm absorbance, were then collected, pooled and concentrated to 1.5 ml using an Amicon Ultra-15 (Amicon, MWCO 100,000). The sample was then applied to a Sephacryl-S300 column (ca. 120 ml) pre-equilibrated in buffer D (25 mM KPi (pH 7.5); 100 mM NaCl; 0.015% DDM; 3 mM NaN_3) and ran at a flow rate of 0.5 ml/min. Purified cytochrome bc_1 fractions were then collected and concentrated to 30 mg/m.

Bovine Enzyme crystallography: Compounds designed using structure-based analyses of cytochrome b co-crystalized with JAG21 as described un Example 1[26]. This was done to optimize medicine-like properties using structure activity principles and analyses. Compounds synthesized as

above were used in these assays as follows: 0.10 mM stock solutions were made with 100% Dimethyl Sulfoxide (DMSO) [Sigma Aldrich] and working concentrations were made with IMDM-C (1×, [+] glutamine, [+] 25 mM HEPES, [−] Phenol red, 10% FBS)[gibco, Denmark]).

Statistical Analysis: A Pearson test was used to confirm a correlation between increasing dose and increasing inhibition. An ANOVA and subsequent pairwise comparison with Dunnett correction was used to determine whether or not inhibition or toxicity at a given concentration was statistically significant. Stata/SE 12.1 was used for this analysis.

Results

Tetrahydroquinolone Compounds:

In Vitro Challenge Assay for Tachyzoites: Seven compounds (Table 1) were tested and each compound was tested at least twice. JAG021 and JAG050 demonstrated effect below 1 μM, and were tested at lower concentrations. A representative graph of this data is shown in FIG. **8**. JAG050 and JAG021 were identified as lead compounds because the IC_{50} values were 55 and 188 nM respectively. Correlation between concentration of compound and inhibition of parasite growth and activity (as measured by fluorescence) was observed for all compounds except JAG046.

Cytotoxicity Assay using HFF and WST-1 and IC50 with HEP G cells: Because *T. gondii* grows inside cells, if a compound was toxic to host Human Foreskin Fibroblast Cells (HFF), then it would make the compound appear to be spuriously effective; in actuality only toxicity for the host cell would be measured. Cytotoxicity to human foreskin fibroblasts was therefore assessed for all compounds at 10 μM and 50 μM. Results of this experiment are in FIG. **9** and Table 3. A two-way ANOVA and subsequent pairwise comparison found none of the differences in absorbance, compared to the controls, to be statistically significant ($p>0.05$). This suggests that these compounds are not toxic at 10 μM or 50 μM and that toxicity to cells is attributed to DMSO in the solution, not the compound. IC50 with HEP G cells was performed as described and toxicity was: HEP G2 IC50 17.70 microM ($r^2=0.97$) JAG 21; JAG 50 7.1 microM $r^2=0.98$.

TABLE 3

Cytotoxicity to human foreskin fibroblasts was therefore assessed for all compounds at 10 μM and 50 μM. Graph is representative of replicate experiment.

Observation	Control	JAG050 10 μM	JAG050 1 μM	JAG021 10 μM	JAG021 1 μM
		a			
True Cysts	4.67 ± 3.06 [2-8]	1 ± 0.82 [0-2]	0.25 ± 0.5 [0-1]	0.25 ± 0.5 [0-1]	0.5 ± 0.6 [0-1]
Pseudocysts	40.3 ± 11.4 [31-53]	20.5 ± 2.9 [17-24]	23.25 ± 10.31 [14-38]	25.5 ± 5.1 [19-30]	29 ± 6 [21-34]
Small organisms	1600 ± 436 [1100-1900]	31 ± 16 [8-43]	58 ± 24 [27-85]	73.25 ± 30.9 [30-101]	90.5 ± 33.5 [63-137]
		b			
True Cysts	452	88	29	22	54
Pseudocysts	3884	1921	2269	2638	2955
Small organisms	16404	3018	5086	7309	9734

In Vitro Challenge Assay for Bradyzoites

Lead compounds JAG050 and JAG021 were tested against EGS because of their effects on tachyzoites (RH—YFP). Under immunofluorescence microscopy, the following forms were observed: "true cysts" with a dolichos-staining wall. "pseudocysts" or tight clusters of parasites, and small organisms. If there were fewer than four parasites visible in a cluster, the organisms were counted individually (as "small organisms"). A statistically significant reduction in the number of true cysts and small organisms was observed at 1 μM and 10 μM for both compounds ($p<0.05$, $p<0.005$, FIGS. **10**A-**10**C).

ADME properties of THQs. In vitro ADME analyse of the THQ compounds were outsourced to ChemPartner Shanghai Ltd. ELQ-271 was tested as a comparison. THQs which were potent inhibitors of *T. gondii* tachyzoites were assessed for their kinetic solubility, metabolic stability in human and mouse liver microsomes, and their ability to permeate across MDCK-MDK1 cell membranes (in vitro measure of blood-brain barrier (BBB) permeability). Solubility, half-life and BBB permeability/efflux results are shown in Table 4. The kinetic solubility (PBS, pH 7.4) of compounds JAG021 and JAG050, 7 and 16 μM respectively, were higher than MJM170 (2 μM) and ELQ-271 (0.2 μM). JAG021 was the most metabolically stable compound in human liver microsomes (>99% remaining after 45 mins) compared with other THQs and ELQ-271, although it displayed a much shorter half-life of 101 mins in mouse liver microsomes. All THQs tested in the MDK1 (MJM170, JAG021 and JAG050) MDCK-MDK1 system exhibit high permeability (P_{app} >10× 10^6 cm/s) and low efflux (efflux ratio <1.5).

TABLE 4

Chart compares properties of solubility and half-life of JAG050 and JAG021 to parent compounds ELQ 271 and MJM170.

Compound	Solubility (pH 7.4)*	Human liver microsomes#	Mouse liver microsomes#
ELQ271	0.15 μM	171.93 min	448.13 min
MJM170	1.97 μM	146.33 min	20.97 min

TABLE 4-continued

Chart compares properties of solubility and half-life of JAG050 and JAG021 to parent compounds ELQ 271 and MJM170.

Compound	Solubility (pH 7.4)*	Human liver microsomes#	Mouse liver microsomes#
JAG021	7.07 μM	∞	101.09 min
JAG050	16.41 μM	99.04 min	68.55 min

The test system was 100 mM Phosphate Buffer (pH 7.4).:
<10 μM is low solubility,
10-80 μM is moderate solubility and
>80 μM is high solubility.
A $T_{1/2}$
< 30 minutes indicates susceptibility to metabolism,
between 30 and 120 minutes indicates moderate metabolism and
>120 minutes indicates stability in the liver.

Enzyme assays: Enzyme reduction of cytochrome c by the parasite extract is mediated by *P. falciparum* bc_1 complex cytochrome c reductase ($Pfbc_1$). All three compounds (1 μM) significantly inhibited the reduction of cytochrome c by the parasite extract, (JAG021=86.4±3.2; JAG099=81.3±6.0; MJM170=69.7±11.3% atovaquone response). This clearly demonstrates the compounds are inhibitors of $Pfbc_1$. Additional data demonstrating effect on bovine and *Plasmodium falciparum* enzyme are shown in Table 5. There is selectivity for the malaria enzyme.

TABLE 5

Inhibition of $Pfbc_1$ by compounds.

Compound (1 μM)	Inhibition of cytochrome c reduction (% atovaquone response)
JAG021	86.4 ± 3.2
JAG099	81.3 ± 6.0
MJM170	69.7 ± 11.3

Data shown are mean ± s.e.m. of 4 independent experiments performed in triplicate.

Binding assays and co crystallography. JAG021 has lower binding affinity to bovine cytochrome be in comparison with previous compounds that we have tested. JAG 21 'inhibits' Cytbc1 but not fully, indicating that it will be less toxic for bovine/human cyt bc (FIG. **11**)

Assessment of Compound Degradation and Microbicidal Effect on *Toxoplasma gondii* Compounds JAG050 and JAG021 were observed for degradation and microbicidal effect. Neither compound was found to be microbicidal; when media was replaced with clean media, the parasites appeared to resume activity and replication. In comparing the 6-day exposure with no addition of compound to the 6-day exposure with the addition of compound, it did not appear that the compound was being degraded over time. In condition II, in which the compound is refreshed, there appears to be a rise in fluorescence on day 6 in the 1 μM treatment group for both compounds. However, these differences were not found to be statistically significant ($p>0.05$).

Effective of JAG21 on *Toxoplasma gondii*. JAG21 at 5 mg/kg eliminates *T. gondii* tachyzoites seen in luminescence studies (FIGS. **12A-12D**).

JAG21 against G0 arrested and normal (no tet repressor) *Toxoplasma* RPS13 delta in Interferon gamma knockout mice plus and minus tetracycline. Our data show that the combination of JAG21 and tafenoquine treatment is superior to either alone against RPS13Δ minus tetracycline (FIG. **13**). The data indicates that this appears to be a dormant parasite that is less susceptible to JAG21 than either the slowly growing EGS bradyzoites or the rapidly proliferating tachyzoites.

Malaria:

In vitro. Results are shown in Table 6. JAG 21 is a 40-65 nM inhibitor of *Plasmodium falciparum* including effect against all drug resistant strains. The effects of the other compounds are also shown in this table and are in the range of 50-200 nM.

TABLE 6

Inhibition of *P falciparum* in vitro including drug resistant isolates

Compound ID	SYBR Green D6 IC50 (uM)	SYBR D6 R^2	SYBR Green C235 IC50 (uM)	SYBR TM91C235 R^2	SYBR Green W2 IC50 (uM)	SYBR W2 R^2	SYBR Green C2B IC50 (uM)	SYBR C2B R^2
JAG006	0.29	0.90	0.88	0.92	2.46	0.92	1.66	0.94
JAG021	0.01435	0.9572	0.06164	0.9706	0.05518	0.9727	0.04042	0.9847
JAG050	0.04664	0.9138	0.06913	0.9562	0.03136	0.9693	0.03635	0.9427
JAG047	3.746	0.9738	12.56	0.9218	9.072	0.9358	7.781	0.9575
JAG039	9.595	0.9532	>20	N/A	>20	N/A	>20	N/A
JAG046	6.716	0.9844	>20	N/A	>20	N/A	>20	N/A
RG38	2.84	0.8936	13.66	0.8338	9.245	0.7954	>20	N/A

In vivo. Single dose causal prophylaxis in 5 C57BL/6 albino mice at 2.5 mpk dosed on day 0, 1 hour after intravenous administration of 10,000 *P. berghei* sporozoites. 3 dose causal prophylaxis treatment in 5 C57BL/6 albino mice at 0.6 mpk dosed on days −1, 0, and +1. A representative figure for higher dose (5 mg/kg) is shown, but all experiments with the amounts mentioned above had efficacy measured as cure measured as survival, luminesence and parasitemia quantitated by flow cytometry are similar to these. (FIGS. **12A-12D**)

Discussion

JAG050 and JAG021 were identified as lead compounds, demonstrating potent inhibition of tachyzoites and bradyzoites and no toxicity to human foreskin fibroblasts in our in vitro model. While compounds inhibited parasite replication and activity, there did not appear to be a microbicidal effect.

Toxoplasmosis is highly prevalent and the impact of this disease can be devastatingly severe. Current treatments have toxic side effects and are not curative. JAG050 and JAG021 are lead compounds in the search for a new curative medicine because they demonstrate effect on both life stages and were not toxic to the human cells in our in vitro model.

Experiments testing the compounds against the EGS strain had some surprising findings. While true cysts in vitro appeared to be completely eliminated by treatment, or their number significantly reduced, parasites did persist in tight, clustered, cyst-like structures, or pseudo-cysts, and small punctate life forms that resemble tachyzoites. One possible explanation is that the dolichos-staining organisms that remain 48 hours after treatment are in a separate, hypnozoite-like life stage that is not affected by the compounds.

JAG050 and JAG021 do not appear to have a microbicidal effect on the RH—YFP parasites. However, in comparing the two conditions in which cells and parasites were exposed to the drug for 6 days, it does not appear that the parasites or host cells are degrading the compounds. In order to cure toxoplasmosis, a companion drug that can work synergistically with the compounds of the present invention may be helpful. Primaquine and tafenoquine, which are the only medicines that can treat the hypnozoite stage of *Plasmodium vivax* and *P. ovale*, may be potential candidates. We had demonstrated synergy with an earlier generation compound with atovaquone and additive effect with cycloguanil.[26]

JAG21 demonstrated high efficacy against Toxopalsma tachyzoites in our vitro and in vivo models, low nanoM efficacy against drug resistant *P. falciparum*, and single dose causal prophylaxis in a mouse model of *P. berghei* sporozoites infection

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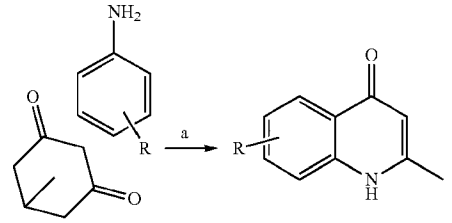
Example 3: Synthesis and Activity of Compounds

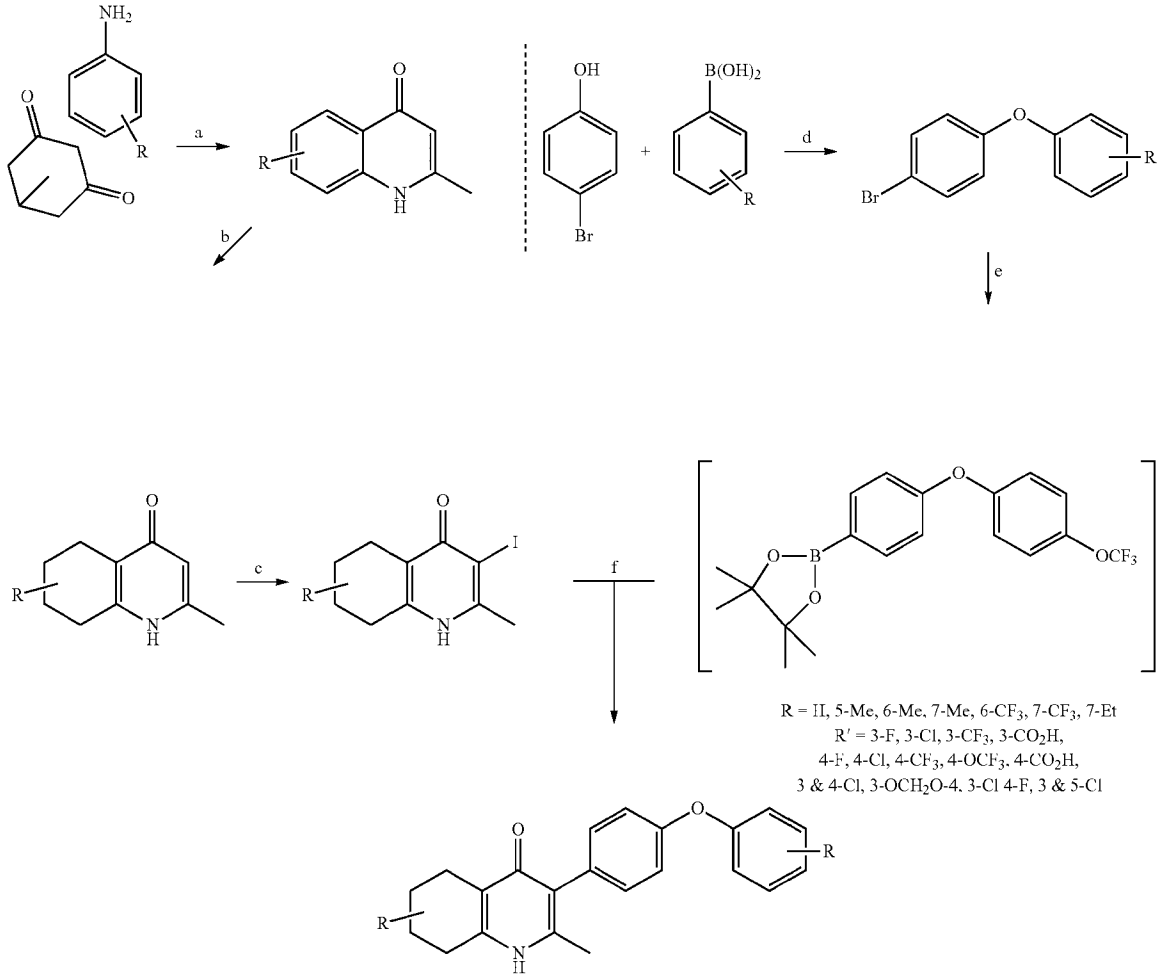
All reagents and solvents were purchased from commercial sources. All commercial reagents and solvents were used as received without further purification. The reactions were monitored using analytical thin layer chromatography (TLC) with 0.25 mm EM Science silica gel plates (60F-254). The developed TLC plates were visualized by short wave UV light (254 nm) or immersion in potassium permanganate solution followed by heating on a hot plate. Flash chromatography was performed with Selecto Scientific silica gel, 32-63 μm particle sizes. All reactions were performed in flame or oven-dried glassware under a nitrogen atmosphere. All reactions were stirred magnetically at ambient temperature unless otherwise indicated. ^{1}H NMR spectra were obtained with a Bruker DRX400, Varian VXR400 or VXR300. ^{1}H NMR spectra were reported in parts per million (δ) relative to TMS (0.0), DMSO-d6 (2.50) or CD_3OD (4.80) as an internal reference. All ^{1}H NMR spectra were taken in CDCl3 unless otherwise indicated.



a) i) Meldrums acid, triethylorthoacetate, 110° C., ii) Aniline, 110° C., iii) Dowtherm A, 250° C.

General Method A

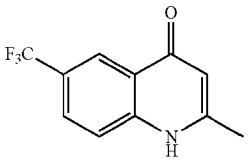
2,2-Dimethyl-1,3-dioxane-4,6-dione (1.5 equiv.) was dissolved in trimethylorthoacetate (2 equiv.) and heated to 115° C. for 2 hrs. The reaction was cooled to allow the addition of the aniline (1 equiv.) before being heated to 115° C. for a further 2 hrs. The reaction mixture was then allowed to cool and was concentrated in vacuo, remaining solvent was washed off with cold methanol. The precipitate was then dissolved in minimum volume of Dowtherm A and refluxed



a) i) Meldrums acid, triethylorthoacetate, 110° C., ii) Aniline, 110° C., iii) Dowtherm A, 250° C., b) PtO_2, H_2, AcOH, c) NIS, Acetonitrile, 80° C., d) $Cu(OAc)_2$, Pyridine, TEA, DCM, e) $Pd(dppf)Cl_2$, Bispinolatodiborane, KOAc, DMF, 80° C. f) $Pd(dppf)Cl_2$, Na_2CO_3, DMF, 80° C.

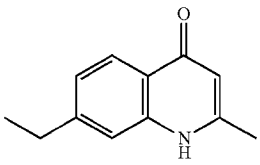
at 250° C. for 1.5 hours. The reaction mixture was allowed to cool and the precipitate filtered followed by washing with hexane to afford the title compound.

2-Methyl-6-(trifluoromethyl)quinolin-4(1H)-one



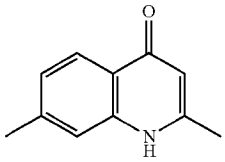
The title compound was synthesised by general method A using 4-trifluoromethyl aniline (2.00 g, 12. 4 mmol) to yield the title compound as a white amorphous solid (466 mg, 2.05 mmol, 17%). ^{1}H NMR (300 MHz, MeOD) δ 8.42 (s, 1H), 7.81 (dd, J=8.8 Hz, 2.1 Hz, 1H), 7.60 (d, J=8.8 Hz, 1H), 6.16 (s, 1H), 2.40 (s, 3H); M/Z (ESI+); 228.06 (Found MH$^+$228.0634, $C_{11}H_8F_3NO$ requires 228.0630).

7-ethyl-2-methylquinolin-4(1H)-one



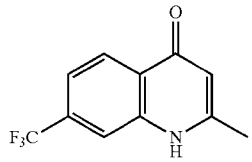
The title compound was synthesised following general procedure A from 3-ethylaniline (1.4 mL, 11.1 mmol). The title compound was isolated as a colourless solid (210 mg, 1.12 mmol, 10%). ^{1}H NMR (500 MHz, CDCl3) δ 9.72 (s, 1H), 8.17 (d, J=8.3 Hz, 1H), 7.16 (s, 1H), 7.10 (d, J=8.3 Hz, 1H), 6.07 (s, 1H), 2.66 (q, J=7.6 Hz, 2H), 2.33 (s, 3H), 1.18 (t, J=7.6 Hz, 3H);

2,7-Dimethylquinolin-4(1H)-one

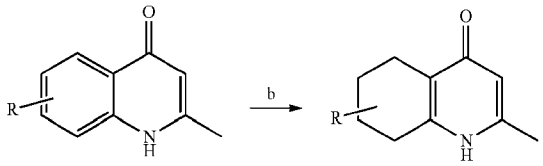


The title compound was synthesised following general procedure A from 3-methylaniline (7.5 mL, 42 mmol). The title compound was isolated as a colourless solid (370 mg, 2.13 mmol, 5%). ^{1}H NMR (400 MHz, CDCl3) δ 9.58 (s, 1H), 8.24 (d, J=8.3 Hz, 1H), 7.22 (s, 1H), 7.16 (d, J=8.3 Hz, 1H), 6.15 (s, 1H), 2.46 (s, 3H), 2.41 (s, 3H).

7-trifluormethyl-2-methylquinolin-4(1H)-one



The title compound was synthesised following general procedure A from 3-trifluoromethyl-aniline (3 mL, 24 mmol). The title compound was isolated with its regiomer and separation was not achieved and so was carried forwards as a mixture (1.5 g).

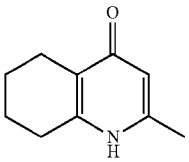


b) PtO_2, H_2, AcOH

General Method B

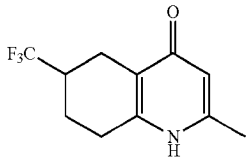
The 4-hydroxylquinolone (1 equiv.) was dissolved in acetic acid (10.0 mL) under inert conditions. platinum dioxide (5% weight equiv.) was added and a hydrogen balloon was attached. The reaction was left to proceed for 12 hours. The resulting suspension was filtered through a pad of Celite and washed with ethyl acetate (10.0 mL). The filtrate was concentrated in vacuo to afford a yellow/brown oil. Purification by column chromatography (10% methanol in chloroform) afforded the title compound.

2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



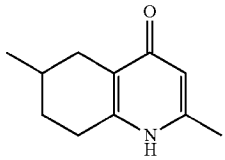
A solution of 4-hydroxyl 2-methyl-quinolone (1.00 g, 6.28 mmol) in acetic acid (10.0 mL) was catalytically hydrogenated over platinum dioxide (0.10 g, 0.44 mmol) for 12 hours. The resulting suspension was filtered through a pad of Celite and washed with ethyl acetate (10.0 mL). The filtrate was concentrated in vacuo to afford a yellow/brown oil. Purification by column chromatography (10% methanol in chloroform) afforded the title compound as a colourless amorphous solid. (1.02 g, 6.25 mmol, 99%). δ H NMR; (500 MHz, Chloroform-d); δ 6.29 (s, 1H), 2.71 (t, J=6.1 Hz, 2H), 2.48 (t, J=6.1 Hz, 2H), 2.32 (s, 3H), 1.78-1.69 (m, 4H); M/Z (ESI+); 164.1122 (Found MH+, 164.11 $C_{10}H_{13}NO$ requires 164.1075).

2-Methyl-6-(trifluoromethyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one



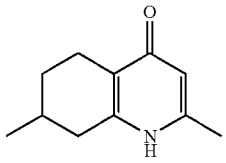
The title compound was synthesised following general procedure B from 2-methyl-6-(trifluoromethyl)quinolin-4(1H)-one (466 mg, 2.0 mmol), The title compound was isolated as colourless solid (230 mg, 0.99 mmol, 49%). ^{1}H NMR (500 MHz, MeOD) δ 6.32 (s, 1H), 2.94 (dd, J=16.7, 5.1 Hz, 1H), 2.86 (dd, J=8.4, 4.1 Hz, 2H), 2.65-2.52 (m, 1H), 2.41 (dd, J=22.8, 11.5 Hz, 1H), 2.36 (s, 3H), 2.28-2.15 (m, 1H), 1.75 (tt, J=12.8, 9.1 Hz); M/Z (ESI+); 232.10 (Found MH$^+$; 232.0955, $C_{11}H_{12}F_3NO$ requires 232.0949).

2,6-Dimethyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



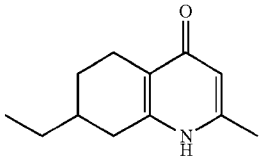
The title compound was synthesised following general procedure B from 2,6-dimethyl-quinolin-4(1H)-one (1.0 g, 5.78 mmol), The title compound was isolated as a colourless solid (780 mg, 4.30 mmol. 76%). ^{1}H NMR (400 MHz, CDCl3) δ 12.26 (s, 1H), 6.09 (s, 1H), 2.88-2.64 (m, 3H), 2.30 (s, 3H), 1.98 (dd, J=16.9, 10.1 Hz, 1H), 1.87 (d, J=12.4 Hz, 1H), 1.77 (m, 1H), 1.39 (ddd, J=23.9, 11.1, 6.0 Hz, 1H), 1.08 (d, J=6.5 Hz, 3H); M/Z (ESI+); 178.13 (Found MH$^+$; 178.1280, $C_{11}H_{15}NO$ requires 177.1154).

2,7-Dimethyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



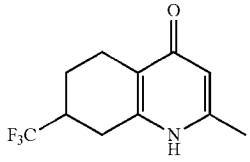
The title compound was synthesised following general procedure B from 2,7-Dimethyl-quinolin-4(1H)-one (350 mg, 2.0 mmol). The title compound was isolated as a colourless solid (311 mg, 1.75 mmol, 88%). ^{1}H NMR (500 MHz, MeOD) δ 6.31 (s, 1H), 2.78 (dd, J=17.0, 5.1 Hz), 2.73 (ddd, J=17.7, 5.2, 2.7 Hz), 2.45-2.31 (m, 2H), 2.00-1.87 (m, 2H), 2.37 (s, Me), 1.37 (m, 2H), 1.13 (d, J=6.6 Hz, 3H); M/Z (ESI+); 178.13 (Found MH$^+$; 178.1278, $C_{11}H_{15}NO$ requires 177.1154).

7-ethyl-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one

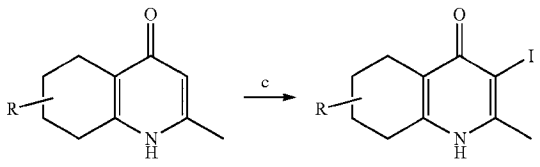


The title compound was synthesised following general procedure B from 7-ethyl-2-methylquinolin-4(1H)-one (420 mg, 1.78 mmol). The title compound was isolated as a colourless solid (360 mg, 1.5 mmol, 84%). H NMR (400 MHz, CDCl3) δ 12.20 (s, 1H), 6.09 (s, 1H), 2.88-2.66 (m, 2H), 2.46-2.19 (m, 5H, 2-Me), 1.95 (d, J=13.0 Hz, 1H), 1.63 (s, 1H), 1.38 (td, J=13.9, 6.9 Hz, 2H), 1.33-1.22 (m, 1H) 0.94 (t, J=7.4, 3H); M/Z (ESI+); 192.14 (Found MH$^+$; 192.1378 $C_{12}H_{17}NO$ requires 192.1383).

7-trifluormethyl-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



The title compound was synthesised following general procedure B from a mixture of 7-trifluormethyl-2-methylquinolin-4(1H)-one & 5-trifluormethyl-2-methylquinolin-4(1H)-one (1.5 g). The title compound was isolated as a colourless solid (495 mg, 2.14 mmol). ^{1}H NMR (500 MHz, $CDCl_3$/MeOD, 1:1) δ 5.99 (s, 1H), 2.73-2.60 (m, 2H), 2.53 (dd, J=16.3, 12.0 Hz, 1H), 2.35 (s, 1H), 2.20 (ddd, J=17.7, 11.4, 5.9 Hz, 1H), 2.11 (s, 3H), 2.09-2.01 (m, 1H), 1.42 (ddd, J=25.0, 12.1, 5.7 Hz, 1H); M/Z (ESI+); 232.10 (Found MH$^+$; 232.0953, $C_{11}H_{12}F_3NO$ requires 232.0949).



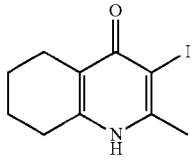
c) NIS, Acetonitrile, 80° C.

General Method C

Potassium iodide solution (sat aq. 5.60 mL mmol^{-1}) and n-butylamine (10 equiv.) were added to a solution of the tetrahydroquinolin-4(1H)-one (1 equiv.) and iodine (1 equiv.) in DMF (10.0 mL). The reaction mixture was stirred at room temperature for 16 hours. Observed colour change from dark purple to orange. Sodium thiosulphate (250 mg in

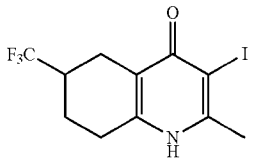
10.0 mL water) was then added causing precipitation of a colourless solid. Filtration (washed 2×10 mL water) afforded the title compound.

4(1H), 3-iodo-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



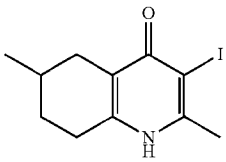
Saturated potassium iodide solution (sat aq, 5.60 mL) and n-butylamine (5.80 mL, 58.3 mmol) was added to a solution of 2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one (0.95 g, 5.83 mmol) and iodine (1.48 g, 5.83 mmol) in DMF (10.0 mL). The reaction mixture was stirred at room temperature for 16 hours. Observed colour change from dark purple to orange. Sodium thiosulphate (250 mg in 10.0 mL water) was then added followed by filtration (washed 2×10 mL water) to afford the title compound (39) as colourless microcrystals (1.45 g, 5.02 mmol, 86%). 1H NMR (500 MHz, methanol-d4); δ 2.53 (t, J=6.1 Hz, 2H), 2.44 (s, 3H), 2.30 (t, J=6.1 Hz, 2H), 1.73-1.67 (m, 2H), 1.67-1.61 (m, 2H); M/Z (ESI+); 290.00 (Found MH+, 290.0037 $C_{10}H_{12}INO$ requires 290.0036).

3-Iodo-2-methyl-6-(trifluoromethyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one



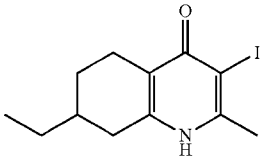
The title compound was synthesised following general procedure C from 2-methyl-6-(trifluoromethyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one (230 mg, 1.0 mmol). The title compound was isolated as colourless solid (300 mg, 0.84 mmol, 84%). ^{1}H NMR (400 MHz, DMSO) δ 11.58 (s, 1H), 2.75 (d, J=5.6 Hz, 1H), 2.72-2.67 (m, 2H), 2.62 (dd, J=7.4, 5.8 Hz, 1H), 2.46 (s, 3H), 2.31 (d, J=22.7 Hz, 1H), 2.15 (dd, J=16.4, 11.1 Hz, 1H), 2.07 (dd, J=6.6, 5.4 Hz, 1H), 1.68-1.51 (m, 1H); M/Z (ESI+); 357.99 (Found MH$^+$; 357.9914, $C_{11}H_{11}F_3INO$ requires 357.9910).

3-Iodo-2·6-dimethyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



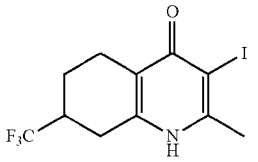
The title compound was synthesised following general procedure C from 2,6-dimethyl-5,6,7,8-tetrahydro quinolin-4(1H)-one (750 mg, 4.24 mmol). The title compound was isolated as colourless solid (740 mg, 2.44 mmol, 58%). ^{1}H NMR (500 MHz, $CDCl_3$/MeOD) δ 2.36 (dd, J=17.3, 4.8 Hz), 2.25 (d, J=4.8 Hz, 2H), 2.15 (s, 3H), 1.57 (dd, J=17.3, 10.4 Hz, 1H), 1.51 (d, J=10.9 Hz, 1H), 1.36 (s, 1H), 1.09-0.94 (m, 1H), 0.69 (d, J=6.6 Hz, 3H); M/Z (ESI+); 304.02 (Found MH$^+$; 304.0190, $C_{11}H_{14}INO$ requires 304.0193).

3-Iodo-7-ethyl-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one

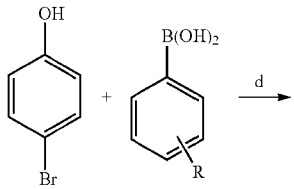


The title compound was synthesised following general procedure C from 7-ethyl-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one (110 mg, 0.57 mmol). The title compound was isolated as colourless solid (180 mg, 0.57 mmol, 99%). ^{1}H NMR (400 MHz, MeOD) δ 2.62 (dd, J=17.2, 4.4 Hz, 2H), 2.47 (s, 3H), 2.34-2.22 (m, 1H), 2.18 (dd, J=17.8, 9.6 Hz, 1H), 1.92-1.81 (m, 1H), 1.64-1.50 (m, 1H), 1.34 (dtd, J=14.1, 7.2, 2.2 Hz, 2H), 1.21 (ddd, J=24.1, 10.9, 5.6 Hz, 1H), 0.91 (t, J=7.4 Hz, 3H); M/Z (ESI+); 318.03 (Found MH$^+$; 318.0261, C12H16INO requires 318.0349).

3-Iodo-7-trifluormethyl-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



The title compound was synthesised following general procedure C from 7-trifluormethyl-2-methylquinolin-4(1H)-one (480 mg, 2.10 mmol). The title compound was isolated as a colourless solid (688 mg, 1.92 mmol, 91%). ^{1}H NMR (500 MHz, $CDCl_3$/MeOD, 1:1) δ 2.40 (dd, J=16.0, 4.9 Hz, 2H), 2.24 (dd, J=17.9, 8.3 Hz, 1H), 2.20-2.12 (m, 1H), 2.12 (s, 2H), 2.00-1.86 (m, 1H), 1.94 (s, 1H), 1.75 (s, 1H), 1.74 (dd, J=13.5, 5.9 Hz, 1H), 1.13 (ddd, J=19.4, 12.1, 5.8 Hz, 1H). M/Z (ESI+); 357.99 (Found MH$^+$; 357.9915, $C_{11}H_{11}F_3INO$ requires 357.9910).



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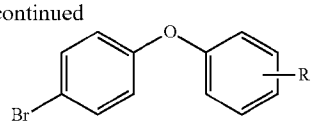
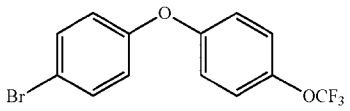


d) $Cu(OAc)_2$, Pyridine, TEA, DCM.

General Procedure d

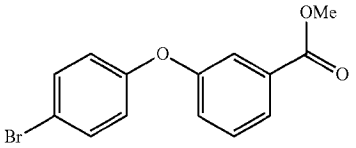
Copper (II) acetate (1 equiv.), triethylamine (5 equiv.), and pyridine (5 equiv.) was added to a solution of the boronic acid (1.5 equiv.) and phenol (1 equiv.) in dichloromethane (10 mL $mmol^{-1}$) over heat-activated 4 Å molecular sieves. The reaction mixture was stirred over 16 hours at room temperature. The reaction mixture was quenched with HCl (0.5 M, 20 mL $mmol^{-1}$) and filtered through a pad of Celite, followed by repeated washing with water (10 mL $mmol^{-1}$). The organic layer was extracted with brine, dried over magnesium sulphate, and concentrated in vacuo. Purification by silica gel chromatography (ethyl acetate/hexane) afforded the title compound.

1-Bromo-4-(4-(trifluoromethoxy)phenoxy)benzene



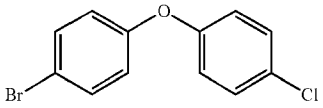
The title compound was synthesised, from 4-bromophenol (0.42 g, 2.43 mmol) and 4-trifluoromethoxy benzene boronic acid (1.00 g, 4.68 mmol), according to general procedure d as a colourless oil (70%, 0.53 g, 1.63 mmol). δ H NMR (500 MHz, Chloroform-d) δ 7.37 (d, J=9.0 Hz, 2H), 7.10 (d, J=9.1 Hz, 2H), 6.91 (d, J=9.1 Hz, 2H), 6.80 (d, J=9.0 Hz, 2H);

Methyl 3-(4-bromophenoxy)benzoate



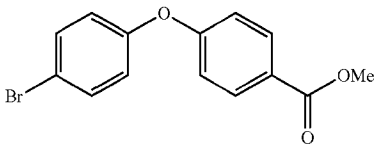
The title compound was synthesised, from 4-bromo-pehonl (0.42 g, 2.43 mmol) and 3-methoxycarbonyl phenyl boronic acid (0.43 g, 2.43 mmol), according to general procedure D. The title compound (43) was isolated as colourless glassy solid (0.21 g, 0.69 mmol, 28%). ^1H NMR (500 MHz, $CDCl_3$) δ 7.73 (dt, J=7.7, 1.2 Hz, 1H), 7.59-7.53 (m, 1H), 7.38 (d, J=9.0 Hz, 2H), 7.34 (t, J=8.4 Hz, 2H), 7.13 (ddd, J=8.4, 2.5, 0.9 Hz, 1H), 6.82 (d, J=9.0 Hz, 2H), 3.83 (s, 3H); M/Z (ESI+); 307.00 (Found MH^+, 306.9962 $C_{14}H_{11}BrO_3$ requires; 306.9964).

1-bromo-4-(4-chlorophenoxy)benzene



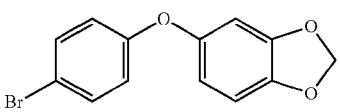
The title compound was synthesised, from 4-bromophenol (0.5 g, 2.80 mmol) and 4-trifluoromethoxy benzene boronic acid (0.6 g, 4.20 mmol), according to general procedure D. The title compound was isolated as a colourless needles (160 mg, 0.56 mmol, 20%). 1H NMR (500 MHz, $CDCl_3$) δ 7.43 (d, J=9.0 Hz, 2H), 7.30 (d, J=9.0 Hz, 2H), 6.93 (d, J=9.0 Hz, 2H), 6.86 (d, J=9.0 Hz, 2H).

Methyl 4-(4-bromophenoxy)benzoate

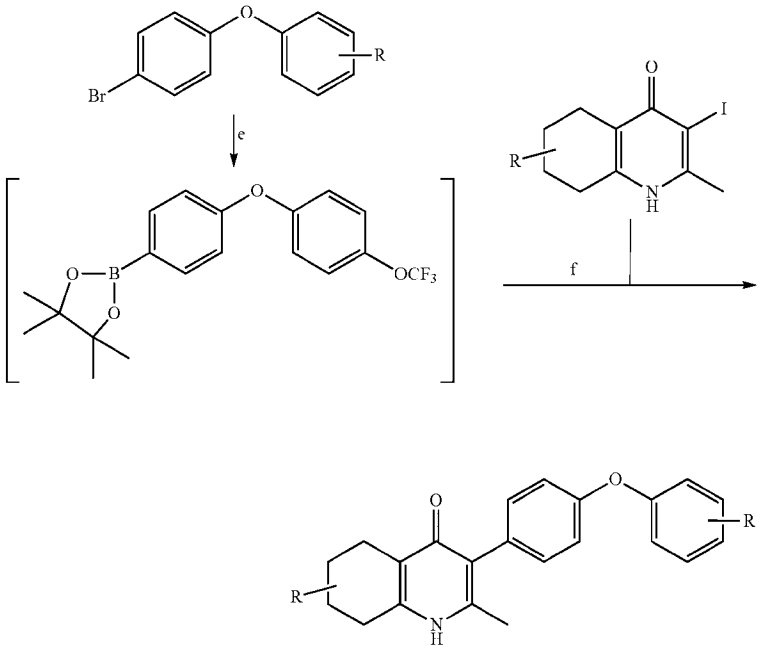


The title compound was synthesised, from 4-bromo-pehonl (0.42 g, 2.43 mmol) and 4-methoxycarbonyl phenyl boronic acid (0.43 g, 2.43 mmol), according to general procedure D. The title compound was isolated as colourless plate crystals (0.25 g, 0.81 mmol, 33%). 1H NMR (500 MHz, $CDCl_3$) δ 7.94 (d, J=8.9 Hz, 2H), 7.41 (d, J=8.9 Hz, 2H), 6.91 (d, J=8.8 Hz, 2H, 6.87 (d, J=8.9 Hz, 2H), 3.83 (s, 3H); M/Z (ESI+); 307.00 (Found MH^+, 306.9961 $C_{14}H_{11}BrO_3$ requires 306.9964).

5-(4-bromophenoxy)-2H-1,3-benzodioxole



The title compound was synthesised, from 4-bromo-pehonl (500 mg, 2.89 mmol) and 3,4-methyleneoxy-phenylboronic acid (719 mg, 4.34 mmol according to general procedure D. The title compound was isolated as a pale yellow oil (196 mg, 0.67 mmol, 23%). δ ^1H NMR (500 MHz, Chloroform-d) δ 7.43 (d, J 8.5 Hz, 2H), 6.86 (d, J 8.5 Hz, 2H), 6.79 (d, J 8.5 Hz, 1H), 6.59 (d, J 2.5 Hz, 1H), 6.51 (dd, J 8.5 & 2.5 Hz, 1H), 6.00 (s, 2H);

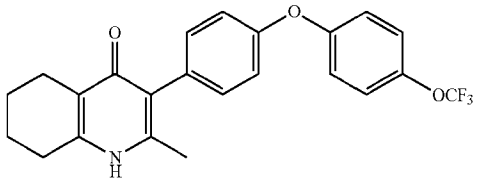


c) Pd(dppf)Cl_2, Bispinocolatodiborane, KOAc, DMF, 80° C. f) Pd(dppf)Cl_2, Na_2CO_3, DMF, 80° C.

1.1 General Method E &F

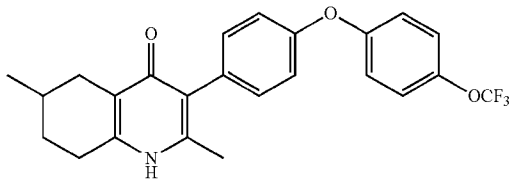
A flask charged with the 4-bromo-diarylether (1 equiv.), bispinocolatodiborane (1.1 equiv.), KOAc (3 equiv.) and Pd(dppf)Cl_2 (3 mol %) was flushed with nitrogen. DMF (2.00 mL) was added and the reaction was stirred at 80° C. for 18 hours. After cooling the solution to room temperature, 3-iodotetrahydroquinoline (2 equiv.), $PdCl_2$(dppf) (3 mol %) and Na_2CO_3 (2M, 5 equiv.) were added and the mixture was stirred at 80° C. under nitrogen for a further 24 hours. The solution was cooled to room temperature, the product was extracted with Et_2O (15.0 mL). The organic layers were combined and washed with H_2O (15.0 mL), brine and dried over $MgSO_4$ and concentrated in vacuo. This was followed by purification by silica gel chromatography (ethyl acetate/petroleum ether).

2-methyl-3-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one



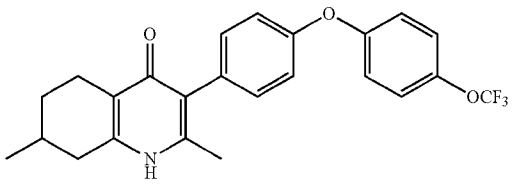
The title compound was synthesised following general procedure E&F from 3-iodo-5,6,7,8-tetrahydroquinolin-4(1H)-one (260 mg, 0.9 mmol) and 4-bromo(4-trifluromethoxyphenoxy)phenyl (200 mg, 0.6 mmol). The title compound was isolated as colourless solid (38 mg, 0.09 mmol, 15%). ^{1}H NMR (500 MHz, DMSO) δ 11.07 (s, 1H), 7.40 (d, J=8.5 Hz, 2H), 7.19 (d, J=8.6 Hz, 2H), 7.13 (d, J=9.0 Hz, 2H), 7.02 (d, J=8.5 Hz, 2H), 2.54 (t, J=6.0 Hz, 2H, H-8), 2.28 (t, J=5.9 Hz, 2H), 2.07 (s, 3H), 1.71 (m, 2H), 1.65 (m, 2H); M/Z (ESI+); 416.15 (Found MH^+, 416.1492 $C_{23}H_{20}F_3NO_3$ requires 416.1473).

3(4-(4-trifluoromethoxyphenoxy)phenyl)-2,6-dimethyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



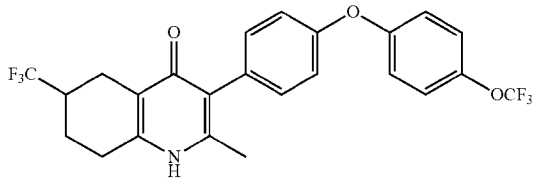
The title compound was synthesised following general procedure E&F from 3-iodo-2,6-dimethyl-5,6,7,8-tetrahydroquinolin-4(1H)-one (270 mg, 0.9 mmol) and 4-bromo(4-trifluromethoxyphenoxy)phenyl (200 mg, 0.6 mmol). The title compound was isolated as colourless solid (40 mg, 0.09 mmol, 15%). ^{1}H NMR (500 MHz, $CDCl_3$/MeOD) δ 6.86 (d, J=8.3 Hz, 4H), 6.79-6.64 (m, 4H), 2.43 (dd, J=17.5, 4.9 Hz, 1H), 2.37 (s, 2H), 1.81 (s, 3H), 1.70-1.54 (m, 2H), 1.45 (m, 1H), 1.09 (dt, J=20.7, 10.5 Hz, 1H), 0.76 (d, J=6.6 Hz, 3H); M/Z (ESI+); 452.14 (Found MNa^+; 452.1446 $C_{24}H_{22}F_3NO_3$ requires 452.1444).

3(4-(4-trifluoromethoxyphenoxy)phenyl)-2,7-dimethyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



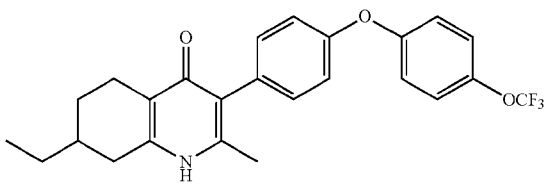
The title compound was synthesised following general procedure E&F from 3-iodo-2,7-dimethyl-5,6,7,8-tetrahydroquinolin-4(1H)-one (120 mg, 0.4 mmol) and 4-bromo(4-trifluromethoxyphenoxy)phenyl (87 mg, 0.27 mmol). The title compound was isolated as colourless solid (30 mg, 0.07 mmol, 26%). ^{1}H NMR (400 MHz, DMSO) δ 10.90 (s, 1H), 7.41 (d, J=8.5 Hz, 2H), 7.20 (d, J=7.9 Hz, 2H), 7.14 (d, J=8.5 Hz, 2H), 7.03 (d, J=7.9 Hz, 2H), 2.61 (m, 2H), 2.20 (dd, J=16.4, 9.6 Hz, 2H), 2.08 (s, 3H), 1.81 (m, 2H), 1.24 (s, 1H), 1.04 (d, J=5.9 Hz, 3H); M/Z (ESI+); 452.14 (Found MNa$^+$; 452.1446 $C_{24}H_{22}F_3NO_3$ requires 452.1444).

6-Ethyl-3-(4-(4-trifluoromethoxyphenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



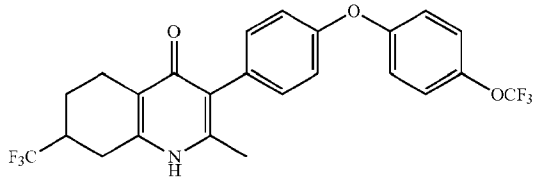
The title compound was synthesised following general procedure E&F from 3-iodo-2-methyl-6-(trifluoromethyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one (300 mg, 0.84 mmol) and 4-bromo-(4-trifluromethoxyphenoxy)phenyl (185 mg, 0.56 mmol). The title compound was afforded as a colourless solid. (20 mg, 0.04 mmol, 7%). ^{1}H NMR (500 MHz, TFA) δ 7.39 (d, J=7.7 Hz, 4H), 7.34 (d, J=7.1 Hz, 2H), 7.24 (d, J=8.5 Hz, 2H), 3.32 (d, J=14.9 Hz, 2H), 3.21 (dd, J=13.8, 5.6 Hz, 1H), 2.89 (dd, J=18.0, 9.6 Hz, 1H), 2.73 (dd, J=15.3, 6.9 Hz, 1H), 2.57 (s, 3H), 2.52 (d, J=13.5 Hz, 1H), 2.07 (d, J=16.0 Hz, 1H, H-7b); M/Z (ESI+); 484.14 (Found MH$^+$ 484.1358, $C_{24}H_{19}F_6NO_3$ requires 484.1342).

7-Ethyl-2-methyl-3(4-(4-trifluoromethoxyphenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one



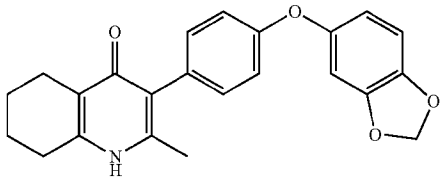
The title compound was synthesised following general procedure E&F from 7-ethyl-2-methylquinolin-4(1H)-one (170 mg, 0.54 mmol) and 4-bromo-(4-trifluromethoxyphenoxy)phenyl (120 mg, 0.36 mmol). The title compound was isolated as a colourless solid (17 mg, 0.04 mmol, 11%). ^{1}H NMR (500 MHz, $CDCl_3$) δ 11.50 (s, 1H), 7.16 (dd, J=15.3, 8.2 Hz, 4H), 6.94 (dd, J=16.6, 8.3 Hz, 4H), 2.78 (d, J=16.1 Hz, 1H), 2.60 (d, J=21.4 Hz, 1H), 2.39 (s, 1H), 2.13 (dd, J=15.8, 10.8 Hz, 1H), 1.96 (s, 3H), 1.59 (s, 1H), 1.43-1.32 (m, 2H), 1.32-1.18 (m, 2H), 0.94 (t, J=7.2 Hz, 3H); M/Z (ESI+); 444.18 (Found MH$^+$; 444.1784, $C_{25}H_{24}F_3NO_3$ requires 444.1781).

7-trifluromethyl-2-methyl-3(4-(4-trifluoromethoxyphenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one

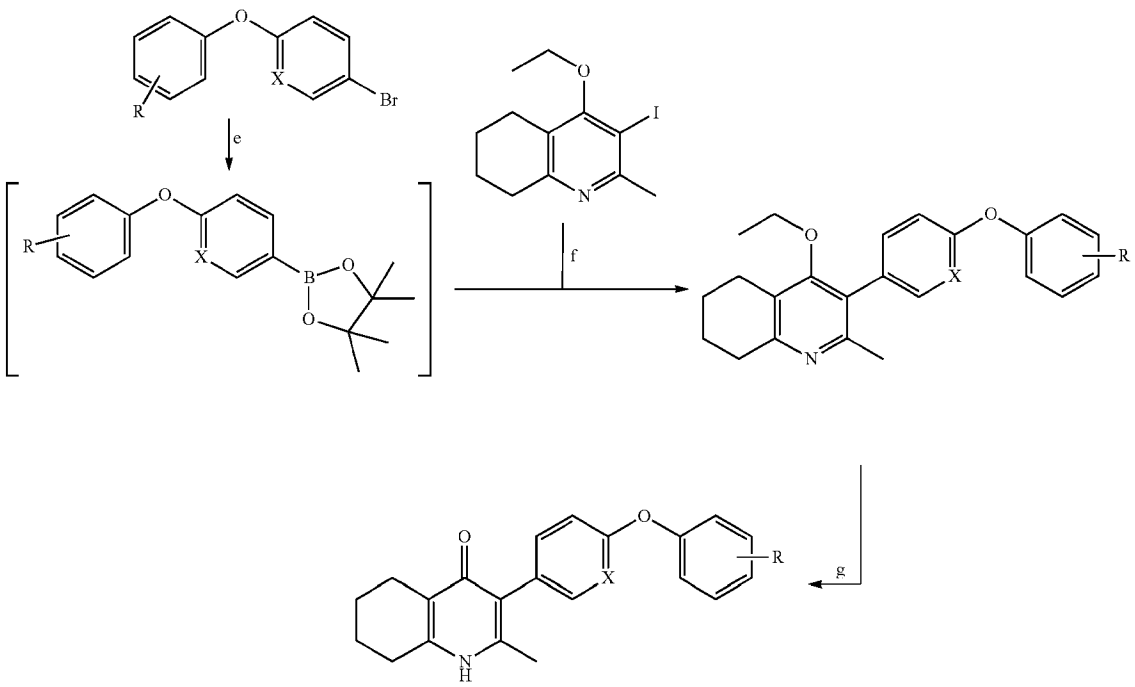


The title compound was synthesised following general procedure x from 7-trifluormethyl-2-methylquinolin-4(1H)-one (360 mg, 0.99 mmol) and 4-bromo-(4-trifluromethoxyphenoxy)phenyl (220 mg, 0.66 mmol). The title compound was isolated as a colourless solid (95 mg, 0.20 mmol. 30%). M/Z (ESI+); 484.14 (Found MH+484.1358, $C_{24}H_{19}F_6NO_3$ requires 484.1342).

3-[4-(2H-1,3-benzodioxol-5-yloxy)phenyl]-2-methyl-5,6,7,8-tetrahydro-1H-quinolin-4-one



The title compound was synthesised from 5-(4-bromophenoxy)-2H-1,3-benzodioxole (200 mg, 0.68 mmol) and 3-iodo-5,6,7,8-tetrahydroquinolin-4(1H)-one (288 mg, 1.02 mmol). The title compound was isolated as a pale grey solid (40 mg, 0.11 mmol, 16%). HPLC; 3.28 min (86%); δ ^{1}H NMR (500 MHz, TFA) δ 7.35 (d, J 8.5 Hz, 2H), 7.31-7.29 (m, 3H), 6.98 (s, 1H), 6.79 (s, 1H), 6.10 (s, 2H), 3.09 (t, J 5.0 Hz, 2H), 2.90 (t, J 5.0 Hz, 2H), 2.52 (s, 3H), 2.11-2.05 (m, 4H); M/Z (ESI); 375.1563, ($C_{23}H_{21}NO_4$ requires 375.1471).

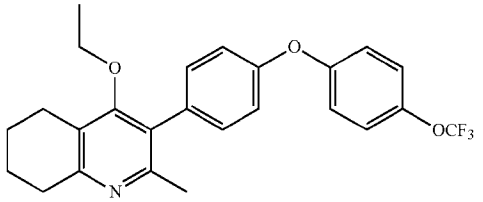


e) $Pd(dppf)Cl_2$, Bispinocolatodiborane, KOAc, DMF, 80° C. f) $Pd(dppf)Cl_2$, Na_2CO_3, DMF, 80° C., g) HBr, AcOH 120° C.

1.2 General Method E &F

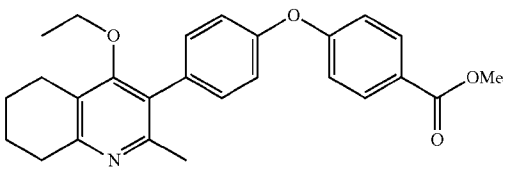
A flask charged with the 4-bromo-diarylether (1 equiv.), bispinocolatodiborane (1.1 equiv.), KOAc (3 equiv.) and $Pd(dppf)Cl_2$ (3 mol %) was flushed with nitrogen. DMF (2.00 mL) was added and the reaction was stirred at 80° C. for 18 hours. After cooling the solution to room temperature, 3-iodotetrahydroquinoline (2 equiv.), $PdCl_2(dppf)$ (3 mol %) and Na_2CO_3 (2M, 5 equiv.) were added and the mixture was stirred at 80° C. under nitrogen for a further 24 hours. The solution was cooled to room temperature, the product was extracted with Et_2O (15.0 mL). The organic layers were combined and washed with H_2O (15.0 mL), brine and dried over $MgSO_4$ and concentrated in vacuo. This was followed by purification by silica gel chromatography (ethyl acetate/petroleum ether).

4-ethoxy-2-methyl-3-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-5,6,7,8-tetrahydro quinoline



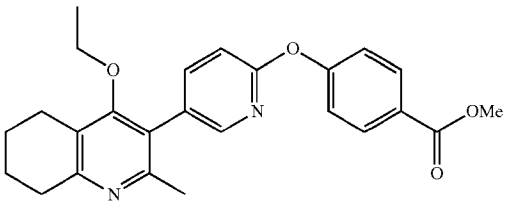
The title compound was synthesised from 1-Bromo-4-(4-(trifluoromethoxy)phenoxy)benzene (100 mg, 0.30 mmol) according to general procedure E&F, to afford the title compound as a colourless gum/viscous oil (30 mg, 0.07 mmol, 23%). ^1H NMR (500 MHz, Acetone) δ 7.28 (d, J=8.7 Hz, 2H), 7.26 (d, J=9.1 Hz, 2H), 7.09 (d, J=9.1 Hz, 2H), 7.07 (d, J=8.7, 2H), 3.52 (q, J=7.0 Hz, 2H), 2.85 (t, J=6.5 Hz, 2H), 2.78 (t, J=6.2 Hz, 2H), 2.26 (s, 3H), 1.89-1.81 (m, 2H), 1.81-1.72 (m, 2H), 0.93 (t, J=7.0 Hz, 3H); M/Z (ESI+); 444.18 (Found MH^+, 444.1792 $C_{25}H_{24}F_3NO_3$ requires 444.1781).

Methyl 4-(4-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)phenoxy)benzoate



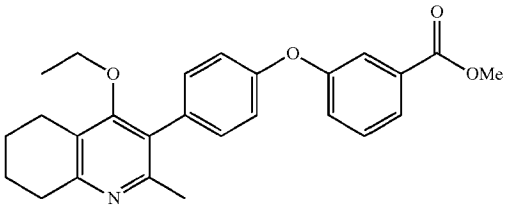
The title compound was synthesised from methyl 4-(4-bromophenoxy)benzoate (150 mg, 0.49 mmol) according to general procedure E&F. The title compound was isolated as colourless microcrystals (56 mg, 0.13 mmol, 27%). ^1H NMR (500 MHz, $CDCl_3$) δ 8.03 (d, J=8.7 Hz, 2H), 7.29 (d, J=8.4 Hz, 2H), 7.11 (d, J=8.4 Hz, 2H), 7.03 (d, J=8.7 Hz, 2H), 3.90 (s, 3H), 3.51 (q, J=7.0 Hz, 2H), 2.91 (t, J=6.2 Hz, 2H), 2.72 (t, J=6.0 Hz, 2H), 2.32 (s, 3H), 1.91-1.84 (m, 2H), 1.83-1.76 (m, 2H), 1.04 (t, J=7.0 Hz, 3H); M/Z; 418.20 (ESI+); 418.20 (Found MH^+, 418.2037 $C_{26}H_{27}NO_4$ requires 418.2018).

Methyl 4-((5-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)pyridin-2-yl)oxy) benzoate



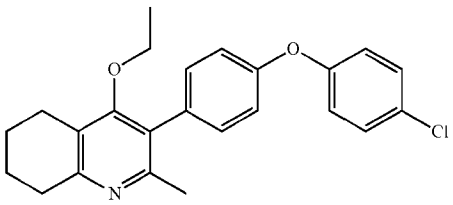
The title compound was synthesised from methyl 3-((5-bromopyridin-2-yl)oxy)benzoate (100 mg, 0.33 mmol) according to general procedure E&F. The title compound was isolated as a colourless gum/semisolid (30 mg, 0.07 mmol, 21%). 1H NMR (500 MHz, $CDCl_3$) δ 8.13 (d, J=2.3 Hz, 1H, H-6'), 8.10 (d, J=8.7 Hz, 2H), 7.67 (dd, J=8.4, 2.4 Hz, 1H, H-4'), 7.24 (d, J=8.7 Hz, 2H), 7.05 (d, J=8.4 Hz, 1H), 3.91 (s, 3H), 3.53 (q, J=7.0 Hz, 2H), 2.92 (t, J=6.3 Hz, 2H), 2.71 (t, J=6.2 Hz, 2H), 1.06 (t, J=7.0 Hz, 3H); M/Z (ESI+); 419.20 (Found MH^+, 419.1993 $C_{25}H_{26}N_2O_4$ requires 419.1970).

Methyl 3-(4-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)phenoxy)benzoate



The title compound was synthesised from methyl 3-(4-bromophenoxy)benzoate (150 mg, 0.49 mmol) according to general procedure E&F. The title compound was isolated as colourless crystals (50 mg, 0.12 mmol, 25%). 1H NMR (500 MHz, $CDCl_3$) δ 7.73 (d, J=7.7 Hz, 1H), 7.65-7.60 (m, 1H), 7.37 (t, J=7.9 Hz, 1H), 7.19 (m, 3H), 7.00 (d, J=8.6 Hz, 2H), 3.83 (s, J=, 3H), 3.45 (q, J=7.0 Hz, 2H), 2.85 (t, J=6.3 Hz, 2H), 2.65 (t, J=6.2 Hz, 2H), 2.26 (s, 3H), 1.88-1.78 (m, 2H), 1.76-1.67 (m, 2H), 0.99 (t, J=7.0 Hz, 3H); M/Z (ESI+); 418.20 (Found MH^+ 418.2030, $C_{26}H_{27}NO_4$ requires 418.2018).

Formation of: 3-(4-(4-chlorophenoxy)phenyl)-4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinoline

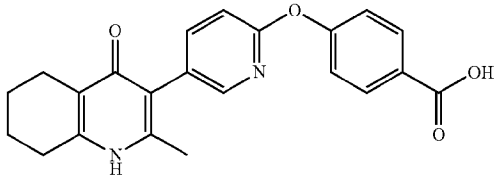


The title compound was synthesised from 1-bromo-4-(4-chlorophenoxy)benzene (70 mg, 0.24 mmol) according to general procedure E&F. The title compound was isolated as colourless oil (20 mg, 0.05 mmol, 27%). 1H NMR (500 MHz, $CDCl_3$) δ 7.25 (d, J=8.9 Hz, 2H), 7.17 (d, J=8.7 Hz, 2H), 6.98 (d, J=8.7 Hz, 2H), 6.93 (d, J=8.9 Hz, 2H), 3.45 (q, J=7.0 Hz, 2H), 2.89 (t, J=6.3 Hz, 2H), 2.65 (t, J=6.1 Hz, 2H), 2.28 (s, 3H), 1.90-1.61 (m, 4H), 0.98 (t, J=7.0 Hz, 3H); M/Z (ESI+); 394.16 (Found MH+394.1575, $C_{24}H_{24}ClNO_2$ requires 394.1568).

1.3 General Method G

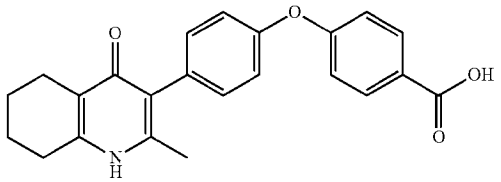
To a solution of the 4-ethoxy-3-(diaryl ether)-hydroxyquinolone (1 equiv.) in acetic acid (2 mL $mmol^{-1}$) was added hydrogen bromide (>48% w/v (aq)) (1 mL $mmol^{-1}$). The reaction mixture was then heated to 90° C. and left to reflux for 72 hours. The reaction mixture was neutralised with sodium hydroxide (2 M, 30.0 mL) and precipitate formed. The reaction mixture was then filtered to afford the title compound and purified.

Methyl 3-((5-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)pyridin-2-yl)oxy) benzoate



The title compound was synthesised from methyl 4-((5-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)pyridin-2-yl)oxy) benzoate (30 mg, 0.07 mmol) according to general procedure G. The title compound was isolated as colourless semi solid (13 mg, 0.03 mmol, 45%). 1H NMR (500 MHz, DMSO) δ 12.83 (s, 1H, NH), 11.03 (s, 1H, CO_2H), 8.00 (d, J=8.7 Hz, 2H), 7.99 (s, 1H) 7.73 (dd, J=8.4. 2.4 Hz, 1H), 7.24 (d, J=8.7 Hz, 2H), 7.12 (d, J=8.4 Hz, 1H), 2.56 (t, J=5.8 Hz, 2H), 2.30 (t, J=5.8 Hz, 2H), 2.11 (s, 3H), 1.82-1.51 (m, 4H); M/Z (ESI+); 377.15 (Found MH^+, 377.1497 $C_{22}H_{20}N_2O_4$ requires 377.1496).

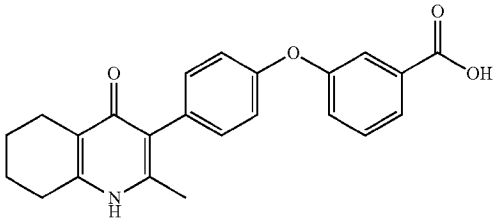
4-(4-(2-methyl-4-oxo-1,4,5,6,7,8-hexahydroquinolin-3-yl)phenoxy)benzoic Acid



The title compound was synthesised from methyl 4-(4-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)phenoxy) benzoate (50 mg, 0.17 mmol) according to general procedure G. The title compound was isolated as colourless crystals (33 mg, 0.09 mmol, 48%). 1H NMR (500 MHz, DMSO) δ 13.79 (s, 1H), 12.80 (s (b), 1H), 7.99 (d, J=8.8 Hz,

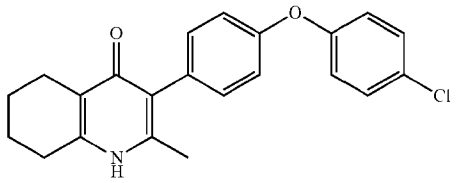
2H), 7.36 (d, J=8.7 Hz, 2H), 7.24 (d, J=8.7 Hz, 2H), 7.14 (d, J=8.8 Hz, 2H), 2.92 (t, J=5.1 Hz, 2H), 2.62 (t, J=5.0 Hz, 2H), 2.31 (s, 3H), 1.90-1.76 (m, 4H); M/Z (ESI+); 376.16 (Found MH^+, 376.1550, $C_{23}H_{21}NO_4$ requires 376.1543).

3-(4-(2-methyl-4-oxo-1,4,5,6,7,8-hexahydroquinolin-3-yl)phenoxy)benzoic Acid

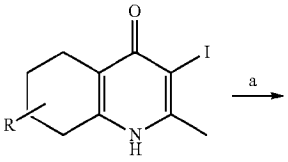


The title compound was synthesised from 3 methyl 3-(4-(4-ethoxy-2-methyl-5,6.7,8-tetrahydroquinolin-3-yl)phenoxy) benzoate (50 mg, 0.17 mmol) according to general procedure G. The title compound was isolated as colourless crystals (7 mg, 0.02 mmol, 12%). ^{1}H NMR (500 MHz, DMSO) δ 13.00 (s, 1H), 11.31 (s, 1H), 7.70 (d, J=7.9 Hz, 1H), 7.52 (t, J=7.9 Hz, 1H), 7.48 (dd, J=2.2, 1.6 Hz, 1H), 7.31 (dd, J=7.9, 1.6 IIz, 1II), 7.21 (d, J=8.6 IIz, 2II), 7.04 (d, J=8.6 Hz, 2H), 2.58 (t, J=5.8 Hz, 2H), 2.32 (t, J=5.8 Hz, 2H), 2.09 (s, 3H), 1.76-1.59 (m, 4H); M/Z (ESI+); 376.15 (Found MH^+ 376.1547, $C_{23}H_{21}NO_4$ requires 376.1543).

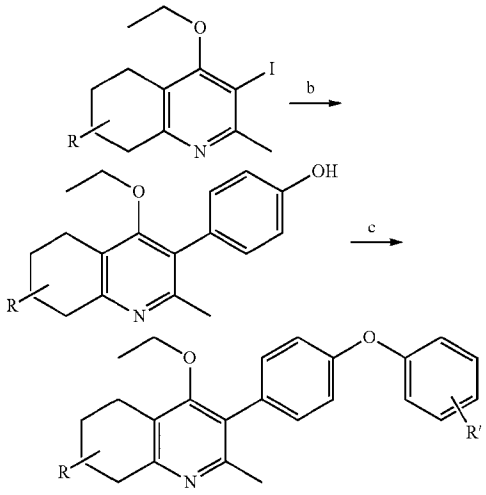
3-(4-(4-chlorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



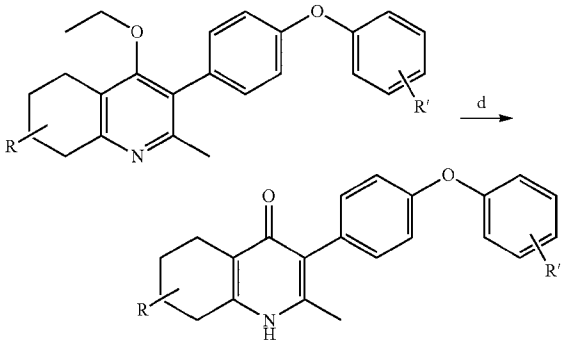
The title compound was synthesised from 3-(4-(4-chlorophenoxy)phenyl)-4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinoline (20 mg, 0.5 mmol) according to general procedure G. The title compound was isolated as colourless solid (18 mg, 0.05 mmol, 95%). ^{1}H NMR (500 MHz, DMSO) δ 10.99 (s, 1H), 7.44 (d, J=8.9 Hz, 2H), 7.18 (d, J=8.6 Hz, 2H), 7.05 (d, J=8.9 Hz, 2H), 7.04 (d, J=8.6 Hz, 2H), 2.56 (t, J=5.3 Hz, 2H), 2.29 (t, J=5.3 Hz, 2H), 2.07 (s, 3H), 1.78-1.55 (m, 4H); M/Z (ESI+); 366.13 (Found MH^+ 366.1262, $C_{22}H_{21}ClNO_2$ requires 366.1253).



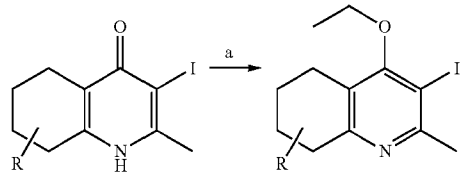
-continued



R = H, 5-Me, 6-Me, 7-Me, 6-CF_3, 7-CF_3, 7-Et R′ = 3-F, 3-Cl, 3-CF_3, 3-CO_2H, 4-F, 4-Cl, 4-CF_3, 4-OCF_3, 4-CO_2H, 3 & 4-Cl, 3-Cl, 4-F, 3 & 5-Cl

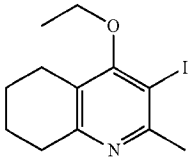


a) Etl, K_2CO_3, DMF, 80° C., b) 4-Hydroxyphenyl boronic acid, $Pd(PPH_3)_4$, Na_2CO_3, DMF, 80° C., c) Phenyl boronic acid, $Cu(OAc)_2$, Pyridine, TEA, DCM, d) HBr (40% aq), Acetic acid, 120° C.

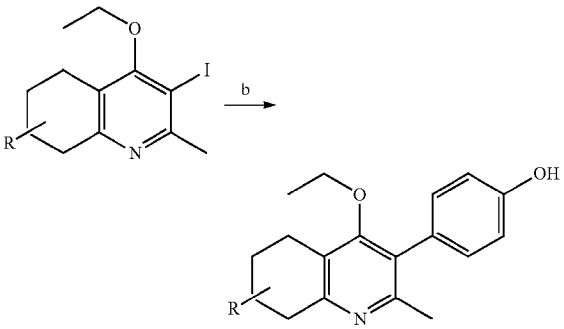


a) Etl, K_2CO_3, DMF, 80° C.

4-ethoxy-3-iodo-2-methyl-5,6,7,8-tetrahydroquinoline

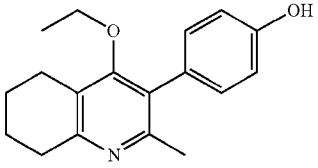


A suspension of 4(1H), 3-iodo-2-methyl, 5,6,7,8-tetrahydroquinolinone (2.20 g, 7.61 mmol) and Potassium carbonate (2.10 g, 15.2 mmol) in DMF (20.0 mL) was heated to 50° C. and stirred for 45 minutes. The reaction mixture was removed from the heat and ethyl iodide (0.89 mL, 11.4 mmol) was added dropwise. The reaction mixture was then heated to 50° C. and stirred for a further 18 hours. Formation of a yellow emulsion was observed. The reaction mixture was then quenched with water (40.0 mL). The organic phase was extracted using the polar extraction technique (ethyl acetate, 3×40.0 mL), and the resulting organic layers were combined and dried over $MgSO_4$ and concentrated in vacuo to afford the title compound as an orange oil. (2.00 g, 6.32 mmol, 83%). δ H NMR (500 MHz, Chloroform-d); δ 3.96 (q, J=7.0 Hz, 2H), 2.84 (t, J=6.3 Hz, 2H), 2.74 (t, J=6.3 Hz, 2H), 2.71 (s, 3H), 1.89-1.82 (m, 2H), 1.78-1.72 (m, 2H), 1.49 (t, J=7.0 Hz, 3H); M/Z (ESI+); 318.04 (Found MH^+, 318.0350, $C_{12}H_{17}INO$ requires 318.0349).



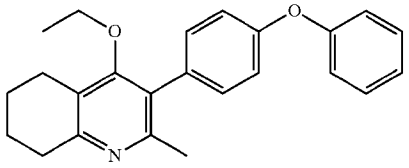
b) 4-Hydroxyphenyl boronic acid, $Pd(PPH_3)_4$, Na_2CO_3, DMF, 80° C.,

4-(4-Ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)phenol

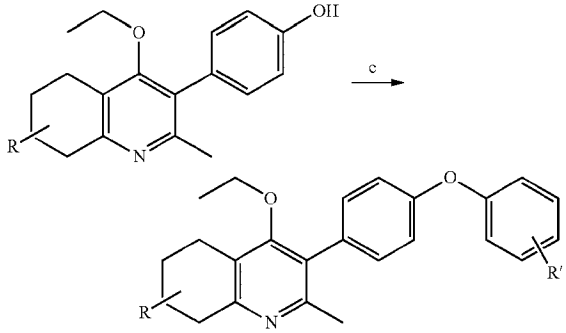


To a nitrogen flushed flask charged with 4-ethoxy-3-iodo-2-methyl-5,6,7,8-tetrahydroquinoline (400 mg, 1.26 mmol), 4 hydroxybenzene boronic acid (260 mg, 1.89 mmol) and palladium tetra(triphenylphosphine) (73 mg, 0.06 mmol) was added degassed DMF (10 mL). Potassium carbonate (aq) (3 mL, 2 M) was added and the reaction mixture brought up to 80° C. and stirred for 3 hours. The reaction mixture was then cooled to room temperature and diluted with water (10 mL). The organic phase was then extracted using ethyl acetate (3×20 mL). The organic phases were combined and washed with water (3×20 mL) and then dried with brine (1×10 mL) and $MgSO_4$, before concentration in vacuo. The resulting reddish brown solid was then recrystallized in ethyl acetate. To yield the title compound as a colourless solid (220 mg, 0.78 mmol, 61%). ^1H NMR (500 MHz, MeOD) δ 7.07 (d, J=8.6 Hz, 2H), 6.86 (d, J=8.6 Hz, 2H), 3.51 (q, J=7.0 Hz, 2H), 2.83 (t, J=6.3 Hz, 2H), 2.72 (t, J=6.1 Hz, 2H), 2.23 (s, 3H), 1.95-1.72 (m, 4H), 1.00 (t, J=7.0 Hz, 3H); M/Z (ESI+); 284.17 (Found MH^+ 284.1664, $C_{18}H_{21}NO_2$ requires 284.1651).

4-ethoxy-2-methyl-3-(4-phenoxyphenyl)-5,6,7,8-tetrahydroquinoline



A solution of 4-ethoxy, 3-iodo, 2-methyl, 5,6,7,8-tetrahydro, quinolone (1.00 g, 3.16 mmol), 4-phenoxy phenyl boronic acid (1.01 g, 4.73 mmol), Palladium (II) tetra(tri-phenyl-phosphine) (0.18 g, 0.16 mmol) and dipotassium carbonate (2.00 M, 6.40 mL) dissolved in degassed DMF (20.0 mL) was heated to 85° C. and stirred for 12 hours. Observed colour change from yellow to black. The reaction mixture was allowed to cool to room temperature before dilution with ethyl acetate (15.0 mL). The organic layer was extracted using polar extraction technique, before being collected and dried over $MgSO_4$. The solution was then concentration in vacuo to afford the title compound as colourless fine needles (0.45 g, 1.25 mmol, 40%). δ H NMR (126 MHz, Chloroform-d); δ 7.35 (t, J=7.6 Hz, 2H), 7.23 (d, J=8.5 Hz, 2H), 7.11 (t, J=7.6 Hz, 1H), 7.05 (d, 4H) 3.42 (q, J=7.0 Hz, 2H), 2.82 (t, J=6.4 Hz, 2H), 2.62 (t, J=6.3 Hz, 2H), 2.23 (s, 3H), 1.80-1.74 (m, 2H), 1.73-1.67 (m, 2H), 1.49 (t, J=7.0 Hz, 3H); M/Z (ESI+); 360.20 (Found MH^+, 360.1963, $C_{24}H_{26}NO_2$ requires 360.1958).



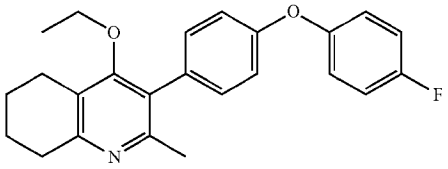
c) Phenyl boronic acid, $Cu(OAc)_2$, Pyridine, TEA, DCM

General Method C

Copper (II) acetate (1 equiv.), triethylamine (5 equiv.), and pyridine (5 equiv.) was added to a solution of the boronic acid (1.5 equiv.) and phenol (1 equiv.) in dichloromethane (10 mL $mmol^{-1}$) over heat-activated 4 Å molecular sieves. The reaction mixture was stirred over 16 hours at room temperature. The reaction mixture was quenched with HCl (0.5 M, 20 mL $mmol^{-1}$) and filtered through a pad of Celite, followed by repeated washing with water (10 mL $mmol^{-1}$). The organic layer was extracted with brine, dried

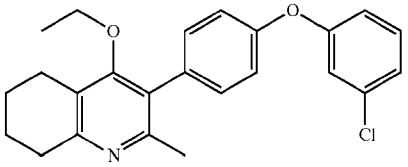
over magnesium sulphate, and concentrated in vacuo. Purification by silica gel chromatography (ethyl acetate/hexane) afforded the title compound.

4-Ethoxy-3-(4-(4-fluorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinoline



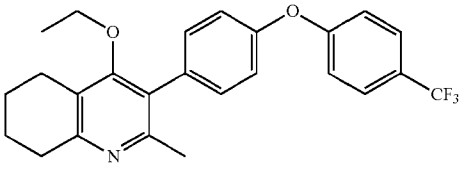
The title compound was synthesised from 4-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)phenol (60 mg, 0.21 mmol) and 4-fluorobenzene boronic acid (45 mg, 0.31 mmol), according to general procedure C as pink micro crystals (70%, 55 mg, 0.15 mmol). ^{1}H NMR (500 MHz, Chloroform-d) δ 7.14 (d, J=8.6 Hz, 2H), 6.97 (m, 6H), 3.44 (q, J=7.0 Hz, 2H), 2.87 (t, J=6.2 Hz, 2H), 2.64 (t, J=6.1 Hz, 2H), 2.26 (s, 3H), 1.78 (m, 4H), 0.97 (t, J=7.0 Hz, 3H); M/Z (ESI+); 378.19 (Found MH$^+$ 378.1877, $C_{24}H_{24}FNO_2$ requires).

3-(4-(3-Chlorophenoxy)phenyl)-4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinoline



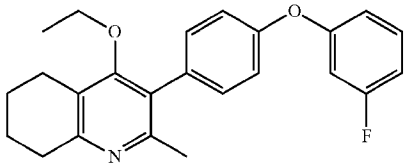
The title compound was synthesised from 4-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)phenol (250 mg, 0.88 mmol) and 3-chlorobenzene boronic acid (205 mg, 1.32 mmol), according to general procedure C as a viscous orange oil (78%, 270 mg, 0.68 mmol). 1H NMR (500 MHz, $CDCl_3$) δ 7.28-7.22 (m, 1H), 7.20 (d, J=8.3 Hz, 2H), 7.02 (d, J=8.3 Hz, 2H), 6.81-6.72 (m, 2H), 6.67 (dd, J=10.2, 2.1 Hz, 1H), 3.44 (q, J=7.0 Hz, 2H), 2.86 (t, J=6.2 Hz, 2H), 2.65 (t, J=6.1 Hz, 2H), 2.26 (s, 3H), 1.82 (m, 2H), 1.77-1.69 (m, 2H), 0.98 (t, J=7.0 Hz, 3H); M/Z (ESI+); 394.16 (Found MH$^+$; 394.1588, $C_{24}H_{24}ClNO_2$ requires 394.1574).

4-Ethoxy-2-methyl-3-(4-(4-(trifluoromethyl)phenoxy)phenyl)-5,6,7,8-tetrahydroquinoline



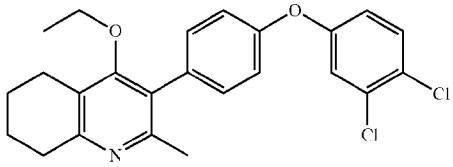
The title compound was synthesised from 4-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)phenol (60 mg, 0.21 mmol) and 4-trifluoromethylbenzene boronic acid (62 mg, 0.29 mmol), according to general procedure C as a viscous orange oil (55%, 47 mg, 0.11 mmol). 1H NMR (500 MHz, $CDCl_3$) δ 7.61 (d, J=8.7 Hz, 2H), 7.28 (d, J=8.5 Hz, 2H), 7.15-7.06 (m, 5H), 3.54 (q, J=7.0 Hz, 2H), 2.98 (t, J=6.3 Hz, 2H), 2.72 (t, J=6.2 Hz, 2H), 2.37 (s, 3H), 1.94-1.85 (m, 2H), 1.84-1.77 (m, 2H), 1.06 (t, J=7.0 Hz, 3H), M/Z (ESI+); 428.18 (Found MH$^+$ 428.1842, $C_{25}H_{24}F_3NO_2$ requires 428.1832).

4-Ethoxy-3-(4-(3-fluorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinoline



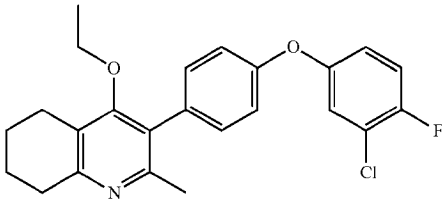
The title compound was synthesised from 4-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)phenol (250 mg, 0.88 mmol) and 3-fluorobenzene boronic acid (184 mg, 1.32 mmol), according to general procedure G as a yellow oil (57%, 190 mg, 0.50 mmol). ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.25 (dd, J=15.0, 8.2 Hz, 1H), 7.18 (d, J=8.6 Hz, 2H), 7.04 (d, J=8.6 Hz, 2H), 6.81-6.75 (m, 2H), 6.69 (dt, J=10.1, 2.3 Hz, 1H), 3.49 (q, J=7.0 Hz, 2H), 3.04 (m, 2H), 2.65 (t, J=6.2 Hz, 2H), 2.39 (s, 3H), 1.87-1.79 (m, 2H), 1.79-1.70 (m, 2H), 1.02 (t, J=7.0 Hz, 3H); M/Z (ESI+); 378.19 (Found MH$^+$ 378.1877, $C_{24}H_{24}FNO_2$ requires).

4-Ethoxy-3-(4-(3,4-dichlorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinoline



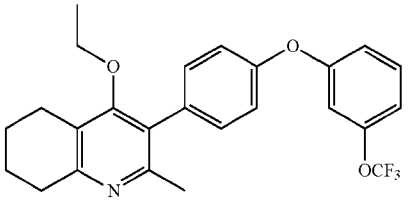
The title compound was synthesised from 4-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)phenol (100 mg, 0.34 mmol) and 3,4-dichlorobenzene boronic acid (93 mg, 0.51 mmol), according to general procedure C as an orange oil (49%, 70 mg, 0.16 mmol). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.34 (d, J=8.8 Hz, 1H), 7.20 (dd, J=7.1, 1.6 Hz, 2H), 7.06 (d, J=2.8 Hz, 1H), 7.03-6.97 (m, 2H), 6.85 (dd, J=8.8, 2.8 Hz, 1H), 3.46 (q, J=7.0 Hz, 2H), 2.91 (t, J=6.2 Hz, 2H), 2.65 (t, J=6.1 Hz, 2H), 2.29 (s, 3H), 1.90-1.62 (m, 4H), 0.99 (t, J=7.0 Hz, 3H); M/Z (ESI+); 428.12 (Found MH$^+$; 428.1202, $C_{24}H_{23}Cl_2NO_2$ requires 428.1184).

4-Ethoxy-3-(4-(3-chloro-4-fluorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinoline



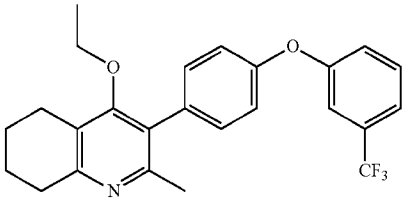
The title compound was synthesised from 4-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)phenol (250 mg, 0.88 mmol) and 3-chloro-4-fluorophenylboronic acid (230 mg, 1.32 mmol), according to general procedure C. The title compound was collected as a pale orange solid (104 mg, 0.25 mmol, 29%). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.26 (d, J=8.2 Hz, 2H), 7.17-7.11 (t, J=8.7 Hz, 1H), 7.09 (dd, J=6.1, 2.9 Hz, 1H), 7.04 (d, J=8.3 Hz, 2H), 6.97-6.91 (m, 1H), 3.50 (dd, J=14.0, 7.0 Hz, 1H), 2.91 (t, J=6.2 Hz, 2H), 2.72 (t, J=5.9 Hz, 2H), 2.31 (s, 3H), 1.95-1.84 (m, 2H), 1.81 (m, 2H), 1.04 (t, J=7.0 Hz, 3H); M/Z (ESI); 412.1489, $C_{24}H_{23}ClFNO_2$ requires 411.1401.

4-Ethoxy-3-(4-(3-trifluoromethoxyphenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinoline



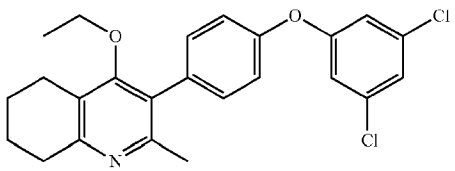
The title compound was synthesised from 4-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)phenol (200 mg, 0.71 mmol) and 3-trifluoromethoxyphenylboronic acid (218 mg, 1.06 mmol) according to general procedure C. The title compound was collected as a purple/brown oil (132 mg, 0.32 mmol, 42%); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.29 (t, J=8.3 Hz, 1H), 7.21 (d, J=8.3 Hz, 2H), 7.03 (d, J=8.2 Hz, 2H), 6.90 (d, J=8.2 Hz, 2H), 6.81 (s, 1H), 3.44 (dd, J=14.0, 7.0 Hz, 2H), 2.84 (t, J=6.2 Hz, 2H), 2.65 (t, J=6.0 Hz, 2H), 2.25 (s, 3H), 1.86-1.78 (m, 2H), 1.74 (dd, J=10.2, 4.6 Hz, 2H), 0.97 (t, J=7.0 Hz, 3H); M/Z (ESI); 444.1786, $C_{25}H_{25}F_3NO_3$ requires 444.1781.

4-Ethoxy-3-(4-(3-trifluoromethylphenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinoline

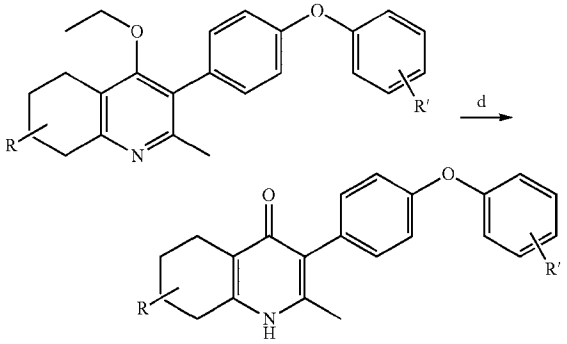


The title compound was synthesised from 4-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)phenol (300 mg, 1.06 mmol) and 3-trifluoromethylphenyl boronic acid (302 mg, 1.59 mmol) according to general procedure C. The title compound was collected as a yellow oil (198 mg, 0.46 mmol, 44%). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.44-7.38 (m, 1H), 7.32 (d, J=7.8 Hz, 1H), 7.17 (s, 2H), 7.12 (d, J=8.3 Hz, 2H), 7.01 (d, J=8.4 Hz, 2H), 3.45 (q, J=7.0 Hz, 2H), 2.85 (t, J=6.0 Hz, 2H), 2.62 (t, J=5.7 Hz, 2H), 2.25 (s, 3H), 1.73 (m, 4H), 1.00 (t, J=7.0 Hz, 3H); M/Z (ESI); 427.1852, $C_{25}H_{24}F_3NO_2$ requires 427.1759.

3-[4-(3,5-dichlorophenoxy)phenyl]-4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinoline



The title compound was synthesised from 4-(4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinolin-3-yl)phenol (250 mg, 0.88 mmol) and 3,5-dichlorophenylboronic acid (278 mg, 1.32 mmol according to general procedure C. The title compound was collected as a yellow solid (50 mg, 0.12 mmol, 13%). δ ^{1}H NMR (500 MHz, Chloroform-d) δ 7.34-7.32 (m, 2H), 7.13-7.11 (m, 3H), 6.93 (d, J=1.8 Hz, 2H), 3.54 (q, J=7.0 Hz, 2H), 2.94 (t, J=6.1 Hz, 2H), 2.75 (t, J=6.3 Hz), 2.39 (s, 3H), 1.95-1.81 (m, 4H), 1.08 (t, J=7.0 Hz, 3H); M/Z (ESI); 428.1194, $C_{24}H_{24}Cl_2NO_2$ requires 428.1179.

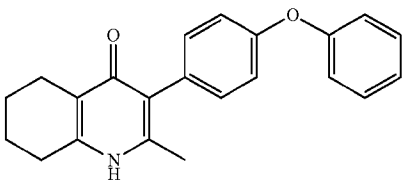


d) HBr (40% aq), Acetic acid, 120° C.

1.4 General Method D

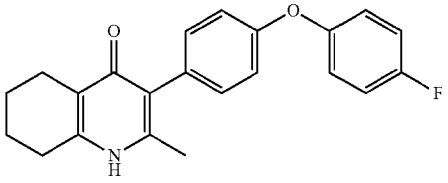
To a solution of the 4-ethoxy-3-(diaryl ether)-hydroxyquinoline (1 equiv.) in acetic acid (2 mL $mmol^{-1}$) was added hydrogen bromide (>48% w/v (aq)) (1 mL $mmol^{-1}$). The reaction mixture was then heated to 90° C. and left to reflux for 72 hours. The reaction mixture was neutralised with sodium hydroxide (2 M, 30.0 mL) and precipitate formed. The reaction mixture was then filtered to afford the title compound and purified.

Formation of 2-methyl-3-(4-phenoxyphenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one



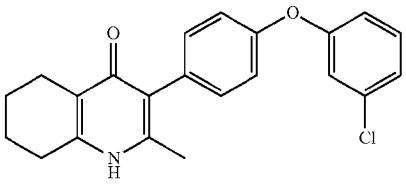
The title compound was synthesised from 4-ethoxy,3-(4-phenoxy, benzene),2-methy,5,6,7,8-hydroquinolone (0.43 g, 1.20 mmol) following general procedure D to afford the title compound as colourless microcrystals (0.39 g, 1.20 mmol, 99%). ^{1}H NMR (500 MHz, DMSO-d_6); δ 7.38 (t, J=7.9 Hz, 2H, H-3" & 5"), 7.17 (d, J=8.5 Hz, 2H), 7.14 (t, J=7.4 Hz, 1H), 7.05 (d, J=7.9 Hz, 2H), 6.98 (d, J=8.5 Hz, 2H), 2.56 (t, J=5.9 Hz, 2H), 2.30 (t, J=6.1 Hz, 2H), 2.08 (s, 3H), 1.75-1.68 (m, 2H), 1.68-1.62 (m, 2HM/Z (ESI+); 332.17 (Found MH$^+$, 332.1673, $C_{22}H_{21}NO_2$ requires 332.1650).

3-(4-(4-Fluorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



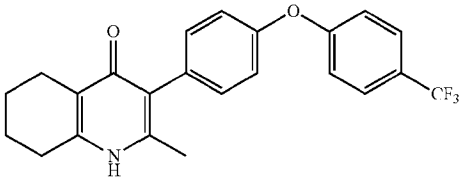
The title compound was synthesised from 4-ethoxy-3-(4-(4-fluorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinoline (45 mg, 0.12 mmol) according to general procedure D. The title compound was isolated as colourless solid (25 mg, 0.07 mmol, 60%). ^{1}H NMR (500 MHz, DMSO) δ 7.25 (t, J=8.7 Hz, 2H), 7.17 (d, J=8.5 Hz, 2H), 7.11 (dd, J=8.9, 4.5 Hz, 2H), 6.96 (d, J=8.5 Hz, 2H), 2.59-2.53 (m, 2H), 2.30 (t, J=5.2 Hz, 2H), 2.08 (s, 3H), 1.72 (m, 2H), 1.66 (m, 2H); M/Z (ESI+); 350.16 (Found MH$^+$ 350.1562, $C_{22}H_{20}FNO_2$ requires 350.1550).

3-(4-(3-Chlorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



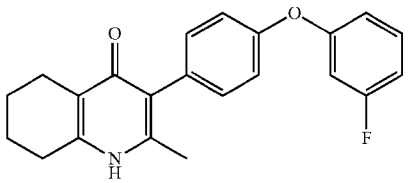
The title compound was synthesised from 3-(4-(3-chlorophenoxy)phenyl)-4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinoline (200 mg, 0.51 mmol) according to general procedure D. The title compound was isolated as colourless solid (120 mg, 0.33 mmol, 66%). ^{1}H NMR (500 MHz, DMSO) δ 11.71 (s, 1H), 7.44 (t, J=8.2 Hz, 1H), 7.25 (d, J=8.6 Hz, 2H), 7.22 (ddd, J=8.0, 1.9, 0.8 Hz, 1H), 7.10 (t, J=2.0 Hz, 1H), 7.09 (d, J=8.6 Hz, 2H), 7.03 (ddd, J=8.2, 2.3, 0.6, 1H), 2.65 (t, J=5.9 Hz, 2H), 2.39 (t, J=5.2 Hz, 2H), 2.14 (s, 3H), 1.84-1.59 (m, 4H); M/Z (ESI+); 366.13 (Found MH$^+$ 366.1262, $C_{22}H_{20}ClNO_2$ requires 366.1255).

2-Methyl-3-(4-(4-(trifluoromethyl)phenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one



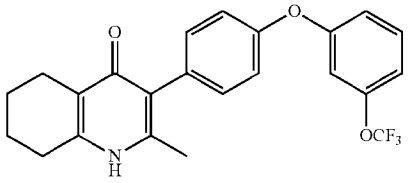
The title compound was synthesised from 4-ethoxy-2-methyl-3-(4-(4-(trifluoromethyl)phenoxy)phenyl)-5,6,7,8-tetrahydroquinoline (47 mg, 0.12 mmol) according to general procedure D. The title compound was isolated as colourless solid (25 mg, 0.07 mmol, 60%). ^{1}H NMR (500 MHz, DMSO) δ 11.14 (s, 1H), 7.77 (d, J=8.6 Hz, 2H), 7.26 (d, J=8.6 Hz, 2H), 7.19 (d, J=8.6 Hz, 2H), 7.12 (d, J=8.6 Hz, 2H), 2.59 (t, J=5.8 Hz, 2H), 2.33 (t, J=6.4 Hz, 2H), 2.12 (s, 3H), 1.73 (m, 2H), 1.70-1.64 (m, 2H); M/Z (ESI+); 400.15 (Found MH$^+$; 400.1528, $C_{23}H_{20}F_3NO_2$ requires 400.1519).

3-(4-(3-Fluorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



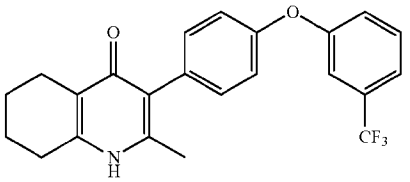
The title compound was synthesised from 4-ethoxy-3-(4-(3-fluorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinoline (170 mg, 0.45 mmol) according to general procedure D. The title compound was isolated as colourless solid (110 mg, 0.31 mmol, 70%). ^{1}H NMR (500 MHz, DMSO) δ 7.47 (dt, J=8.6, 7.1 Hz, 1H), 7.31 (d, J=8.7 Hz, 2H), 7.17 (d, J=8.7 Hz, 2H), 7.02 (tdd, J=8.5, 2.3, 0.7 Hz, 1H), 6.97-6.88 (m, 2H), 2.83 (t, J=6.1 Hz, 2H), 2.54 (t, J=5.8 Hz, 2H), 2.25 (s, 3H), 1.87-1.69 (m, 4H); M/Z (ESI+); 350.16 (Found MH$^+$; 350.1569, $C_{22}H_{21}FNO_2$ requires 350.1556).

3-(4-(3,4-Dichlorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



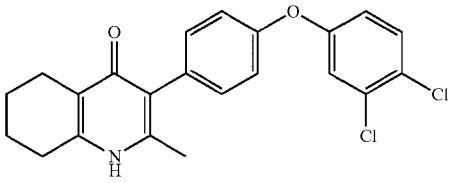
The title compound was synthesised from 4-Ethoxy-3-(4-(3-trifluoromethoxyphenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinoline (100 mg, 0.23 mmol) according to general procedure D. The title compound was isolated as colourless solid (72 mg, 0.16 mmol, 76%). ^{1}H NMR (501 MHz, DMSO) δ 11.27 (s, 1H, NH), 7.51 (t, J=8.3 Hz, 1H), 7.22 (d, J=8.5 Hz, 2H), 7.12 (d, J=7.6 Hz, 1H), 7.07 (d, J=8.5 Hz, 2H), 7.03 (dd, J=8.6, 1.7 Hz, 1H), 7.00 (s, 1H), 2.58 (t, J=5.2 Hz, 2H), 2.32 (t, J=6.2 Hz, 2H), 2.09 (s, 3H), 1.76-1.69 (m, 2H), 1.69-1.61 (m, 2H); M/Z (ESI+); 416.15 (Found MH$^+$; 416.1469, $C_{23}H_{21}F_3NO_3$ requires 416.1468).

3-(4-(3,4-Dichlorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



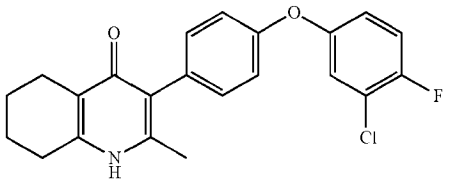
The title compound was synthesised from 4-Ethoxy-3-(4-(3-trifluoromethylphenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinoline (193 mg, 0.45 mmol) according to general procedure D. The title compound was isolated as colourless solid (174 mg, 0.44 mmol, 96%). ^{1}H NMR (501 MHz, DMSO) δ 11.15 (s, 1H), 7.63 (t, J=8.6 Hz, 1H), 7.48 (d, J=8.2 Hz, 1H), 7.30 (s, 2H), 7.22 (d, J=8.4 Hz, 2H), 7.07 (d, J=8.4 Hz, 2H), 2.56 (t, J=3.3 Hz, 2H), 2.30 (t, J=5.3 Hz, 2H), 2.08 (s, 3H), 1.71 (dd, J=6.0, 4.6 Hz, 2H), 1.68-1.60 (m, 2H); M/Z (ESI+); 400.15 (Found MH$^+$; 400.1519 requires $C_{23}H_{21}F_3NO_2$ requires 400.1519).

3-(4-(3,4-Dichlorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one



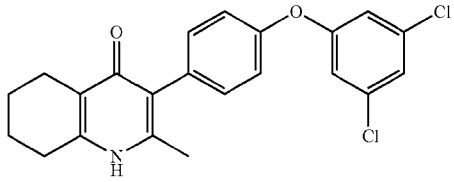
The title compound was synthesised from 4-ethoxy-3-(4-(3,4-dichloro phenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinoline (60 mg, 0.15 mmol) according to general procedure D. The title compound was isolated as colourless solid (35 mg, 0.08 mmol, 59%). ^{1}H NMR (500 MHz, DMSO) δ 10.95 (s, 1H), 7.66 (d, J=8.5 Hz, 1H), 7.31 (s, 1H), 7.23 (d, J=8.2 Hz, 2H), 7.08 (d, J=7.6 Hz, 2H), 7.04 (d, J=8.9 Hz, 1H), 2.55 (m, 2H), 2.30 (m, Hz, 2H), 2.09 (s, 3H), 1.72 (m, 2H), 1.66 (m, 2H); M/Z (ESI+); 400.09 (Found MH$^+$; 400.0881, $C_{22}H_{20}Cl_2NO_2$ requires 400.0866).

3-[4-(3-chloro-4-fluorophenoxy)phenyl]-2-methyl-5,6,7,8-tetrahydro-1H-quinolin-4-one

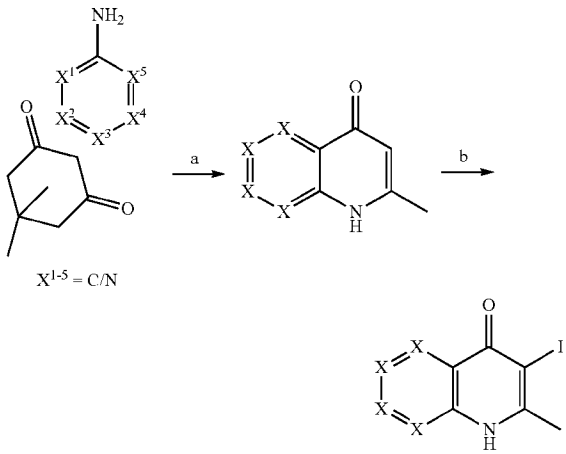


The title compound was synthesised from 4-Ethoxy-3-(4-(3-chloro-4-fluorophenoxy)phenyl)-2-methyl-5,6,7,8-tetrahydroquinoline (90 mg, 2.18 mmol) The title compound was isolated as a pale grey precipitate (55 mg, 0.14 mmol, 66%). 1H NMR (500 MHz, TFA) δ 7.39 (d, J 8.6 Hz, 2H), 7.32 (d, J 8.5 Hz, 2H). 7.27-7.24 (m, 2H), 7.12-7.09 (m, 1H), 3.10 (t, J 5.8 Hz, 2H), 2.91 (t, J 5.9 Hz, 2H), 2.54 (s, 3H), 2.15-2.03 (m, 4H); M/Z (ESI); 384.1167, ($C_{22}H_{20}ClFNO_2$ requires 384.1161).

3-[4-(3,5-dichlorophenoxy)phenyl]-2-methyl-5,6,7,8-tetrahydro-1H-quinolin-4(1H)-one



The title compound was synthesised from 3-[4-(3,5-dichlorophenoxy)phenyl]-4-ethoxy-2-methyl-5,6,7,8-tetrahydroquinoline (40 mg, 0.93 mmol) according to general procedure D. The title compound was isolated as colourless solid (31 mg, 0.08 mmol, 83%). δ ^{1}II NMR (500 MIz, TFA) δ 7.44 (d, J=8.5 Hz, 2H), 7.39 (d, J=8.0 Hz, 2H), 7.30 (s, 1H), 7.10 (s, 2H), 3.12 (t, J=5.5 Hz, 2H), 2.93 (t, J=5.5 Hz, 2H), 2.57 (s, 3H), 2.09-2.00 (m, 4H); M/Z (ESI); 400.0874, $C_{22}H_{20}Cl_2NO_2$ requires 400.0866.



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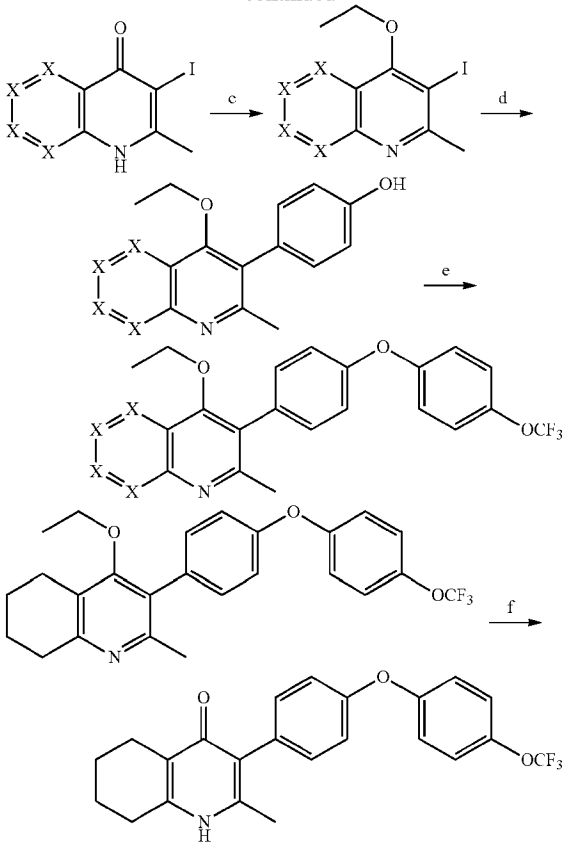
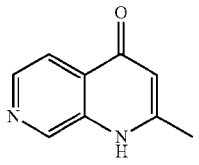


a) i) Meldrums acid, triethylorthoacetate, 110° C., ii) Aniline, 110° C., iii) Dowtherm A, 250° C., b) NIS, Acetonitrile, 80° C., c) EtI, K_2CO_3, DMF, 80° C., d) 4-Hydroxyphenyl boronic acid, $Pd(PPH_3)_4$, Na_2CO_3, DMF, 80° C., e) Phenyl boronic acid, $Cu(OAc)_2$, Pyridine, TEA, DCM, f) HBr (40% aq), Acetic acid, 120° C.

General Method A

2,2-Dimethyl-1,3-dioxane-4,6-dione (1.5 equiv.) was dissolved in trimethylorthoacetate (2 equiv.) and heated to 115° C. for 2 hrs. The reaction was cooled to allow the addition of the aniline (1 equiv.) before being heated to 115° C. for a further 2 hrs. The reaction mixture was then allowed to cool and was concentrated in vacuo, remaining solvent was washed off with cold methanol. The precipitate was then dissolved in minimum volume of Dowtherm A and refluxed at 250° C. for 1.5 hours. The reaction mixture was allowed to cool and the precipitate filtered followed by washing with hexane to afford the title compound.

2-Methyl-1,7-napthyrid-4(1H)-one

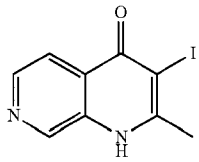


The title compound was synthesised using 3-amino pyridine (3.0 g, 32 mmol) following general procedure A. To give the title compound as colourless solid (489 mg, 3.1 mmol, 10%). 1H NMR (500 MHz, $CDCl_3$) δ 8.52 (s, 1H, H-8), 7.72 (d, J=8.0 Hz, 1H, H-5), 7.59-7.31 (m, 1H, H-6), 6.18 (s, 1H, H-3), 2.28 (s, 3H, Me). M/Z (ESI+); (Found MH^+; requires).

General Method B

2-Methyl-napthyrid-4(1H)-one (1 equiv.) and N-Iodosuccinamide (1.2 equiv.) were dissolved in acetonitrile (5 mL $mmol^{-1}$) and stirred and heated at 80° C. for 3 hours. The reaction mixture was then allowed to cool and the mixture filtered, the precipitate was then washed with water (15 mL) to afford the title compound as a colourless solid.

3-Iodo-2-methyl-1,7-napthyrid-4(1H)-one

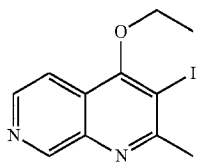


The title compound was synthesised using 2-Methyl-1,7-napthyrid-4(1H)-one (480 mg, 3.0 mmol) following general procedure B. To give the title compound as colourless solid (740 mg, 2.6 mmol, 86%). ^{1}II NMR (500 MIIz, $CDCl_3$) δ 8.36-7.91 (m. 1H), 7.49 (dd, J=8.4, 1.1 Hz, 1H), 7.18 (dd, J=8.6, 4.2 Hz, 1H), 2.27 (s, 3H). M/Z (ESI+); (Found MH^+; requires).

General Method C

A suspension of the 4(1H), 3-Iodo-2-methyl-napthyrid-4(1H)-one (1 equiv.) and potassium carbonate (2 equiv.) in DMF (20.0 mL) was heated to 50° C. and stirred for 45 minutes. The reaction mixture was removed from the heat and ethyl iodide (1.5 equiv.) was added dropwise. The reaction mixture was then heated and kept at 50° C. with stirring for a further 18 hrs. Formation of a yellow emulsion was observed. The reaction mixture was then quenched with water (40.0 mL). The organic phase was extracted using the polar extraction technique (ethyl acetate, 3×40.0 mL), and the resulting organic layers were combined and dried over $MgSO_4$ and concentrated in vacuo to afford the title compound.

4-Ethoxy-3-iodo-2-methyl-1,7-napthyridine



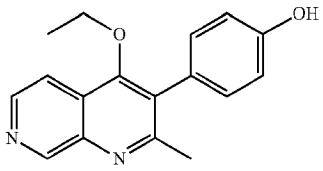
The title compound was synthesised using, 3-Iodo-2-methyl-napthyrid-4(1H)-one (720 mg, 2.5 mmol) following general procedure C. To give the title compound as brown gum (244 mg. 0.8 mmol, 33%). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.87 (dd, J=4.0, 1.5 Hz, 1H), 8.31 (dd, J=8.5, 1.6 Hz, 1H),

7.63 (dd, J=8.5, 4.1 Hz, 1H), 4.84 (q, J=7.0 Hz, 2H), 3.00 (s, 3H), 1.59 (t, J=7.0 Hz, 3); M/Z (ESI+); (Found MH^+; requires).

General Method F

To a nitrogen flushed flask charged with the 4-Ethoxy-3-iodo-2-methyl-napthyridine (400 mg, 1.26 mmol), 4-hydroxybenzene boronic acid (260 mg, 1.89 mmol) and palladium tetra(triphenylphosphine) (73 mg, 0.06 mmol) was added degassed DMF (10 mL). Potassium carbonate (3 mL, 2 $M_{(aq)}$) was added and the reaction mixture brought up to 80° C. and stirred for 3 hours. The reaction mixture was then cooled to room temperature and diluted with water (10 mL). The organic phase was then extracted using ethyl acetate (3×20 mL). The organic phases were combined and washed with water (3×20 mL) and then dried with brine (1×10 mL) and $MgSO_4$, before concentration in vacuo. The resulting solid was then recrystallized in ethyl acetate to afford the title compound.

4-Ethoxy-3-phenol-2-methyl-1,7-napthyridine

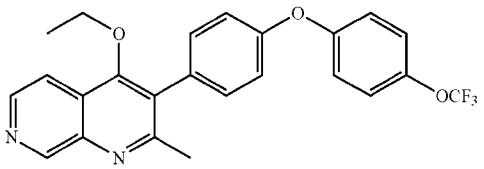


The title compound was synthesised using, 4-Ethoxy-3-iodo-2-methyl-1,7-napthyridine (230 mg, 0.73 mmol) following general procedure F. To give the title compound an orange powder (80 mg, 0.8 mmol, 28%). 1H NMR (400 MHz, MeOD) δ 8.78 (dd, J=4.1, 1.4 Hz, 1H), 8.24 (dd, J=8.6, 1.4 Hz, 1H), 7.64 (dd, J=8.6, 4.2 Hz, 1H), 7.08 (d, J=8.5 Hz, 2H), 6.84 (d, J=8.5 Hz, 2H), 4.12 (q, J=7.0 Hz, 2H), 2.39 (s, 3H), 1.05 (t, J=7.0 Hz, 3H); M/Z (ESI+); (Found MH^+; requires).

General Method G

Copper (II) acetate (1 equiv.), triethylamine (5 equiv.), and pyridine (5 equiv.) was added to a solution of the boronic acid (1.5 equiv.) and phenol (1 equiv.) in dichloromethane (10 mL $mmol^{-1}$) over heat-activated 4 Å molecular sieves. The reaction mixture was stirred over 16 hours at room temperature. The reaction mixture was quenched with HCl (0.5 M, 20 mL $mmol^{-1}$) and filtered through a pad of Celite, followed by repeated washing with water (10 mL $mmol^{-1}$). The organic layer was extracted with brine, dried over magnesium sulphate, and concentrated in vacuo. Purification by silica gel chromatography (ethyl acetate/hexane) afforded the title compound.

4-Ethyl-3(4-(4-trifluoromethoxyphenoxy)phenyl)-2-methyl-1,7-napthyridone

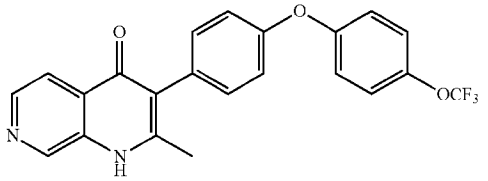


The title compound was synthesised using 4-Ethoxy-3-iodo-2-methyl-1,7-napthyridine (70 mg, 0.25 mmol) and 4-trifluoromethoxybenzenboronic acid (79 mg, 0.38 mmol) following general procedure G. To give the title compound as a red crystalline solid (34 mg, 0.08 mmol, 20%). 1H NMR (400 MHz, $CDCl_3$) δ 8.90 (dd, J=4.0, 1.3 Hz, 1H), 8.33 (dd, J=8.5, 1.3 Hz, 1H), 7.61 (dd, J=8.5, 4.1 Hz, 1H), 7.31 (d, J=8.5 Hz, 2H), 7.24 (d, J=8.7 Hz, 2H), 7.13 (d, J=8.7 Hz, 2H), 7.10 (d, J=9.1 Hz, 2H), 4.43 (q, J=7.0 Hz, 2H), 2.54 (s, 3H), 1.19 (t, J=7.0 Hz, 3H); M/Z (ESI+); (Found MH^+; requires).

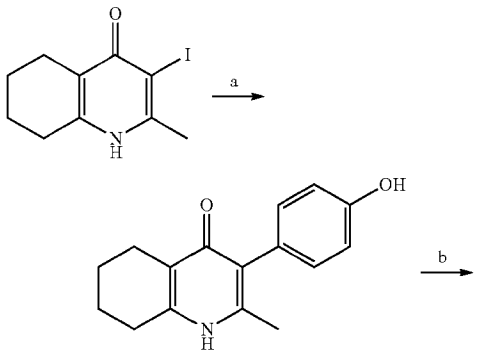
General Method J

To a solution of the 4-Ethyl-3(4-(4-trifluoromethoxyphenoxy)phenyl)-2-methylnapthyridone (1 equiv.) in acetic acid (2 mL $mmol^{-1}$) was added hydrogen bromide (>48% w/v (aq)) (1 mL $mmol^{-1}$). The reaction mixture was then heated to 90° C. and left to reflux for 72 hours. The reaction mixture was neutralised with sodium hydroxide (2 M, 30.0 mL) and precipitate formed. The reaction mixture was then filtered to afford the title compound.

3(4-(4-trifluoromethoxyphenoxy)phenyl)-2-methyl-1,7-napthyrid-4(1H)-one



The title compound was synthesised from 4-Ethyl-3(4-(4-trifluoromethoxyphenoxy)phenyl)-2-methyl-1,7-napthyridone (31 mg, 0.07 mmol). To give the title compound as a colourless solid (13 mg, 0.03 mmol, 45%). ^1H NMR (400 MHz, $CDCl_3$) δ 8.44 (d, J=3.6 Hz, 1H), 7.75 (d, J=8.2 Hz, 1H), 7.36 (dd, J=8.3, 4.1 Hz, 1H), 7.02 (d, J=8.6 Hz, 2H), 6.96 (d, J=8.5 Hz, 2H), 6.83 (d, J=9.2 Hz, 4H), 2.09 (s, 3H); M/Z (ESI+); 413.11 (Found MH^+; 413.1104 $C_{22}H_{15}F_3N_2O_3$ requires 413.1107).



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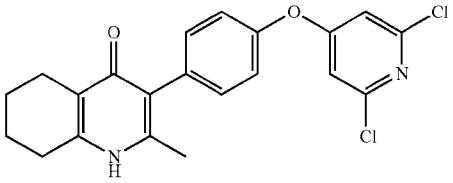
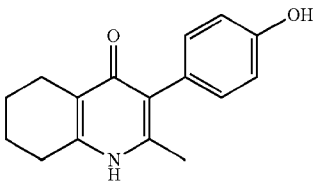


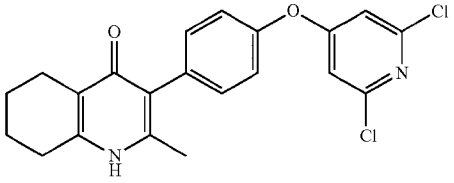
a) 4-Hydroxybenzene boronic acid, $Pd(PPH_3)_4$, Na_2CO_3 (aq), DMF 80° C., b) 2,4,6 trichloropyridine, K_2CO_3, DMF, 100° C.

3-phenol-2-methyl-5,6,7,8-tetrahydroquinolin-4 (1H)-one

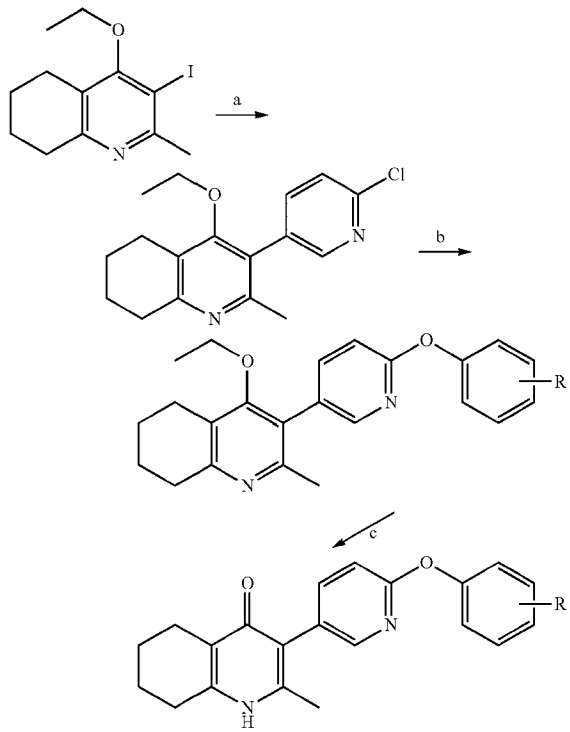


The title compound was synthesised from 4(1H)-3-iodo-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one (500 mg, 1.73 mmol) and 4-hydroxyphenylboronic acid (318 mg, 2.30 mmol) and palladium tetra(triphenylphosphine) (73 mg, 0.06 mmol) was added degassed DMF (10 mL). Potassium carbonate (aq) (3 mL, 2 M) was added and the reaction mixture brought up to 80° C. and stirred for 3 hours. The reaction mixture was then cooled to room temperature and diluted with water (10 mL). Organics were extracted with ethyl acetate (3×10 mL). The aqueous layer was neutralised using hydrochloric acid (2 M), causing the title compound, a grey precipitate to crash out which was collected by vacuum filtration (150 mg, 0.58 mmol, 44%). ^{1}H NMR (400 MHz, MeOD) δ 6.91 (d, J=8.6 Hz, 2H), 6.71 (d, J=8.6 Hz, 2H), 2.57 (t, J=6.0 Hz), 2.39 (t, J=5.6 Hz, 2H), 2.03 (s, 3H), 1.77-1.61 (m, 4H); M/Z (ESI); 255.1341 ($C_{16}H_{17}NO_2$ requires 255.1259).

3-{4-[(2,6-dichloropyridin-4-yl)oxy]phenyl}-2-methyl-5,6,7,8-tetrahydro-1H-quinolin-4-one

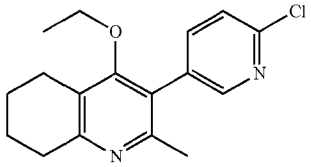


3-phenol-2-methyl-5,6,7,8-tetrahydroquinolin-4(1H)-one (120 mg, 0.47 mmol) and potassium carbonate (78 mg, 0.56 mmol) were dissolved in DMF (3 mL) and the reaction mixture was stirred for 15 mins. Following this, 2,4,6-Trichloropyridine (86 mg, 0.47 mmol) was added and the mixture was heated to 100° C. and stirred for 18 hours under an inert atmosphere. The reaction mixture was allowed to cool to room temperature and was diluted with water (5 ml). The resulting pale grey precipitate was collected by vacuum filtration, washed with water (5 ml) and dried (118 mg, 0.29 mmol, 63%). δ ^{1}H NMR (500 MHz, TFA) δ 7.66 (d, J 8.5 Hz, 2H), 7.56 (d, J 8.5 Hz, 2H), 7.44 (s, 2H), 3.10 (t, J 5.0 Hz, 2H), 2.90 (t, J 5.0 Hz, 2H), 2.55 (s, 3H), 2.10-2.08 (m, 4H); M/Z (ESI); 400.0826, ($C_{21}H_{18}Cl_2N_2O_2$ requires 400.0745).



a) 2-Chlorpyridine-5-boronic acid, $Pd(PPH_3)_4$, Na_2CO_3 (aq), DMF 80° C., b) Phenol, K_2CO_3, DMF, 100° C.

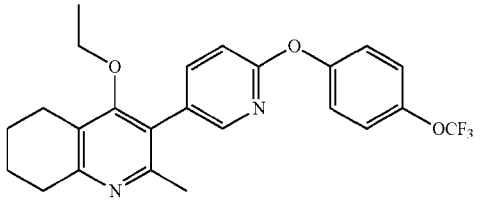
4-Ethoxy-3-(6-chloropyrdin-3-yl)-2-methyl-5,6,7,8-tetrahydroquinoline



To a nitrogen flushed flask charged with 4-ethoxy-3-iodo-2-methyl-5,6,7,8-tetrahydroquinoline (1.5 g, 4.7 mmol), 2-chloropyridine-5-boronic acid (1.12 g, 7.1 mmol) and palladium tetra(triphenylphosphine) (271 mg, 0.24 mmol) was added degassed DMF (20 mL). Potassium carbonate (3 mL, 2 $M_{(aq)}$) was added and the reaction mixture brought up to 80° C. and stirred for 3 hours. The reaction mixture was then cooled to room temperature and diluted with water (10 mL). The organic phase was then extracted using ethyl acetate (3×20 mL). The organic phases were combined and washed with water (3×20 mL) and then dried with brine (1×10 mL) and $MgSO_4$, before concentration in vacuo. The resulting residue was purified by column chromatography (Pet:EtOAc), to yield the title compound as a yellow plate-

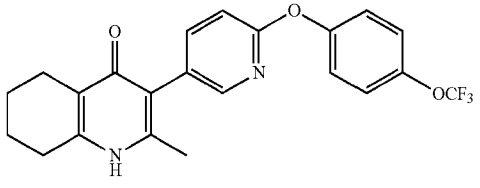
lets (330 mg, 1.09 mmol, 23%). HPLC; 2.17 min (100% ref area); ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.33 (d, J=2.4 Hz, 1H), 7.60 (dd, J=8.2, 2.4 Hz, 1H), 7.44-7.42 (d, J=8.2, 1H), 3.53 (q, J=7.0 Hz, 2H), 2.98 (t, J=6.2 Hz, 2H), 2.72 (t, J=6.2 Hz, 2H), 2.35 (s, 3H), 1.93-1.85 (m, 2H), 1.84-1.76 (m, 2H), 1.06 (t, J=7.0 Hz, 3H); M/Z (ESI+); 303.13 (Found MH^+; 303.1266, $C_{17}H_{19}ClN_2O$ requires 303.1259).

4-Ethoxy-3-(6-(4-trifluoromethoxyphenoxy)pyridin-3-yl)-2-methyl-5,6,7,8-tetrahydroquinoline

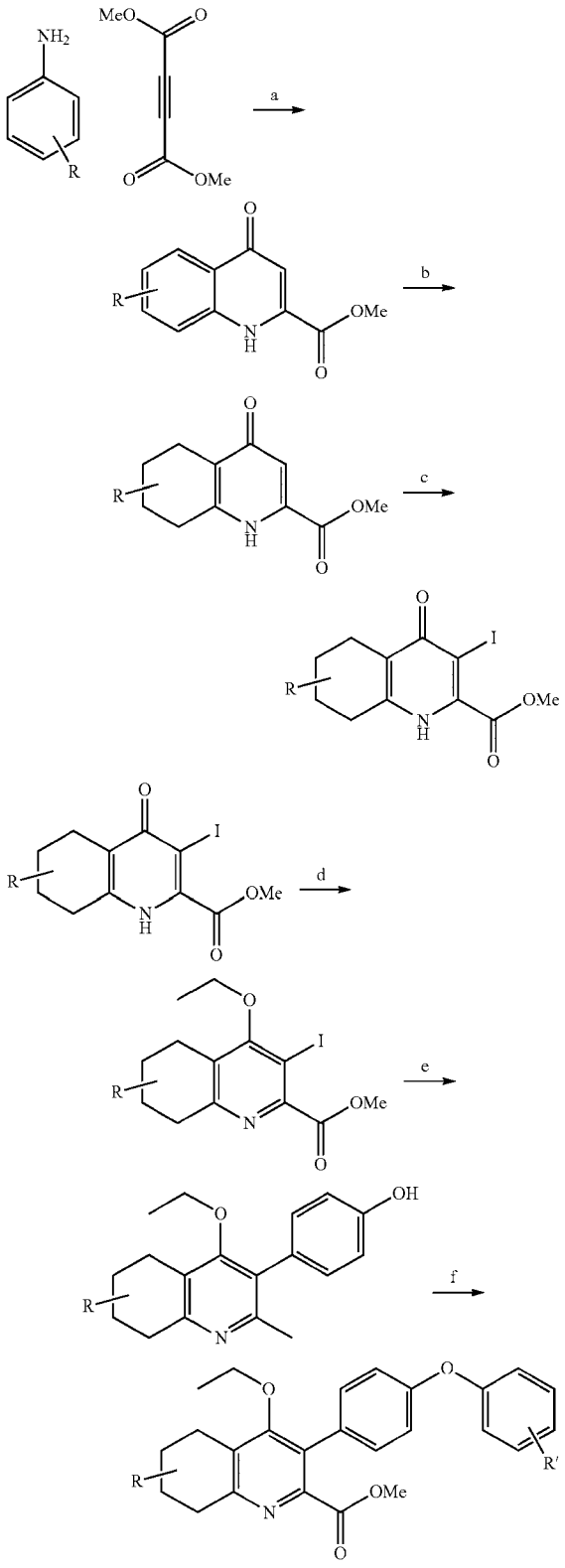


4-Ethoxy-3-(6-chloropyrdin-3-yl)-2-methyl-5,6,7,8-tetrahydroquinoline (250 mg, 0.82 mmol), 4-trifluoromethoxyphenol (178 mg, 1.0 mmol) and potassium carbonate (276 mg, 1.7 mmol) were dissolved in DMF and refluxed at 110° C. for 24 hrs. The reaction mixture was then cooled to room temperature and diluted with water (10 mL). The organic phase was then extracted using ethyl acetate (3×20 mL). The organic phases were combined and washed with water (3×20 mL) and then dried with brine (1×10 mL) and $MgSO_4$, before concentration in vacuo. The resulting residue was purified by reverse phase column chromatography (H_2O: acetonitrile), to yield the title compound as a yellow oil (75 mg, 0.17 mmol, 20%). 1H NMR (400 MHz, $CDCl_3$) δ 8.03 (d, J=2.2 Hz, 1H), 7.58 (dd, J=8.4, 2.2 Hz, 1H), 7.29-7.07 (m, 4H), 6.95 (d, J=8.4 Hz, 1H), 3.46 (q, J=7.0 Hz, 2H), 2.85 (t, J=6.3 Hz, 2H), 2.64 (t, J=6.1 Hz, 2H), 2.25 (s, 3H), 1.81 (dt, J=12.2, 6.3 Hz, 2H), 1.77-1.68 (m, 2H), 0.99 (t, J=7.0 Hz, 3H); M/Z (ESI+); 445.18 (Found MH^+; 445.1759, $C_{24}H_{23}F_3N_2O_3$ requires 445.1739).

3-(6-(4-Trifluoromethoxyphenoxy)pyridin-3-yl)-2-methyl-5,6,7,8-tetrahydroquinolin-(4)-one



The title compound was synthesised from 4-ethoxy-3-(6-(4-trifluoromethoxyphenoxy)pyridin-3-yl)-2-methyl-5,6,7,8-tetrahydroquinoline (70 mg, 0.15 mmol) according to general procedure J. The title compound was isolated as colourless solid (13 mg, 0.03 mmol, 20%). ^{1}H NMR (400 MHz, DMSO) δ 11.07 (s, 1H), 7.94 (d, J=2.2 Hz, 1H), 7.70 (dd, J=8.4, 2.4 Hz, 1H), 7.43 (d, J=8.7 Hz, 2H), 7.29 (d, J=9.0 Hz, 2H), 7.09 (d, J=8.4 Hz, 1H), 2.56 (t, J=5.1 Hz, 2H), 2.30 (t, J=5.9 Hz, H), 2.10 (s, 3H), 1.79-1.57 (m, 4H); M/Z (ESI+); 417.14 (Found MH^+; 417.1432, $C_{22}H_{19}F_3N_2O_3$ requires 417.1421).



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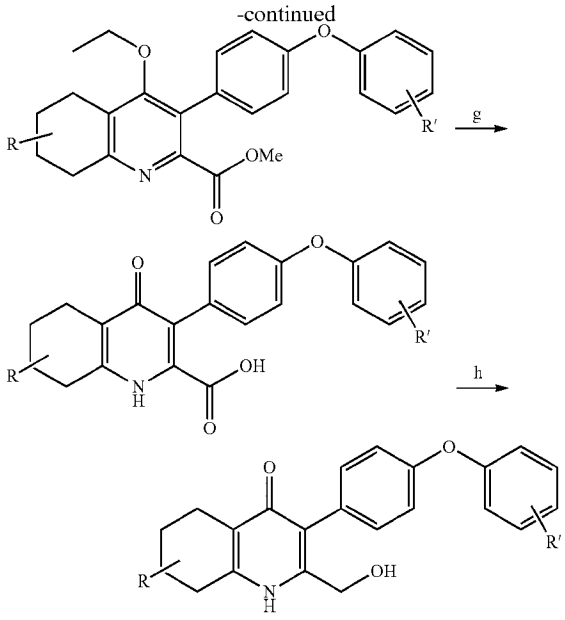
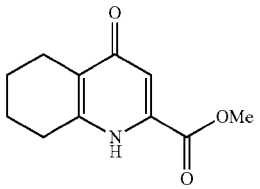


a) i) Ethanol, ii) Dowtherm A, 250° C., b) PtO_2, H_2, AcOH, c) NIS, Acetonitrile, 80° C., D) EtI, K_2CO_3, DMF, 80° C., e) 4-Hydroxyphenyl boronic acid, $Pd(PPH_3)_4$, DMF, 80° C., f) Phenyl boronic acid, $Cu(OAc)_2$, Pyridine, TEA, DCM, g) HBr (40% aq), Acetic acid, 120° C., h) i) MeOH, ii) DiBAI.

General Method B

The 4-hydroxylquinolone (1 equiv.) was dissolved in acetic acid (10.0 mL) under inert conditions. platinum dioxide (5% weight equiv.) was added and a hydrogen balloon was attached. The reaction was left to proceed for 12 hours. The resulting suspension was filtered through a pad of Celite and washed with ethyl acetate (10.0 mL). The filtrate was concentrated in vacuo to afford a yellow/brown oil. Purification by column chromatography (10% methanol in chloroform) afforded the title compound.

2-(Methoxycarboxylate)-5,6,7,8-tetrahydro quinolin-4(1H)-one

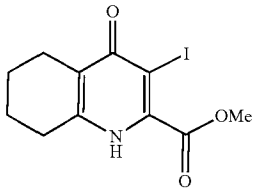


The title compound was synthesised following general procedure B from 2-(methoxycarboxylate)quinolin-4(1H)-one (500 mg, 2.5 mmol). The title compound was isolated as colourless solid (470 mg, 2.4 mmol, %). ^1H NMR (500 MHz, $CDCl_3$) δ 8.89 (s, 1H), 7.07 (s, 1H), 3.95 (s, 3H), 2.74 (t, J=6.1 Hz, 2H), 2.56 (t, J=6.2 Hz, 2H), 1.82 (dt, J=8.0, 6.1 Hz, 1H), 1.79-1.69 (m, 1H); M/Z (ESI+); 208.10 (Found MH^+; 208.0977, $C_{11}H_{13}NO_3$ requires 208.0974).

1.5 General Method C

The tetrahydroquinolin-4(1H)-one (1 equiv.) and N-Iodosuccinamide (1.2 equiv.) were dissolved in acetonitrile (5 mL $mmol^{-1}$) and stirred and heated at 80° C. for 3 hours. The reaction mixture was then allowed to cool and the mixture filtered, the precipitate was then washed with water (15 mL) to afford the title compound as a colourless solid

3-Iodo-2-(methoxycarboxylate)-5,6,7,8-tetrahydro quinolin-4(1H)-one

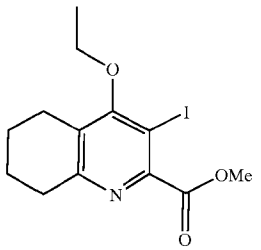


The title compound was synthesised following general procedure C from 2-(methoxycarboxylate)-5,6,7,8-tetrahydro quinolin-4(1H)-one (450 mg, 2.20 mmol). The title compound was isolated as colourless solid (612 mg, 1.8 mmol, 84%). ^1H NMR (400 MHz, DMSO) δ 11.93 (s, 1H), 3.91 (s, 3H), 2.58 (s, 2H), 2.34 (t, 2H), 1.90-1.45 (m, 4H); M/Z (ESI+); 333.99 (Found MH^+; 333.9935, $C_{11}H_{12}INO_3$ requires; 333.9935).

General Method D

A suspension of the 4(1H), 3-iodo-tetrahydroquinolinone (1 equiv.) and potassium carbonate (2 equiv.) in DMF (20.0 mL) was heated to 50° C. and stirred for 45 minutes. The reaction mixture was removed from the heat and ethyl iodide (1.5 equiv.) was added dropwise. The reaction mixture was then heated and kept at 50° C. with stirring for a further 18 hrs. Formation of a yellow emulsion was observed. The reaction mixture was then quenched with water (40.0 mL). The organic phase was extracted using the polar extraction technique (ethyl acetate, 3×40.0 mL), and the resulting organic layers were combined and dried over $MgSO_4$ and concentrated in vacuo to afford the title compound.

4-Ethoxy-3-iodo-2-(methoxycarboxylate)-5,6,7,8-tetrahydroquinoline



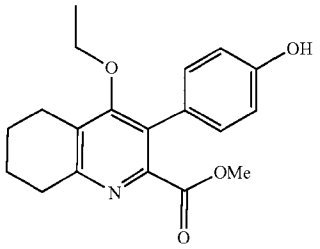
The title compound was synthesised following general procedure D from 3-iodo-2-(methoxycarboxylate)-5,6,7,8-tetrahydro quinolin-4(1H)-one (500 mg, 1.5 mmol). The title compound was isolated as colourless crystals (450 mg, 1.25 mmol, 83%). ^1H NMR (300 MHz, $CDCl_3$) δ 3.94 (q, J=7.0 Hz, 2H), 3.90 (s, 3H), 2.84 (t, J=6.4 Hz, 2H), 2.73 (t, J=6.2

Hz, 2H), 1.87-1.64 (m, 4H), 1.43 (t, J=7.0 Hz, 3H); M/Z (ESI+); 362.03 (Found MH^+; 362.0256, $C_{13}H_{16}INO_3$ requires 362.0248).

1.6 General Method E

To a nitrogen flushed flask charged with the 4-ethoxy-3-iodo-tetrahydroquinoline (400 mg, 1.26 mmol), 4-hydroxybenzene boronic acid (260 mg, 1.89 mmol) and palladium tetra(triphenylphosphine) (73 mg, 0.06 mmol) was added degassed DMF (10 mL). Potassium carbonate (3 mL, 2 $M_{(aq)}$) was added and the reaction mixture brought up to 80° C. and stirred for 3 hours. The reaction mixture was then cooled to room temperature and diluted with water (10 mL). The organic phase was then extracted using ethyl acetate (3×20 mL). The organic phases were combined and washed with water (3×20 mL) and then dried with brine (1×10 mL) and $MgSO_4$, before concentration in vacuo. The resulting solid was then recrystallized in ethyl acetate to afford the title compound.

4-(4-Ethoxy-(methoxycarboxylate)-5,6,7,8-tetrahydroquinolin-3-yl)phenol

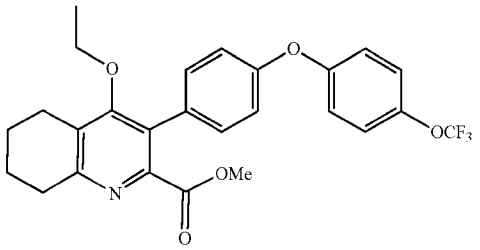


The title compound was synthesised following general procedure E from 4-ethoxy-3-iodo-2-(methoxycarboxylate)-5,6,7,8-tetrahydro quinoline (400 mg, 1.11 mmol). The title compound was isolated as a solid (200 mg, 0.61 mmol, 55%). ^1H NMR (500 MHz, $CDCl_3$) δ 7.11 (d, J=8.6 Hz, 2H), 6.77 (d, J=8.6 Hz, 2H), 3.60 (s, 3H), 3.43 (q, J=7.0 Hz, 2H), 2.91 (t, J=6.4 Hz, 2H), 2.71 (t, J=6.3 Hz, 2H), 1.82 (m, 2H), 1.78-1.69 (m, 2H), 0.98 (t, J=7.0 Hz, 3H); M/Z (ESI+); 328.16 (Found MH^+; 328.1551, $C_{19}H_{21}NO_4$ requires; 328.1543).

General Method F

Copper (II) acetate (1 equiv.), triethylamine (5 equiv.), and pyridine (5 equiv.) was added to a solution of the boronic acid (1.5 equiv.) and phenol (1 equiv.) in dichloromethane (10 mL $mmol^{-1}$) over heat-activated 4 Å molecular sieves. The reaction mixture was stirred over 16 hours at room temperature. The reaction mixture was quenched with HCl (0.5 M, 20 mL $mmol^{-1}$) and filtered through a pad of Celite, followed by repeated washing with water (10 mL $mmol^{-1}$). The organic layer was extracted with brine, dried over magnesium sulphate, and concentrated in vacuo. Purification by silica gel chromatography (ethyl acetate/hexane) afforded the title compound.

4-Ethoxy-3(4-(4-trifluoromethoxyphenoxy)phenyl)-2-(methoxycarboxylate)-5,6,7,8-tetrahydroquinoline

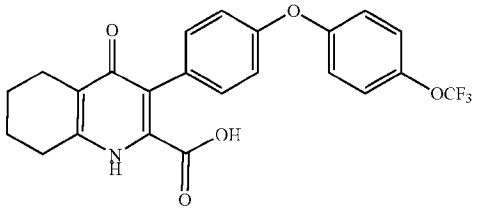


The title compound was synthesised following general procedure F from 4-(4-ethoxy-(methoxycarboxylate)-5,6,7,8-tetrahydroquinolin-3-yl)phenol (180 mg, 0.55 mmol). The title compound was isolated as a yellow oil (210 mg, 0.43 mmol, 78%). ^1H NMR (500 MHz, $CDCl_3$) δ 7.32 (d, J=8.6 Hz, 2H), 7.22 (d, J=8.6 Hz, 2H), 7.05 (dd, J=8.8, 3.0 Hz, 4H), 3.73 (s, 3H), 3.55 (q, J=7.0 Hz, 2H), 3.05 (t, J=4.7 Hz, 2H), 2.80 (t, J=6.2 Hz, 2H), 1.99-1.88 (m, 2H), 1.88-1.79 (m, 2H), 1.09 (t, J=7.0 Hz, 3H).

General Method G

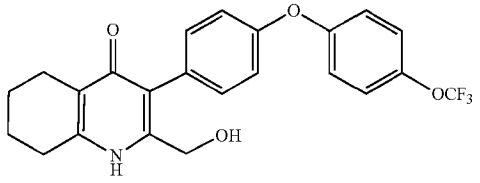
To a solution of the 4-ethoxy-3-(diaryl ether)-hydroxyquinoline (1 equiv.) in acetic acid (2 mL $mmol^{-1}$) was added hydrogen bromide (>48% w/v (aq)) (1 mL $mmol^{-1}$). The reaction mixture was then heated to 90° C. and left to reflux for 72 hours. The reaction mixture was neutralised with sodium hydroxide (2 M, 30.0 mL) and precipitate formed. The reaction mixture was then filtered to afford the title compound.

3(4-(4-Trifluoromethoxyphenoxy)phenyl)-2-(carboxylate)-5,6,7,8-tetrahydroquinolin-4(1H)-one

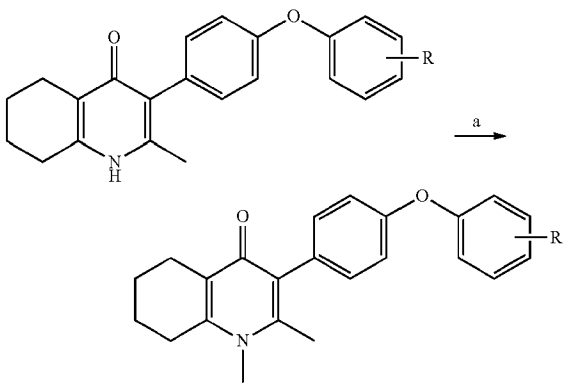


The title compound was synthesised following general procedure G from 4-ethoxy-3(4-(4-trifluoromethoxyphenoxy)phenyl)-2-(methoxycarboxylate)-5,6,7,8-tetrahydroquinoline (200 mg, 0.42 mmol). The title compound was isolated as a colourless solid (80 mg, 0.18 mmol, 43%). ^1H NMR (400 MHz, DMSO) δ 7.41 (d, J=8.8 Hz, 2H), 7.21 (d, J=8.3 Hz, 2H), 7.12 (d, J=8.8 Hz, 2H), 7.01 (d, J=8.3 Hz, 2H), 2.64 (t, J=5.6 Hz, 2H), 2.35 (t, J=5.6 Hz, 2H), 1.94-1.33 (m, 4H); M/Z (ESI+); 446.12 (Found MH^+; 446.1207, $C_{23}H_{18}F_3NO_5$ requires 446.1210).

3(4-(4-Trifluoromethoxyphenoxy)phenyl)-2-(methylhydroxy)-5,6,7,8-tetrahydroquinolin-4(1H)-one

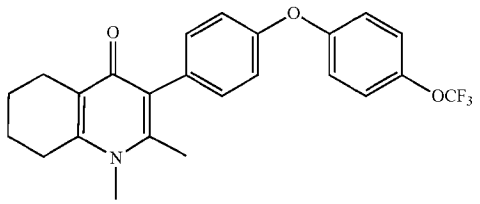


3(4-(4-trifluoromethoxyphenoxy)phenyl)-2-(carboxylate)-5,6,7,8-tetrahydroquinolin-4(1H)-one (80 mg, 0.17 mmol) was dissolved in methanol (5 mL) with the addition of HCl (1 mL, conc.) heated to 80° C. for 24 hours. The reaction mixture was diluted with water (10 mL) and the organics extracted with EtOAc (3×10 mL) before being dried and concentrated in vacuo. The crude material was then dissolved in dry THF under inert atmosphere and cooled to 0° C. Diisobutylaluminium hydride was then added slowly with stirring. After two hours the reaction pH was lowered to 3 through addition of HCl (2M). Dilution of the solution with water caused precipitation. The title compound was isolated as via filtration as a pale yellow solid (38 mg, 0.09 mmol, 55%). 1H NMR (400 MHz, DMSO) δ 11.06 (s, 1H), 7.41 (d, J=8.8 Hz, 2H), 7.25 (d, J=8.6 Hz, 2H), 7.15 (d, J=9.1 Hz, 2H), 7.05 (d, J=8.6 Hz, 2H), 5.53 (s, 1H), 4.25 (s, 2H), 2.67 (t, J=5.6 Hz, 2H), 2.35 (t, J=6.2 Hz, 2H), 1.82-1.56 (m, 4H); M/Z (ESI+); 432.14 (Found MH+; 432.1445, $C_{23}H_{20}F_3NO_4$ requires 432.1423).

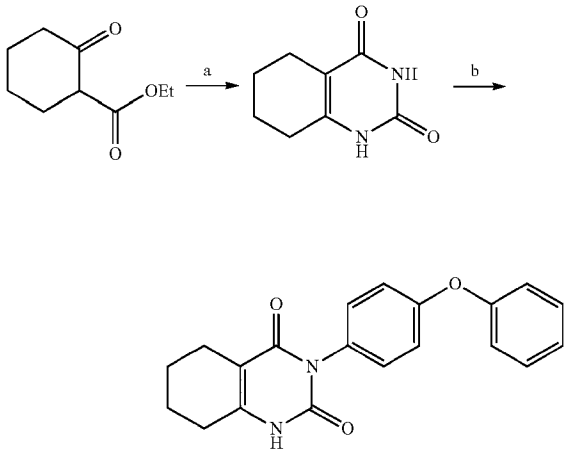


a) MeI, NaH, DMF, 90° C.

1,2-dimethyl-3-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4-one

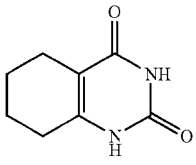


2-methyl-3-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-5,6,7,8-tetrahydroquinolin-4(1H)-one (60 mg, 0.14 mmol) was dissolved in DMF (0.1 mL) under Nitrogen. Sodium Hydride (4 mg, 0.17 mmol) was added and the reaction heated to 90° C. for 30 minutes. Methyl iodide (80 mg, 0.56 mmol) was added and the reaction continued for a further 2 hours. Reaction mixture was allowed to cool and organics extracted with ethyl acetate, followed by drying with brine and magnesium sulphate. Organics were concentrated in vacuo and purified by column chromatography (DCM). To give the title compound as a colourless solid (20 mg, 0.04 mmol, 35%). ^{1}H NMR (400 MHz, DMSO) δ 7.41 (d, J=8.6 Hz, 2H), 7.19-7.08 (m, 4H), 7.06 (d, J=8.5 Hz, 2H), 3.53 (s, 3H), 2.71 (t, J=6.2 Hz, 2H), 2.38 (t, J=6.0 Hz, 2H), 2.19 (s, 3H), 1.75 (d, J=5.7 Hz, 2H), 1.66-1.56 (m, 2H).



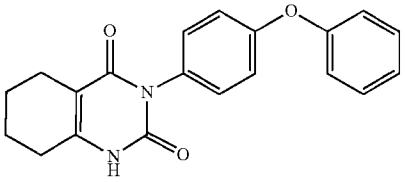
a) Urea, NaOME, Ethanol, 80° C., b) 4-phenoxyphenyl boronic acid, $CuOAc_2$, Pyridine, triethylamine, Ethanol

1,2,3,4,5,6,7,8-Octahydroquinazoline-2,4,dione

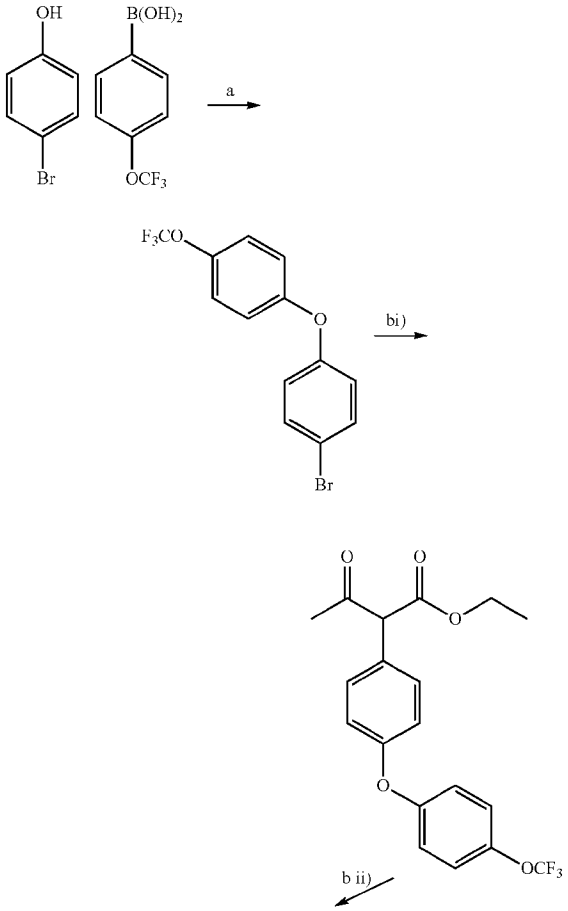


Urea (0.48 g, 8.0 mmol) and ethyl-2-oxocyclohexancarbonylate (1.0 g, 6.0 mmol) were dissolved in ethanol (10 mL). Sodium methoxide (3 mL, 12.0 mmol) was added and the reaction mixture refluxed at 80° C. for 15 hours. The reaction mixture was allowed to cool and the resulting precipitate was washed with diethyl ether (2×10 mL) to afford the title compound as a white solid. (526 mg 3.17 mmol, 53%). ^{1}H NMR (501 MHz, DMSO) δ 10.72 (s, 1H), 8.50 (s, 1H), 2.28 (t, J=6.2 Hz, 2H), 2.12 (t, J=6.2 Hz, 2H), 1.69-1.60 (m, 2H), 1.59-1.51 (m, 2H); M/Z (ESI+); 167.08 (Found MH^+; 167.0815, $C_8H_{10}N_2O_2$ requires 167.0815).

3-(4-Phenoxyphenyl)-1,2,3,4,5,6,7,8-octahydroquinazoline-2,4,dione



1,2,3,4,5,6,7,8-Octahydroquinazoline-2,4,dione (300 mg, 1.8 mmol), 4phenoxyphenyl boronic acid (600 mg, 2.7 mmol) and copper acetate (330 mg, 1.8 mmol) were dissolved in ethanol (20 mL). Triethylamine (1.2 mL, 9.0 mmol), and pyridine (0.66 mL, 9.0 mmol) were added immediately and the reaction stirred overnight. The reaction was filtered through celite neutralised with HCl (0.5 M, 60 mL) to give crude solid, this was then recrystallized in DCM to afford product as colourless needles (60 mg, 0.18 mmol, 10%). ^{1}H NMR (500 MHz, DMSO) δ 11.04 (s, 1H), 7.45 (t, J=7.1 Hz, 2H), 7.20 (d, J=7.5 Hz, 3H), 7.09 (d, J=7.8 Hz, 2H), 7.04 (d, J=7.9 Hz, 2H), 2.39 (s, 2H), 2.22 (s, 2H), 1.68 (m, 4H); M/Z (ESI+); 335.14 (Found MH$^+$; 335.1394, $C_{20}H_{18}N_2O_3$ requires 335.1390).



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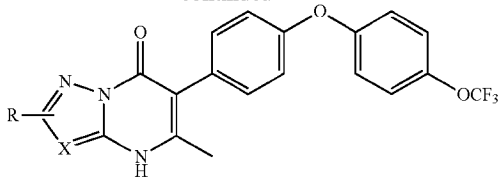
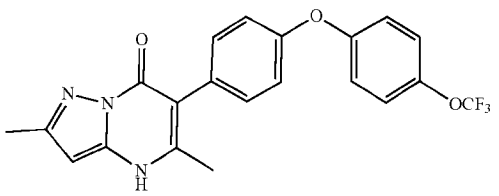


a) Cu(OAc)$_2$, Pyridine, TEA, DCM, b)i) Ethylacetoacetate, PdOAc$_2$, JohnPhos, K_3PO_4, Tol, 90° C., b)ii) pyrazole/triazole, AcOH, 120° C.

General Procedure B

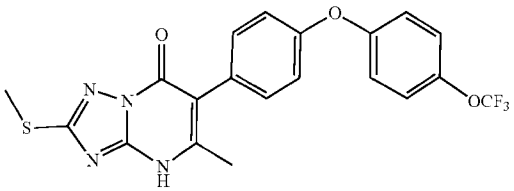
Toluene (2.00 mL) was added to a flask flushed with nitrogen and charged with 1-Bromo-4-(4-trifluoromethoxy) phenoxy)benzene (0.30 g, 0.90 mmol), ethyl acetoacetate (0.252 mL, 0.99 mmol), palladium acetate (10.1 mg, 0.045 mmol), (2-Biphenyl)di-tert-butylphosphine (26.8 mg, 0.09 mmol) and potassium phosphate (0.252 g, 1.2 mmol). The reaction mixture was then heated to 90° C. for 16 hours. The reaction mixture was cooled to room temperature followed by dilution in DCM (15.0 mL) and filtration through a pad of Celite. The reaction mixture was then concentrated in vacuo and passed through a silica plug. The resulting oil containing crude ethyl 3-oxo-2-4-(4-(trifluoromethoxy)phenoxy)phenyl)butanoate (1 equiv.) was dissolved in acetic acid (2.00 ml). 3-amino-tria-/pyrazole (1 equiv.) was added to the solution. The solution was heated to 120° C. and refluxed for 16 hours. The reaction mixture was then allowed to cool to room temperature. Addition of H_2O (2.00 ml) caused precipitation of a white solid. Precipitate was filtered and washed with H_2O (2×10.0 mL). The solid was then recrystallized in appropriate solvent to give the title compound.

2,5-dimethyl-6-(4-(4-(trifluoromethoxy)phenoxy) phenyl)pyrazolo[1,5-a]pyrimidin-7(4H)-one



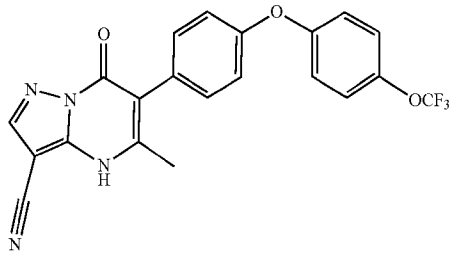
The title compound was synthesised from 3-amino-5 methyl-pyrazole (30.0 mg, 0.31 mmol) according to general procedure B (recrystallized in EtOAc) to afford the title compound as colourless platelets (47.0 mg, 0.11 mmol, 36%). δ H NMR (500 MHz, Chloroform-d); δ 7.27 (d, J=8.6 Hz, 2H), 7.16 (d, J=8.7 Hz, 2H), 6.99 (d, J=8.9 Hz, 2H), 6.95 (d, J=8.6 Hz, 2H), 5.80 (s, 1H), 2.34 (s, 3H), 2.21 (s, 3H) M/Z (ESI); 419.12 (Found MH$^+$ 416.1221, $C_{21}H_{16}F_3N_3O_3$ requires 416.1217).

5-methyl-2-(methylthio)-6-(4-(4-(trifluoromethoxy) phenoxy)phenyl)-[1,2,4]triazolo [1,5-a]pyrimidin-7 (4H)-one



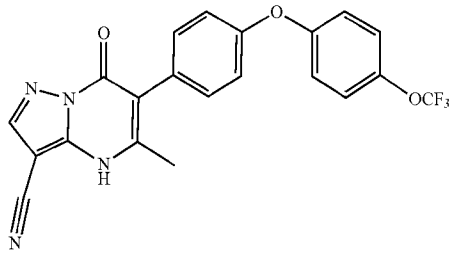
The title compound was synthesised from 3-amino-5-methio-1.2,4triazole (30.0 mg, 0.31 mmol) according to general procedure B to afford the title compound as colourless flake crystals (recrystallized EtOAc) (54.0 mg, 0.12 mmol, 39%). δ H NMR (500 MHz, Chloroform-d); δ 7.30 (d, J=8.6 Hz, 2H), 7.22 (d, J=8.7 Hz, 2H), 7.09 (d, J=8.7 Hz, 2H), 7.07 (d, J=8.6 Hz, 2H), 2.74 (s, 3H), 2.49 (s, 2H); M/Z (ESI); 449.09 (Found MH^+ 449.0893, $C_{20}H_{15}F_3N_4O_3S$ requires 449.0893).

5-methyl-7-oxo-6-(4-(4-(trifluoromethoxy)phenoxy) phenyl)-4,7-dihydropyrazolo[1,5-a]pyrimidine-3-carbonitrile



The title compound was synthesised from 3-amino-4-carbonitrile-1,2,pyrazole (45 mg, 0.42 mmol) according to general procedure B to afford the title compound as white flake crystals (recrystallized EtOAc), (43 mg, 0.10 mmol, 23%); ^1H NMR (500 MHz, DMSO) δ 8.49 (s, 1H), 7.51 (d, J=8.9 Hz, 2H), 7.44 (d, J=8.6 Hz, 2H), 7.28 (d, J=8.9, Hz, 2H), 7.20 (d, J=8.6 Hz, 2H), 2.32 (s, 3H). M/Z (ESI+); 427.10 (Found MH^+ 427.1018, $C_{21}H_{13}F_3N_4O_3$ requires 427.1012).

5-methyl-2-phenyl-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl)pyrazolo[1,5-a]pyrimidin-7(4H)-one



The title compound was synthesised from 3-amino-4-carbonitrile-1,2,pyrazole (0.067 g, 0.42 mmol) according to general procedure B (recrystallized in DMSO) to afford the title compound as a grey micro crystals (0.15 g, 0.31 mmol, 74%). ^1H NMR (500 MHz, DMSO) δ 12.47 (s, 1H, NH), 8.01 (d, J=6.8 Hz, 2H, H-2* & 6*), 7.53-7.40 (m, 5H, H-3*, 4*, 5*,2' & 6'), 7.38 (d, J=8.7 Hz, 2H, H-2" & 6"), 7.20 (d, J=9.1 Hz, 2H, H-3' & 5'), 7.11 (d, J=8.7 Hz, 2H, H-3" & 5"), 6.62 (s, 1H, H-3), 2.22 (s, 3H, Me); M/Z (ESI+); 478.1378 (Found MH+, $C_{26}H_{18}F_3N_3O_3$ requires 478.1373).

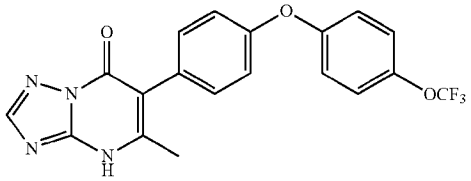
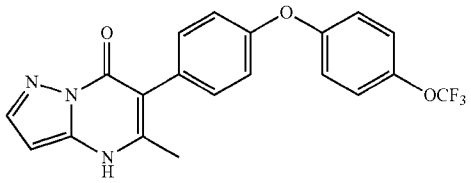
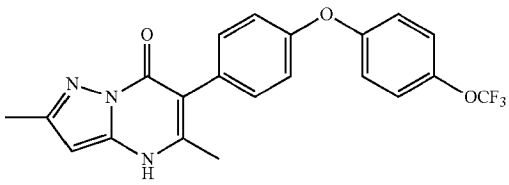
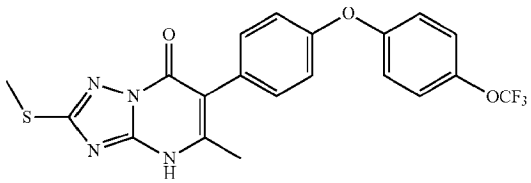
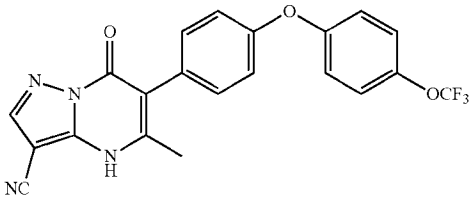
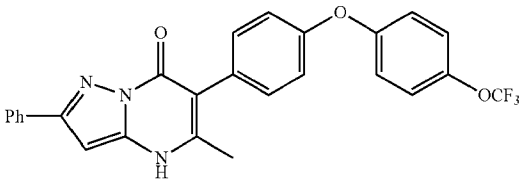
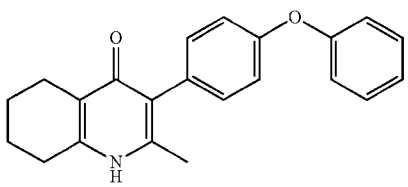
In Vitro Challenge Assay for *Toxoplasma* Tachyzoites

Protocol adapted from Fomovska, et. al. (Fomovska A, Huang Q, El Bissati K, Mui E J, Witola W H, Cheng G, et al. Novel N-Benzoyl-2-Hydroxybenzamide Disrupts Unique Parasite Secretory Pathway. Antimicrob Agents Chemother [Internet]. 2012 May [cited 2015 Jul. 8]; 56(5):2666-82; Fomovska A, Wood R D, Mui E, Dubey J P, Ferreira L R, Hickman M R, et al. Salicylanilide inhibitors of *Toxoplasma gondii*. J Med Chem. 2012 Oct. 11; 55(19):8375-91). Human foreskin fibroblasts (HFF) were cultured on a flat, clear-bottomed, black 96-well plate to 90% to 100% confluence. IMDM (1×, [+] glutamine, [+] 25 mM HEPES, [+] Phenol red, 10% FBS [gibco, Denmark]) was removed from each well and replaced with IMDM-C (1×, [+] glutamine, [+] 25 mM HEPES, [−] Phenol red, 10% FBS)[gibco, Denmark]). Type I RH parasites expressing Yellow Fluorescent Protein (RH—YFP) were lysed from host cells by double passage through a 27-gauge needle. Parasites were counted and diluted to 32,000/mL in IMDM-C. Fibroblast cultures were infected with 3200 tachyzoites of the Type I RH strain expressing Yellow Fluorescent Protein (RH—YFP) and returned to incubator at 37° C. for 1-2 hours to allow for infection (Gubbels M-J, Li C, Striepen B. High-Throughput Growth Assay for *Toxoplasma gondii* Using Yellow Fluorescent Protein. Antimicrob Agents Chemother [Internet]. 2003 January [cited 2015 Jul. 8]; 47(1):309-16). Various concentrations of the compounds were made using IMDM-C, and 20 μl were added to each designated well, with triplicates for each condition. Controls included pyrimethamine/sulfadiazine (current standard of treatment), 0.1% DMSO only, fibroblast only, and an untreated YFP gradient with 2 fold dilutions of the parasite. Cells were incubated at 37° C. for 72 hours. Plates were read using a fluorimeter (Synergy H4 Hybrid Reader, BioTek) to ascertain the amount of yellow fluorescent protein, in relative fluorescence units (RFU), as a measure of parasite burden after treatment. Data was collected using Gen5 software. IC50 was calculated by graphical analysis in Excel.

In Vitro Challenge Assay for Bradyzoites

HFF cells were grown in IMDM (1×, [+] glutamine, [+] 25 mM HEPES, [+] Phenol red, 10% FBS, [gibco, Denmark]) on removable, sterile glass disks in the bottom of a clear, flat-bottomed 24-well plate. Cultures were infected with 3×104 parasites (EGS strain) per well, in 0.5 mL media and plate was returned to incubator at 37° C. overnight. The following day, the media was removed and clear IMDM and compounds were added to making various concentrations of the drug, to a total volume of 0.5 mL. 2 wells were filled with media only, as a control. Plates were returned to the 37° C. incubator for 72 hours, and checked once every 24 hours. If tachyzoites were visible in the control before 72 hours, the cells were fixed and stained. Cells were fixed using 4% paraformaldehyde and stained with Fluorescein-labeled *Dolichos biflorus* Agglutinin, DAPI, and BAG1. Disks were removed and mounted onto glass slides and visualized using microscopy (Nikon T17). Slides were scanned using a CRi Pannoramic Scan Whole Slide Scanner and viewed using

Panoramic Viewer Software. Effects of the compounds were quantified by counting cysts in the controls and treated cells. Cysts and persisting organisms were counted in a representative field of view and then multiplied by a factor determined by the total area of the disk in order to estimate the number of cysts and organisms in each condition. Data was collected using Gen5 software. IC50 was calculated by graphical analysis in Excel.

ID	Structure	Tachy IC_{50} μM	Brady IC_{50} μM	Pf D6 IC_{50} μM
MJM136		1.0-10.0	>10	0.2
MJM141		1.0-10.0	1.0-10.0	0.16
JAG006		>1	N.D.	0.29
JAG013		>1	N.D.	1.31
JAG014		>1	N.D.	0.71
JAG015		>1	N.D.	>20
MJM170		0.03	1.0-10.0	0.01

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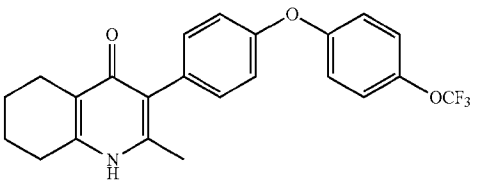
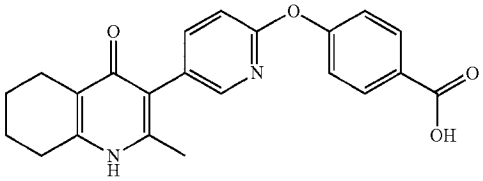
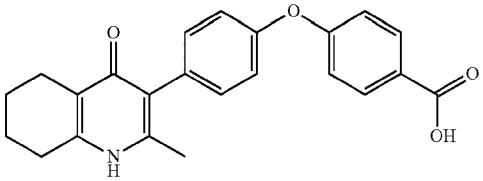
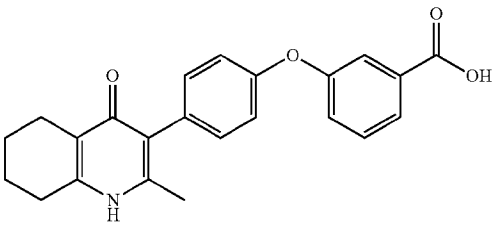
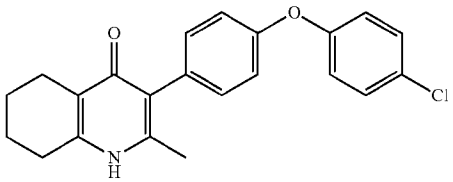
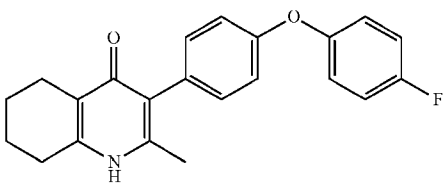
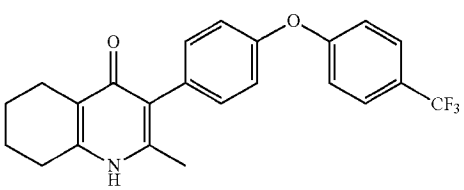
ID	Structure	Tachy IC_{50} μM	Brady IC_{50} μM	Pf D6 IC_{50} μM
JAG21		0.09	N.D.	0.01435
JAG039		7.6	N.D.	9.595
JAG046		>10	N.D.	6.716
JAG047		>10	N.D.	3.746
JAG50		0.055	N.D.	0.04664
JAG58		0.04-0.08	N.D.	Awaiting Testing
JAG63		0.1-0.3	N.D.	Awaiting Testing

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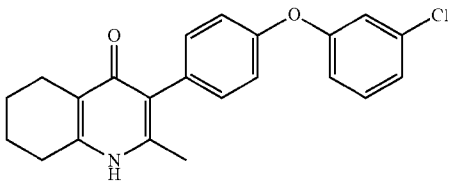
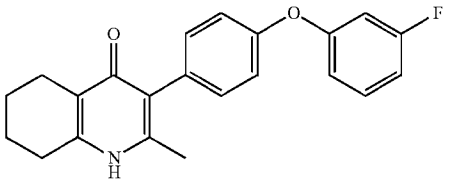
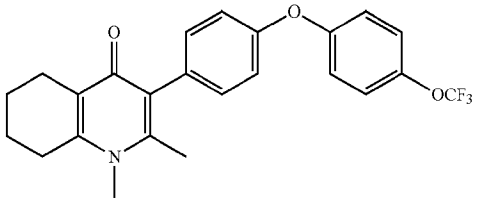
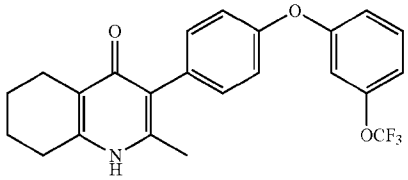
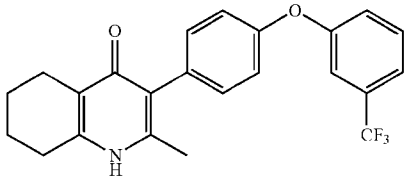
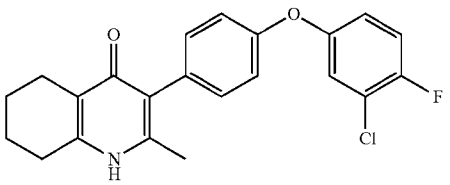
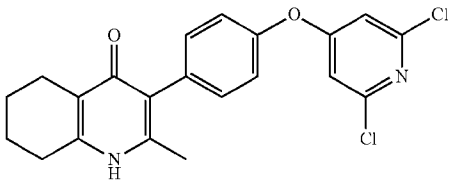
ID	Structure	Tachy IC_{50} μM	Brady IC_{50} μM	Pf D6 IC_{50} μM
JAG062		0.016	N.D.	N.D.
JAG069		0.02	N.D.	N.D.
JAG023		1	N.D.	N.D.
AS006/ JAG143		0.06-0.08	N.D.	N.D.
AS012/ JAG144		0.3	N.D.	N.D.
AS021/ JAG145		0.08	N.D.	N.D.
AS034/ JAG148		0.1-0.5	N.D.	N.D.

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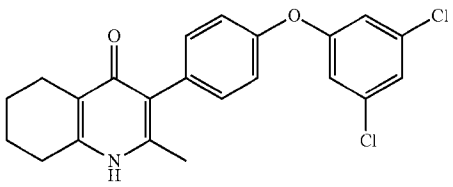
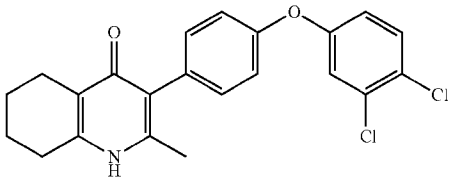
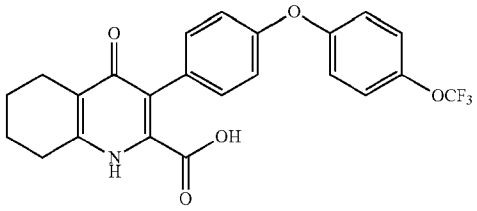
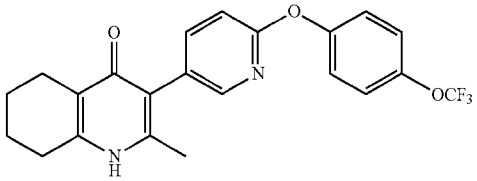
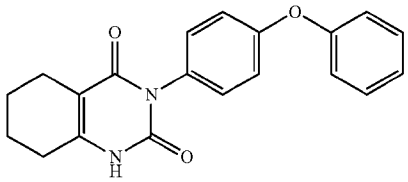
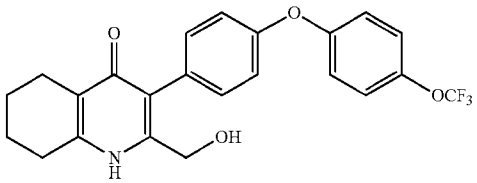
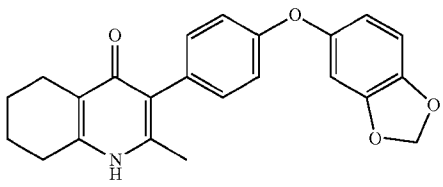
AS022		0.02-0.04	N.D.	N.D.
JAG084		0.04-0.08	N.D.	N.D.
JAG091		>1	N.D.	N.D.
JAG092		1	N.D.	N.D.
JAG095		>10	N.D.	N.D.
JAG099		0.32	N.D.	N.D.
AS032		0.1-0.3	N.D.	N.D.

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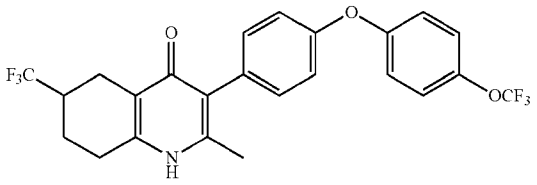
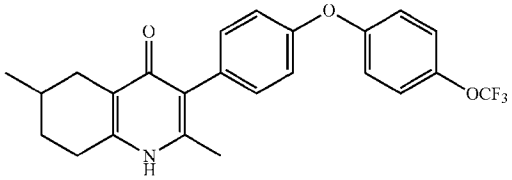
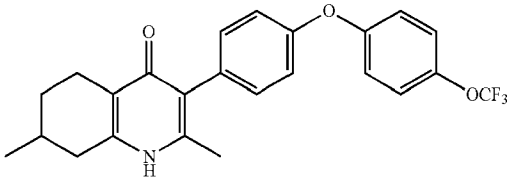
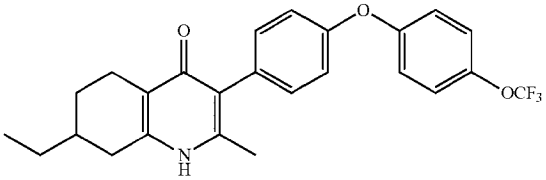
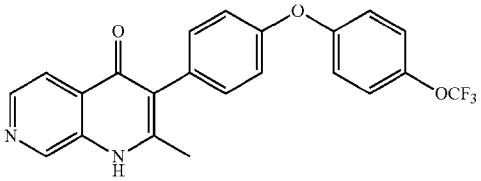
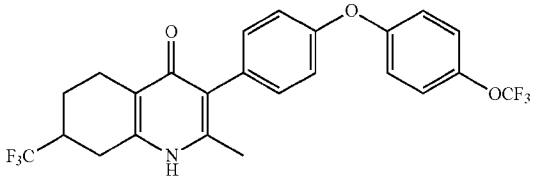
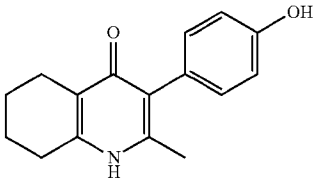
JAG100		>10	N.D.	N.D.
JAG106		~1	N.D.	N.D.
JAG107		1	N.D.	N.D.
JAG121		0.1	N.D.	N.D.
JAG129		0.1	N.D.	N.D.
JAG162		0.5	N.D.	N.D.
JAG094		1	N.D.	N.D.

Biological Activity Studies

Malaria In Vitro Studies:

D6 is a drug sensitive strain from Sierra Leone, C235 is a multi-drug resistant stain from Thailand, W2 is a chloroquine resistant strain from Thailand, and C2B has resistance to a variety of drugs including atovaquone. These assays were performed using standard protocols.

Compound Activity Against *Plasmodium* falciparum

Compound activity against *P. falciparum*, a causative agent of malaria, was tested using the Malaria SYBR Green I—Based Fluorescence (MSF) Assay. This; microtiter plate drug sensitivity assay uses the presence of malarial DNA as a measure of parasitic proliferation in the presence of antimalarial drugs or experimental compounds based on

modifications of previously described methods known in the art. As the intercalation of SYBR Green I dye and its resulting fluorescence is relative to parasite growth, a test compound that inhibits the growth of the parasite will result in a lower fluorescence. Selected compounds were examined for activity against four strains of *P. falciparum*: D6 (CDC/Sierra Leone), a drugsensitive strain readily killed by chloroquine, TM91-C235, a multi-drug resistant strain resistant to chloroquine, W2, a chloroquine resistant strain from Thailand, and C2B has resistance to a variety of drugs including atovaquone.

ID	Parasite strains	SYBR Green D6 IC50 (uM)	SYBR D6 R^2	SYBR Green C235 IC50 (uM)	SYBR TM91C235 R^2
MJM129	D6, C235, W2, C2B	0.03	0.94	0.07	0.94
MJM136	D6, C235, W2, C2B	0.20	0.99	0.58	0.98
MJM141	D6, C235, W2, C2B	0.16	0.96	0.57	0.95
MJM170	D6, C235, W2, C2B	0.01	0.98	0.03	0.99
JAG006	D6, C235, W2, C2B	0.29	0.90	0.88	0.92
JAG013	D6, C235, W2, C2B	1.31	0.98	2.60	0.96
JAG014	D6, C235, W2, C2B	0.71	0.94	1.35	0.99
JAG015	D6, C235, W2, C2B	>20	N/A	>20	N/A
JM10	D6, C235, W2, C2B	0.88	0.94	4.48	0.97
JAG021	D6, C235, W2, C2B	0.01435	0.9572	0.06164	0.9706
JAG047	D6, C235, W2, C2B	3.746	0.9738	12.56	0.9218
JAG046	D6, C235, W2, C2B	6.716	0.9844	>20	N/A
JAG039	D6, C235, W2, C2B	9.595	0.9532	>20	N/A
JAG050	D6, C235, W2, C2B	0.04664	0.9138	0.06913	0.9562
RG38	D6, C235, W2, C2B	2.884	0.8936	13.66	0.8338

ID	Parasite strains	SYBR Green W2 IC50 (uM)	SYBR W2 R^2	SYBR Green C2B IC50 (uM)	SYBR C2B R^2
MJM129	D6, C235, W2, C2B	0.08	0.93	0.01	0.94
MJM136	D6, C235, W2, C2B	0.55	0.98	0.38	0.99
MJM141	D6, C235, W2, C2B	0.63	0.88	0.48	0.95
MJM170	D6, C235, W2, C2B	0.03	0.99	0.01	0.99
JAG006	D6, C235, W2, C2B	2.46	0.92	1.66	0.94
JAG013	D6, C235, W2, C2B	2.35	0.96	1.39	0.94
JAG014	D6, C235, W2, C2B	1.27	0.99	0.92	0.98
JAG015	D6, C235, W2, C2B	>20	N/A	>20	N/A
JM10	D6, C235, W2, C2B	5.36	0.90	6.75	0.81
JAG021	D6, C235, W2, C2B	0.05518	0.9727	0.04042	0.9847
JAG047	D6, C235, W2, C2B	9.072	0.9358	7.781	0.9575
JAG046	D6, C235, W2, C2B	>20	N/A	>20	N/A
JAG039	D6, C235, W2, C2B	>20	N/A	>20	N/A
JAG050	D6, C235, W2, C2B	0.03136	0.9693	0.03635	0.9427
RG38	D6, C235, W2, C2B	9.245	0.7954	>20	N/A

Example 5: Effect of Active Forms of *T. gondii* on Transcriptomes, Proteomes and Mechanisms Whereby this Occurs and Reflection of the Same Type of Damage to Neuronal Cells in Circulating Biomarkers from Children

Since we found signature pathways reflecting influence of the bradyzoite stage (characteristic of the chronic *Toxoplasma gondii* infection) in primary human neuronal stem cells in tissue culture on pathways of neurodevelopment, neuroplasticity, and neurotoxicity, we asked whether the active form of the parasite would also affect those pathways. It did. We found alterations in pathways similar to those shown with EGS bradyzoites in transcriptomics and proteomics (FIGS. **14A-14D** with explanatory figure legends and methods). These abnormalities suggested that there might be circulating biomarkers reflecting such damage to neuronal cells in patients. In that context we then asked whether serum biomarkers from ill children would reflect neuronal damage and neurodegeneration.

Methods.

Biomarkers: Serum collection was from children in the National Collaborative Congenital ToxoplasmosisStudy (NCCCTS). Children have serum drawn at each visit. Sera characterized were obtained at a visit when new seizures were noted for ill children. These sera were analyzed with nano proteomic and miR analyses as described earlier by Hood, Wang et al. This was done using a panel of markers known to be abnormal in patients with Alzheimer's and other neurodegenerative diseases. This was done to determine whether the same biomarkers present in serum or plasma from persons with these neurodegenerative disease might be present in sera from the ill children. The children are described more fully in the figure legend.

Murine study of Apolipoprotein A1: Wildtype mice on a C57Bl6/J background, mice in which the Apolipoprotein A1 gene was knocked out (Apo A1−/−) were utilized in this experiment. They were immunized with an attenuated strain of the RH strain of *Toxoplasma* in which ribosomal proteins small subunit 13 was placed under the control of a tetracycline repressor by placing 4 tet 0 elements in the promoter and a tetracycline regulatable repressor with YFP was stably transfected. This immunized the mice and subsequently the mice were challenged with the Me49 strain *T. gondii* and cysts were counted or luminescence in brain measured.

EGS and Canonical Type 1,2,3 transcriptomics details and Type 1, 2, 3 proteomics, analysis of alternatively spliced genes, and immunoflurescense studies: Details of the specific genes with altered transcription caused by EGS in Example 1 are discussed above. Trascriptomics were carried out as described in Example 1.

iTRAQ data from *T. gondii* infected cell cultures. Protein quantified, extracted, subjected to mass spectroscopy, and sequence analysis from each flask, ~180-190 ug proteins were extracted and 50 ug were used for 8-plex iTRAQ. A raw table listing relative ratios for all peptide identified in 8 samples was created. The ratio should be 0.125 (1.000/8) if one peptide/protein evenly distributes in 8 samples. Ratios of peptides from the same proteins are then calculated to protein ratios. A "Ratio to Channel 0" then included a total of 4,367 proteins identified with iTRAQ ratio. The protein ratios crossed 8 samples (4 conditions in duplicates) and were raw data from mass spec and converted to ratios against Channel 0, i.e. Control sample. They are then normalized and ratios made. "Prot with high score" has 3,359 proteins identified by more than 1 peptide and with ProteinProphet probability >0.8 (=FDR<1%). Among these 3,3359 proteins with high confidence, 10 proteins up >2-fold in either of the 3 infected cells vs. controls, while 28 proteins down >2-fold were identified. Occurrence of differences in alternative splicing between infected and uninfected cells was done with rMATS. Method for IFA are as described in Example 1. The antibodies are to SAG1, P50-NFkB.

Results:

Human serum biomarkers in ill congenitally infected children reflect *T. gondii* infection and neuronal damage. Three pairs of children were studied. In each demographically-matched pair, one child had severe disease and the other had mild or no clinical illness. Each child had serum stored from evaluations at the same ages. The second pair are dizygotic, discordant twins. Each of the three ill children had new myoclonic-"infantile" spasms, or hypsarrhythmic seizures. For two, this was associated with a rise in or high *T. gondii* specific IgG antibody titers (FIG. **14**A). IgG was not measured for the third ill child. A panel of nanoproteomics and miR sequencing was performed on serum obtained at the time of this new illness. The two ill children diagnosed more recently had T2 weighted abnormalities on brain MRIs similar to active inflammatory and parasitic caused brain disease seen in a murine model. Ill children compared with their paired healthy controls had alterations in miRs and increases in serum proteins associated with neurodegeneration, inflammation, a misfolded protein response and protein misfolding. Elevated proteins included clusterin, and oxytocin (FIGS. **14**A-**14**D). PGLYRP2 (N-acetylmuramoyl-L-alanine amidase) and Apolipoprotein B1 were depressed. miR-17-92, which *T. gondi* RH strain markedly increases in HFF cultures, also was increased in sera of the ill children, as was miR-124 (FIGS. **14**B-**14**C). miR-124 is associated with neurodegeneration. This indicates active brain destruction by the parasite or the response to it. These circulating proteins and miRs are clinically useful biomarkers to identify active toxoplasmic brain (and possibly retinal) disease.

To determine whether the presence of one of these biomarkers could be confirmed, a murine model was used. In this example of biomarkers in a murine experiment recapitulating the data of biomarkers in the serum of the ill children, APOA1 knockout and wild type mice were infected with *Toxoplasma*. The wild type and uninfected mice had less radiance from luciferase parasites and fewer cysts, and less immunologic reaction to the lower parasite burden in brain (data not shown). This demonstrates that the circulating ApoA1 diminishing in the boys who were ill as a biomarker. This provides evidence that these biomarkers that were abnormal in the children had counterparts in murine models.

To determine whether similar pathways as were abnormal in EGS Example 1 were perturbed by the canonical U.S./European types of parasites and mechanisms whereby this might occur, transcriptomics, proteomics, analysis of alternatively spliced genes and immunofluorescent were also preformed. Experimental data showed similar perturbation of pathways by canonical U.S./European type parasites that infected the children with the biomarkers through similar transcriptomics analyses demonstrating biological effect of Type I, II, III parasites on localization of NFkB, STAT 3 and STAT 6 in primary human neuronal stem cells. These abnormalities are caused by the canonical U.S. and European types of parasites growing as tachyzoites in the human primary neuronal stem cells and monocytic cells. These finding along with those demonstrated by the Omic studies of EGS (Example 1) suggest a mechanism whereby circulating biomarkers reflecting damage to neuronal cells in patients can occur. Placed in this context, we hypothesize that serum biomarkers from ill children reflect neuronal damage and neurodegeneration, as confirmed by our murine models, and findings seen in tissue culture and/or in patients.

Discussion

The signature pathways we noted in studies of human primary neuronal stem cells which reflected abnormalities and gene products associated with neurodegenerative disease and the mechanisms whereby *T. gondii* can cause such pathology prompted study of biomarkers in a small number of ill versus well children. Biomarkers of active brain *T. gondii* infection in humans were found. The serum biomarkers shown in FIGS. **14**A-**14**D are increased (e.g., clusterin, oxytocin, amyloid, and mir 17-92 and mir 124) or diminished, including PGLYRP2 (N-acetylmuramoyl-L-alanine amidase) and Apolipooprotein-A1 which are indicative of infection. These are consistent with the transcriptome demonstrating signature pathways in GO slim and KEGG analyses with effect on ribosomes, alternative splicing and neurodegenerative diseases, including Alzheimer's disease, Huntingtons disease, and Parkinson's disease by encysted EGS, and for example pathways of response to oxidative stress, regulation of apotosis, and alternative splicing of toll receptors that were abnormal in the same cells infected by the canonical US/European parasites (active tachyzoites) that the children had. Other manifestations of active disease in the brain diminished with treatment and are not abnormal in the dizygotic healthy twin of one child or demographically matched well children. This is consistent with these biomarkers being selected to be assayed with MiR sequencing and proteomics based on their differences in diseases of neurodegeneration. These ill children had developed new seizures, elevations in antibody titer. elevated cerebrospinal fluid protein in one child, and abnormal T2 weighted alterations in T2 weighted brain magnetic resonance imaging. The biomarkers that were characteristic of neurodegeneration in the ill children and when diminished were associated with greater severity of disease in a murine model will be useful to monitor disease and response to treatment of disease due to this parasite. Restoration to normal values being indicative of favorable response to treatment and presence may also mark recrudescence of disease. ApoA1 may also be a useful treatment.

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His Leu Ser Ser Ala Ala Leu Leu Gln Leu Val Gln Thr Gln Ser Ala
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Gly Gly Val Pro Gln Ala Val Leu Arg Asn Leu Phe Ser Ser Ile His
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Ser Ser Leu Tyr Ala Ser Leu Thr Ser Leu Ser Ser Ala Ala Ala Leu
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Pro Asn Glu Ala Ser Pro Ile Leu Ala Leu Leu Gly Glu Ala Ala Arg
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Gly Phe Pro Ser Gln Thr Pro Tyr Glu Leu Ser Ala Ser Pro Ser His
    690                 695                 700

Gln Leu Asp Leu Leu Arg Leu Gly Ala Phe Leu Gly Gly Ala Gly Lys
705                 710                 715                 720

Gln Asp Ala Ser Val His Ser Asp Glu Thr Gly Thr Leu Ser Gly Glu
                725                 730                 735

Pro Ser His Arg Ser Cys Ser Leu Ser Arg Gly Leu Thr Gln Glu Ser
            740                 745                 750

Val Leu Gln Leu Ser Asp Thr Thr Ser Thr Ser Arg Glu Gly Glu Pro
        755                 760                 765

Asn Glu Pro Ser Gln Gly Cys Val Asn Val Ala Ala Ser Leu Pro Ala
    770                 775                 780

Phe Gly Pro Gln Pro Ser Ser Gly Ala Ala Lys Ala Arg Glu Gly Arg
785                 790                 795                 800

Arg Gly Ala Gly Gly Ala Gly Ala Ala Pro Pro Val Pro Leu Arg Ala
                805                 810                 815

Asp Val Thr Leu Gly Gly Asn Arg Pro His Tyr His Val Ala Lys Gln
            820                 825                 830

Glu Trp Arg Val Arg Tyr Tyr Met Asn Gly Lys Arg Lys Met Arg Thr
        835                 840                 845

Tyr Ser Ala Lys Phe Tyr Gly Tyr Glu Thr Ala His Thr Met Ala Glu
    850                 855                 860

Asp Phe Ala His Tyr Val Asp Lys His Glu Ala Leu Pro Asp Ser Met
865                 870                 875                 880

Met Met Thr Ala Met Met Leu Gln Ala Gln Ala Asn Ser Ala Ala Ser
                885                 890                 895

Ser Gly Gln Thr Val Pro Leu Ala Arg Gly Ile Arg Ala Ser Ser Ala
            900                 905                 910

Ser Ala Gly Ala Gly Gly His Val Ser Lys Ser Ala Thr Lys Gly Ser
        915                 920                 925

Val Ala Ala Ser Ser Glu Gly Ser Thr Ser Met Gly Ser Asp Ala Thr
    930                 935                 940

Arg Ser Gln Glu Gly Glu Ala Ala Glu Leu Cys Pro Leu Ala Ala Gly
945                 950                 955                 960

Leu Ser Arg Pro Leu Ala Ser Met His Ser Ala Ala Gly Asn Ala Val
                965                 970                 975

Ala Gln Gly Arg Gln Glu Ser Lys Glu Glu Ala Pro Gly Gly Gln Ala
            980                 985                 990

Trp Phe Gly Glu Pro Gly Lys Phe  Arg Ala Ser Ser Glu  Ala Ala Leu
        995                 1000                1005

Cys Gly  Ser Gly Ser Ser Ala  Glu Gly Arg Asp Gly  His Glu Ser
    1010                1015                1020

Glu Val  Leu Trp Ala Thr Leu  Gly Lys Val His Asp  Ala Ser Gln
    1025                1030                1035

Gly Lys  Lys Ile Lys Pro Glu  Lys Pro Leu Thr Val  Ala Arg Gly
    1040                1045                1050

Arg Leu  Ala Leu Gly Ala Glu  Asp Lys Ser Gln Asn  Leu Gly Val
    1055                1060                1065

Asp Leu  Gly Asp Ser Gly Gly  Ala Gln Gly Leu Pro  Gly Val Arg
    1070                1075                1080

Gln Pro  Arg Gln Met Lys Asn  Ser Glu Glu Cys Ser  Leu Arg Asp
    1085                1090                1095
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Ser Asp  Lys Gly Met Ala Leu  Ser Lys Arg Phe Gly  Phe Leu Pro
    1100                1105                1110

Ser Gln  Thr Pro Ser Cys Asp  Ser Met Thr Leu Pro  Phe Pro Gly
    1115                1120                1125

Gly Phe  Asp Ala Leu Ser Leu  Ser Ser Ala Leu Ser  Ser Cys Ala
    1130                1135                1140

Ser Leu  Pro Val Ala His Glu  Gly Asn Asn Phe Gln  Lys Gly His
    1145                1150                1155

Thr Gly  Asp Ile Val Ala Leu  Ala Ser Gln Ser Gly  Thr Gln Arg
    1160                1165                1170

Pro Ala  Ser Val Val Leu Ser  Arg Asp Ala Asn Val  Ser Gly Ser
    1175                1180                1185

Ser Pro  Ser His Pro Thr Trp  Gln Arg Glu Gly Ala  Ala Val Ser
    1190                1195                1200

Gly Arg  Ala Asp Glu Phe Ser  Ser Leu Ser Val Thr  Pro Ser Thr
    1205                1210                1215

Val Pro  Leu Ser Ser Phe Thr  Met Glu Asp Ile Lys  Gly Glu Glu
    1220                1225                1230

Gly Asp  Pro Ser Arg Arg Phe  Ala Leu Val Gly Glu  Ser Met Lys
    1235                1240                1245

Asn Val  Ser Ala Pro Glu Val  Gln Ala Leu Phe Pro  Thr Ser Ser
    1250                1255                1260

Ile Ala  Asn Ala Glu Leu Leu  Pro Val Asp Phe Leu  His Ser Asn
    1265                1270                1275

Ser Cys  Ser Ala Asp Lys Leu  Glu Ser Ser Ile Pro  Arg Gly Leu
    1280                1285                1290

Ala Gly  Asn Asn Pro Ser Met  Thr Ala Thr Ala Val  Ala Ala Thr
    1295                1300                1305

Ala Val  Ser His Gln Ile Phe  Asp Thr Ile Thr Leu  Phe Gly Glu
    1310                1315                1320

Phe Leu  Arg Glu Phe Ala Lys  Glu Lys Val Asn Glu  Phe His Glu
    1325                1330                1335

Tyr Gly  Leu Glu Ala Ser Pro  Leu Thr Val Glu Ala  Ser Pro Glu
    1340                1345                1350

Val Ser  Leu Phe Gly Lys Ala  Thr Phe Gly Arg Cys  Pro Val Ala
    1355                1360                1365

Gly Gly  Ser Thr Pro Ala Gly  Ile Ser Lys Met Ser  Gly Glu Thr
    1370                1375                1380

Leu Ser  Gly Leu Ser Ala Ser  Glu Leu Ser Leu Val  Ser Ala Arg
    1385                1390                1395

Thr Asn  Thr Thr Thr Gly Glu  Glu Gln Phe Ala Leu  Ala Arg Gly
    1400                1405                1410

Leu Phe  Pro Gly Asp Ser Glu  Gly Asp Arg Asp Glu  Lys Lys Pro
    1415                1420                1425

Gln Leu  Ser Gln Gln Glu Leu  Leu Val Leu Ser His  Ala Leu Val
    1430                1435                1440

Asn Leu  Thr Ser Ser Thr Tyr  Val Leu Met His Thr  Leu Lys Ala
    1445                1450                1455

Ser Leu  Ser Lys Ser Thr Glu  Ala Val Gln Leu His  Gln Pro Leu
    1460                1465                1470

Leu Glu  Ala Ala Ser Glu Ala  Lys Ala Thr Asp Glu  Ala Lys Thr
    1475                1480                1485

Arg Glu  Glu Gln Glu Ser Ser  Glu Cys Asp His Glu  Tyr Pro Pro
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    1490                1495                1500

Gly Ser  Ser Leu Glu Ala Thr  Thr Gly Ala Leu Pro  Phe Arg Leu
    1505                1510                1515

Ser Pro  Ala Leu Ser Ala Ser  Ser Lys Asp Leu Pro  Ser Leu Ser
    1520                1525                1530

Ala Ser  Ala Ser Leu Glu Ser  Val Thr Pro Phe Ala  Gly Leu Pro
    1535                1540                1545

Leu Glu  Glu Gly Thr Leu Ser  Ala Ser Val Gly Leu  Ala Ser Ser
    1550                1555                1560

Asp Asp  Glu His Asp Thr Ser  Leu Leu Phe Lys Thr  Glu Ala Ala
    1565                1570                1575

Lys Lys  Arg Ser Leu Phe Ser  Thr Ala Ala Asp Gly  Asp Glu Ser
    1580                1585                1590

Arg Thr  Tyr Asn Asp Gly Leu  Gly Gln Pro Met Glu  Glu Glu Ile
    1595                1600                1605

Arg Ser  Cys Val Ser Thr Ser  Cys Gly Glu Ala Val  Ala Thr Thr
    1610                1615                1620

Thr Leu  Ser Ala Ile Gly Pro  Gly Thr Gly Ala Ser  Gly Ala Leu
    1625                1630                1635

Leu Asp  Ser Glu Ser Arg Glu  Ser Leu Gly Glu Lys  Pro Gly Ala
    1640                1645                1650

Ala Leu  Arg Ala Gly Ala His  Thr Pro Ala Pro Ser  Arg Ala Pro
    1655                1660                1665

Thr Pro  Ser Arg Thr Phe Ser  Phe Thr Ser Ser Ser  Thr Ala Thr
    1670                1675                1680

Ser Ala  Ala Leu Leu Cys Asp  Ser Asn Val Val His  Glu Lys Leu
    1685                1690                1695

Ser Ala  Gln Gly Lys Asp Ser  Glu Ala Gly Glu Arg  Lys Gly Asp
    1700                1705                1710

Ser Glu  Lys Glu Glu Glu Val  Glu Met Trp Lys Glu  Glu Asp Glu
    1715                1720                1725

Glu Val  Gln Arg Cys Thr Gly  Ser Ala Glu Thr Asp  Ser Thr Glu
    1730                1735                1740

Ala Thr  Arg Gly Glu Glu Ala  Trp Arg Arg Gly Lys  Gln Ser Glu
    1745                1750                1755

Lys Lys  Pro Ser Val Ile Thr  Thr Ala Leu Asn Leu  Leu Glu Thr
    1760                1765                1770

His Arg  His Leu Ala Leu Thr  Ile Ser Gln Leu Lys  Arg Pro Val
    1775                1780                1785

Ala Gln  Gln Leu Arg Phe Ile  Leu Pro Ile Ala Ala  Pro Gln Leu
    1790                1795                1800

Leu Pro  Cys Ile Leu Pro Pro  Ala Ser Phe Gln Gly  Thr Gly Glu
    1805                1810                1815

Ser Gly  Asp Gly Lys Ala Glu  Ala Glu Ala Lys Gly  Ser Ser Ser
    1820                1825                1830

Leu Gly  Gln Val Leu Glu Thr  Ala Leu Gly His Gly  Thr Arg Leu
    1835                1840                1845

Ala Pro  Ser Ala Ser Ala Met  Val Pro Pro Arg Lys  Asp Glu Ala
    1850                1855                1860

Ala Ser  Ala Val Pro Glu Ala  Lys Thr Leu Thr Gly  Leu Ala Asn
    1865                1870                1875

Ala Gly  Val Thr Arg Glu Ala  Ala Ser Arg Thr Leu  Glu Ala Glu
    1880                1885                1890
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Gln Val  Ser Arg Lys Arg Ser  Arg Glu Glu Val Val  Asp Ser Glu
    1895                 1900                 1905

Thr Ala  Gly Asp Glu Gly Asp  Met Glu Asn Val Pro  Glu Thr Arg
    1910                 1915                 1920

Asp Gly  Thr Thr Arg Pro Gly  Ser Arg Gln Tyr Asp  Thr Ser Pro
    1925                 1930                 1935

Ser Asn  Asp Gly Thr Lys Pro  Pro Ala Thr Ala Lys  Ser Arg Val
    1940                 1945                 1950

Ile Arg  Asp Gln Ala Ala Leu  Glu Arg Leu Leu Leu  Ala Pro Phe
    1955                 1960                 1965

Gln Asp  Thr Pro Thr Cys Ser  Cys Thr Asp Arg Pro  Cys Pro Cys
    1970                 1975                 1980

Asp Arg  Gln Gln Val Ala Asp  Met Ile Tyr Leu Phe  Tyr Ala Val
    1985                 1990                 1995

Pro Ala  Arg Gln Gln Ala Glu  Ser Ser Lys Glu Gly  Ser Thr Gln
    2000                 2005                 2010

Arg Leu  Gln Phe Ala Ala Arg  Asp Thr Asn Glu Arg  Lys Asp Ala
    2015                 2020                 2025

Arg Thr  Gly Glu Glu Thr Gln  Gly Gly Glu Thr Glu  Ala Lys Glu
    2030                 2035                 2040

Val Ile  Arg Asp Pro Glu Glu  Arg Gly Val Cys Glu  Gly Ser Ser
    2045                 2050                 2055

Ser Gln  Asn Ala His Thr Gln  Phe Asp Ala Glu Thr  Ala Ser Ser
    2060                 2065                 2070

Ser Met  Ser Ser Asp Pro Arg  Ala Asp Lys Glu Ser  Asn Ala Gln
    2075                 2080                 2085

Asp Ala  His Met Ala Asp Lys  Thr Ser Phe Val Ser  Asp Leu Pro
    2090                 2095                 2100

Gln Pro  Ser Gly Glu Phe Ala  Pro Ser Leu Leu Ser  Glu Thr Ser
    2105                 2110                 2115

Leu Asp  Val Ala Met Ala Asp  Ser Arg Gly Thr Pro  Ser Glu Ile
    2120                 2125                 2130

His Gly  Phe Phe Thr Arg Ser  Asp Glu Gln Lys Arg  Ala Ser Phe
    2135                 2140                 2145

Ser Ser  Ser Ser Leu Leu Ala  Ala Gly His Ala Val  Ala Ser Phe
    2150                 2155                 2160

Ser Ser  Ser Leu Ala Gly Val  Val Ser Gly Ala Gly  Glu Arg Arg
    2165                 2170                 2175

Glu Cys  Ala Gly Pro Ser Leu  Gly Asp Leu Ser Thr  Ile Gly Leu
    2180                 2185                 2190

Leu Ser  Leu Ser Tyr Pro Ala  Met Leu Ala Phe Ile  Leu Pro Leu
    2195                 2200                 2205

Gln Ser  Leu Leu His Thr Val  Ser Gly Met Ile Leu  Thr Leu His
    2210                 2215                 2220

Lys Lys  Leu Ile His Arg Phe  Ile Cys Ala His Leu  Arg Leu Val
    2225                 2230                 2235

Leu Asp  Asp Asp Met Arg Arg  Pro Ala Gly Gly Ala  Leu Lys Ser
    2240                 2245                 2250

Arg Gly  Ala His Gly Asp Thr  Glu Ala Ala Glu Ala  Gln Val Glu
    2255                 2260                 2265

Arg Arg  Arg Arg Glu His Glu  Arg Glu Glu Thr Thr  Asn Leu Ala
    2270                 2275                 2280
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Ile Gly  Tyr Arg Glu Gly Asn  Ala Glu Ala Ala Asn  Thr Phe Pro
    2285                 2290                 2295

Leu Val  Asp Thr Val Ser Ser  Leu Leu Ser Pro Gly  Ser Leu Arg
    2300                 2305                 2310

Gln Glu  Asn Ser Glu Val Glu  Arg Arg Asp Asn Asp  Glu Glu Arg
    2315                 2320                 2325

Leu Glu  Leu Ile Thr Gly Ile  Ala Arg Glu Ser Pro  Lys Pro Ser
    2330                 2335                 2340

Glu Lys  Asp Ser Val Ser Pro  Phe Leu Ser Thr Ala  Pro Cys Pro
    2345                 2350                 2355

Gly Thr  Glu Ala Glu Ser Ser  Asp Cys Ser Ala Ser  Ser Ala Cys
    2360                 2365                 2370

Ser Gly  Thr Pro Thr Glu Gly  Thr Glu Gly Gly Glu  Thr Gly Asp
    2375                 2380                 2385

Ile Ala  Ser Phe Leu Ser Pro  Ser Gly Glu Val Lys  Gln Thr Ile
    2390                 2395                 2400

Met Leu  Ala
    2405
```

<210> SEQ ID NO 3
<211> LENGTH: 1217
<212> TYPE: DNA
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 3

```
tatccagttg cccggctctg tctaccttct tctcgatgta gcggcggcgt tctgtgttcc     60

ttagagcgag tccacgcacc agcctcgtgg agtcgaacag ttttcgccag agcggggaca    120

gccggtggaa gacagaacag gagggattca ccgtcacctc gtttgcttcc ttctggtgat    180

ggagggaata agccgacaga ctcacttctt aacagttcat ggcagagggg caacgacgtc    240

accatccagc tttccattca cttaaccgca caagcgttct gctcgttagt caagcctcac    300

tcgccagtcg aaagccatta aaagttcgcg tttctggctc tcgtttgtgc cggaagtctg    360

ccaaagcaaa caaaggatct gaaaccgagt agcagtgaac gcgtgacagt gcagaggggt    420

gcccgcgtcg gcgtcctctc tgtccacgaa cagccacaat tctcatcacg agaagtcctc    480

ctcccctgga tcttcctcca gttccccatg actgcctccg gttctcgcac ggacggcgat    540

ccgccttcac gctccccggc ctttgccagt tttgtgcaag aaggtcgccc gtcttattct    600

tgtcacgttc cctgtatcta ctcggttcac tgcgctctac agaagctctt cttcgccttc    660

tactgatcag tgcactctac gagatgcact atcatcagtc ctgaactcca tgtttgggtg    720

gaaacctggg cgtgctgtgt cgcagcgagt cgcgagattg ccggcctgtc gacgagttcc    780

cgtttttcct gcactggcta cagcgtagcg gcttcgcctg ctcactggcg acggtactgg    840

ccgcacggtt tcacgatcct gaggtcggcg agatgccccc gtctggcgag gccttttgtc    900

tacgcagcga cctctgtcta ctagagaaag gcagaaggcc ggagcgtttt ctcagatgtg    960

ctactctttg tcttctgcga tcttccgtgc gttcagctgt gcttttgcca aaggagacct   1020

gtgtgcagag gacttcgctg ctaaaaagca gaagagtgcg cggcgtgtgt agctcagtgg   1080

catttcggga ctcggtcttg cgtcgttcgc gactggacgt cgtcgtctgt gagagcgtca   1140

aactagggag aaggggcggg ccagagcgtt cggaaaatta tctgcaaagc ccaggtcccg   1200

tatgatattc aaaaaag                                                  1217
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<210> SEQ ID NO 4

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<211> LENGTH: 267
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 4

Met Lys Ala Ala Val Leu Thr Leu Ala Val Leu Phe Leu Thr Gly Ser
1 5 10 15

Gln Ala Arg His Phe Trp Gln Gln Asp Glu Pro Pro Gln Ser Pro Trp
20 25 30

Asp Arg Val Lys Asp Leu Ala Thr Val Tyr Val Asp Val Leu Lys Asp
35 40 45

Ser Gly Arg Asp Tyr Val Ser Gln Phe Glu Gly Ser Ala Leu Gly Lys
50 55 60

Gln Leu Asn Leu Lys Leu Leu Asp Asn Trp Asp Ser Val Thr Ser Thr
65 70 75 80

Phe Ser Lys Leu Arg Glu Gln Leu Gly Pro Val Thr Gln Glu Phe Trp
85 90 95

Asp Asn Leu Glu Lys Glu Thr Glu Gly Leu Arg Gln Glu Met Ser Lys
100 105 110

Asp Leu Glu Glu Val Lys Ala Lys Val Gln Pro Tyr Leu Asp Asp Phe
115 120 125

Gln Lys Lys Trp Gln Glu Glu Met Glu Leu Tyr Arg Gln Lys Val Glu
130 135 140

Pro Leu Arg Ala Glu Leu Gln Glu Gly Ala Arg Gln Lys Leu His Glu
145 150 155 160

Leu Gln Glu Lys Leu Ser Pro Leu Gly Glu Glu Met Arg Asp Arg Ala
165 170 175

Arg Ala His Val Asp Ala Leu Arg Thr His Leu Ala Pro Tyr Ser Asp
180 185 190

Glu Leu Arg Gln Arg Leu Ala Ala Arg Leu Glu Ala Leu Lys Glu Asn
195 200 205

Gly Gly Ala Arg Leu Ala Glu Tyr His Ala Lys Ala Thr Glu His Leu
210 215 220

Ser Thr Leu Ser Glu Lys Ala Lys Pro Ala Leu Glu Asp Leu Arg Gln
225 230 235 240

Gly Leu Leu Pro Val Leu Glu Ser Phe Lys Val Ser Phe Leu Ser Ala
245 250 255

Leu Glu Glu Tyr Thr Lys Lys Leu Asn Thr Gln
260 265

<210> SEQ ID NO 5
<211> LENGTH: 25
<212> TYPE: DNA
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 5

agtgttctcg aaaccatgct aacac 25

<210> SEQ ID NO 6
<211> LENGTH: 25
<212> TYPE: DNA
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 6

cctcttacct cagttacaat ttata 25

<210> SEQ ID NO 7

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<211> LENGTH: 968
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 7

Met Leu Ser Arg Arg Cys Lys Arg Ser Ser Ala Glu Asp Ser Gly Ala
1 5 10 15

Asn Glu Val Gly Ala Asn Ala Asp Gln Ser Lys Arg Leu Arg His Ser
20 25 30

Leu Asp Ser Ile Ile Glu Lys Gly Gly Asp Pro Val Asn His Asp Ser
35 40 45

Ala Met Leu Leu Asp Cys Ala Pro Thr Gln Thr Gly Arg Ala Phe Ala
50 55 60

Phe Leu Ser Met Pro Ala Pro Ile Glu Pro Ser Gly Asn Glu Glu Ser
65 70 75 80

Ala Pro Ala Val His Arg Asp Ser Gly Val Gly Gly Ile Asp Tyr Pro
85 90 95

Arg Pro Val Ala Ser Ile Ser Val Glu Ser Ser Ser Gln Val Val Ala
100 105 110

Pro Arg Asp Glu Asn Pro Ser Ala Ser Tyr Gln Arg Arg Gly Asp Ser
115 120 125

Pro Pro Ser Leu Arg Asn Gly Gly Asp Arg Gln Glu Arg Lys Arg Thr
130 135 140

Ala Val Ala Pro Glu Ala Asn Glu Pro Gln Asp Asn Glu Thr Lys Asn
145 150 155 160

Glu Glu Trp Leu Gln Leu Ala Arg Leu Lys Pro Lys Val Glu Gly Val
165 170 175

Cys Phe Asp Arg Phe Phe Arg Arg Trp Val Ala Lys Arg Ala Gly Leu
180 185 190

Lys Lys Val Tyr Phe Pro Val Tyr Lys Tyr Gly Phe Asp Arg Ala Tyr
195 200 205

Glu Leu Ala Val Ala Thr Arg Arg Gly Leu Glu Asn Asp Ala Ala Ala
210 215 220

Gly Ile Arg Ala Val Gly Ala Leu Arg Pro Arg Ile Ser Glu Ala Ala
225 230 235 240

Gly Cys Thr Ser Ser Pro Gly Met Leu Ser Glu Asp Ala Cys Pro Glu
245 250 255

Lys Pro Pro Val Pro Val Gln Pro Pro Arg Thr Leu Ser Thr Arg Ala
260 265 270

Thr Ala Ala Gln Ala Glu Val Lys Ser Gly Asp Ser Ala Glu Ser Thr
275 280 285

Lys Asn Asp Ser Glu Gly Ala His Val Leu Glu Gly Ala Glu Leu Gln
290 295 300

Thr Pro Glu Arg Ser Thr Ser Asn Thr Ile Cys Trp Ala Thr Ala Ala
305 310 315 320

Glu Gly Ser Ile Ser Lys Thr Asp Gly Phe Gln Asn Arg Ser Ser Pro
325 330 335

Ser Gly Phe Gly His Gly Ser Arg Asn Lys Pro Glu Leu Ser Gln Gln
340 345 350

Lys Val Glu Thr Thr Ser Arg Gly Ile Arg Ser Ala Ser Ala Ser Cys
355 360 365

Asn Arg Glu Lys Asp Gln Gly Gly Ser Ala Cys Ser Val Leu Ser Ile
370 375 380

Ala Ser Phe Ser Leu Ser Gln Ile Asp Glu Glu Leu Glu Gly Ile Asn

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385                 390                 395                 400

Asp Glu Ala Tyr Glu Ala Glu Arg Leu Gln Ala Asp Glu Asp Arg Ser
                405                 410                 415

His Ala Pro Ala Ala Ser His Gly Glu Gly Gly Thr Ser Ala Gly Glu
            420                 425                 430

Ser Thr Ala Ala Ser Thr Thr Gly Ser Glu Asp Ser Gly Pro Leu Arg
        435                 440                 445

Ala Ala Thr Ser Pro Leu Met Phe Pro Gln Gly Ser Glu Gly Ser Ala
    450                 455                 460

Ser Ser Ala Ser Thr Glu Ile Met Val Leu Asp Asp Glu Ser Met Gln
465                 470                 475                 480

Gln Ala Leu Val Thr Ala Ser Ala Glu Thr Leu Asn Lys Leu Arg Ser
                485                 490                 495

Thr Leu Pro Ala Gly Val His Phe Asp Phe Ala Ser Lys Arg Trp Phe
            500                 505                 510

Ala Val Tyr Ser Ser His Glu Ser Pro Glu Ala Thr Gln Arg Asp Pro
        515                 520                 525

Val Arg Pro Lys Glu Arg Val Arg Ile Phe Asp Pro Thr Gln Tyr Glu
    530                 535                 540

Gly Ser Met Leu Lys Ala Phe His Ala Cys Arg Ser Phe Cys Gly Ser
545                 550                 555                 560

Val Glu Ala Gly Ala Ser Asp Trp Asp Ser Val Pro Gln Leu Val Pro
                565                 570                 575

Glu Gln Arg Lys Gln Gly Glu Cys Gln Asp Thr Ser Gly Ser Ser Asp
            580                 585                 590

Gln Gly Ala Asn Arg Leu Ser Pro Thr Glu Thr Glu Asn Pro Pro Thr
        595                 600                 605

Ala Asp His Pro Arg Ser Leu Ser Ala Thr Thr Arg Pro Glu Gly Ser
    610                 615                 620

Leu Glu Gln Thr Gln His Pro Gln Arg Asn Arg Gly Ile Leu Gly Ile
625                 630                 635                 640

Gln Pro Gly Glu Thr Glu Gly Leu Gln Val Pro Ser Asn Gly His Gly
                645                 650                 655

Val Asn Ala Gly Asp Ile Glu Thr Asn Leu Leu Asp Ala Glu Phe Gly
            660                 665                 670

Ser Glu Thr Arg Ala Arg Thr Thr Ala Leu Pro His Leu Arg Arg Ser
        675                 680                 685

Gln Arg Arg Ala Asp Pro Ala Arg Ser Val His Ser Asn Thr Phe Ala
    690                 695                 700

Gly Gln Glu Leu His Gln Ser Pro Lys Pro Gly Asn Gln Thr Ser Arg
705                 710                 715                 720

Gly Glu Ser Gly Arg Ser Ser Leu Arg Arg Lys Asn Gln Val Ser Thr
                725                 730                 735

Asn Glu Lys Gly Leu Pro Gly Glu Gly Gly Cys Arg Thr Asp Glu Lys
            740                 745                 750

Ser Lys Gln Val Ser Tyr Val Ser Phe Ser Glu Pro Ile Thr Val Arg
        755                 760                 765

Tyr Gln Gln Val Pro Thr Glu Ser Ala Ser Thr Arg Gly Cys Ser Gln
    770                 775                 780

Arg Arg Pro Gln Asn Ala Glu Glu Leu Glu Asp Arg Arg Ser Pro Leu
785                 790                 795                 800

Thr Arg Gln Glu Glu Arg Thr Glu Ser Asp Pro Arg Thr Thr Ala Gly
                805                 810                 815
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Leu Cys Gln Glu Asn Pro His Pro Ser Tyr Arg Phe Leu Arg Gln Gln
            820                 825                 830

Ser Arg Glu Leu Ala Val Arg Cys Leu Leu Val Ile Phe Gly Asn Leu
        835                 840                 845

Ala Asp Val Cys Thr Pro Ala Leu Phe Arg Leu Phe Pro Gln Asp Arg
    850                 855                 860

Cys Arg Arg Val Arg Ala Val Leu Gln His Arg Asp Leu Leu Gln Ser
865                 870                 875                 880

Gly Lys His Thr Arg Val Leu Leu Ser Ala Tyr Phe Gln Leu Phe Trp
                885                 890                 895

Pro Leu Leu Glu Thr Arg Thr Leu Pro Gln His Tyr Ser Ala Asp Tyr
            900                 905                 910

Ile Arg Arg Leu Leu Asn Gly Met His Asn Val Ala Ala Met His Lys
        915                 920                 925

Ser Leu Phe Pro Glu Tyr Pro Leu Arg Gly Glu Leu Asp Asn Arg Glu
    930                 935                 940

Gly Pro Tyr Ala Phe Leu Asp Asp Thr Ala Ala Glu Gly Ile Asn Phe
945                 950                 955                 960

Phe Glu Thr Asp Phe Asp Glu Pro
                965


<210> SEQ ID NO 8
<211> LENGTH: 425
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 8

Met Ala Tyr Gln Arg Arg Arg Ala Ala Ala Cys Thr Ile Glu Val Ser
1               5                   10                  15

Arg Asp Leu Phe Ser Pro Glu Arg Asn Leu His Asp Asn Phe Ser Ser
            20                  25                  30

Leu Ala Ile Thr Met Ala Phe Arg Thr Val Ser Lys Val Leu Pro Thr
        35                  40                  45

Leu Ser His Cys Phe Pro Gly Pro Leu Ser Val Ala Ala Ser Ser Ser
    50                  55                  60

Leu Pro Gly His Ser Lys Gly Glu Glu Ser Val Pro Cys Arg Val His
65                  70                  75                  80

Arg Ser Phe Arg Leu Ser Pro Val Ala Asp His Glu Ala Glu Ala Leu
                85                  90                  95

Ser Gly Ser Gly Asn Asp Thr Ser Gly Cys Ser Gln Arg Asp Arg Phe
            100                 105                 110

Cys Ala Gly Asn Gly Ser Asp Cys Lys Ala Arg Arg Thr Ser Asp Gly
        115                 120                 125

Asn Gly Ser Pro Pro Thr Asn Ala Arg Met Ser Glu Lys Leu Ser Leu
    130                 135                 140

Phe Lys Asn His Ala Tyr Ser Cys Leu Glu Gln Arg Ala Cys Pro Ala
145                 150                 155                 160

Ser Asn Arg Asn Leu Gly Asp Thr Ala Ala Cys Pro Leu Ser Ala Phe
                165                 170                 175

Cys Arg Ser Leu Val Arg Arg Thr Pro Ser Arg Leu Trp Leu Pro Pro
            180                 185                 190

Gln Cys Ser Leu Leu Ser Gly Cys Ser Ala Arg Ser Cys Pro Pro Lys
        195                 200                 205

Ile Ser Val Arg Ala Thr Asn Gly Ala Ser Glu Gln Ala Ser Trp Gln
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    210                 215                 220

Asp Phe His Pro Ala Leu Gly Arg Ala Val Val Pro Leu Ser Leu Ser
225                 230                 235                 240

Gly Arg Gly Thr Ala Gly Thr His Leu Val Arg Lys Phe Gly Thr Gln
                245                 250                 255

Arg Val Val Gln Lys Arg Arg His Gln Met Arg Ile Leu His Pro Ala
            260                 265                 270

Gln Thr Ala Tyr Val Pro Val Glu Gln Arg Pro Pro Pro Ile Pro His
        275                 280                 285

Ser Leu Thr Ala Ser Ser Thr Val Lys Arg Leu Leu Asn Asn Asn Thr
    290                 295                 300

Val Ala Ala Lys Glu Ala Ala Lys Arg Ile Asn Trp Gly Ala Tyr Ile
305                 310                 315                 320

Ser His Gln Arg Gly Val Arg Trp His Pro Gln Gly Ala Trp Arg Val
                325                 330                 335

Gln Phe Ser Arg Arg Asn His Glu Arg Asn Phe Phe Val Arg Cys Glu
            340                 345                 350

Cys Tyr Phe Arg Val Gly Thr Tyr Gly Phe Gln Met Ala Lys Asp Leu
        355                 360                 365

Ala Ile Arg Tyr Arg Gln Arg Leu Glu Lys Glu Trp Glu Glu Leu Gln
    370                 375                 380

Glu Gln Trp Thr Lys Leu Asp Ile Leu Glu Ala Glu Gln Arg Ala Lys
385                 390                 395                 400

Tyr Lys Glu Lys Arg Glu Glu His Leu Leu Leu Gly Ala Gly Glu Glu
                405                 410                 415

Pro Glu Leu His Ser Arg Arg Ser Lys
            420                 425


<210> SEQ ID NO 9
<211> LENGTH: 2674
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 9

Met Asp Gly Ser Gly Glu Ser Ser Gly His Leu Phe Lys Pro Gly His
1               5                   10                  15

Gly Glu Ala Arg Val Ser Val His Arg Gly Ser Leu Thr Asp Ser Gly
            20                  25                  30

Ser Leu Pro Ala Ala Ser Arg Cys His Ser Gln Asp Asn Lys Leu Ser
        35                  40                  45

Leu Pro Cys Ala Gly Ser Met Leu Pro Ala Ser Ser Gly Arg Phe Ser
    50                  55                  60

Cys Asp Ser Ala Leu Phe Gly Gly Pro Val Asp Ser Ala Cys Ser Ser
65                  70                  75                  80

Asp Trp Thr Pro Val Val Ser Pro Ser Arg Asp Leu Ser Ala Asp Gly
                85                  90                  95

Thr Asp Ser Ser Ser Val Ser Gly Ser Arg Gly Ser Ser Leu Pro Phe
            100                 105                 110

Gly Ser Pro Thr Ser Ala Leu Leu Arg Pro Ser Ser Glu Ala Ser Ala
        115                 120                 125

Asn Phe Pro Arg Leu His Lys Ser Val His Ala Leu Asp Asp Lys Met
    130                 135                 140

Arg Gly Leu Asp Ala Gln Leu Tyr Val Arg Pro His Gln Thr Leu Pro
145                 150                 155                 160
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Leu Gln Pro Arg Leu Arg Glu Thr Asp Leu Cys Arg Asn Gly Glu Asp
                165                 170                 175

Gly Arg Pro Gly Lys Phe Asp Ser Pro His Leu Gly Ser Ser Ala Gly
            180                 185                 190

Pro Tyr Gly His Ser Phe Leu Ala Asn Pro Gln Leu Thr Pro Phe Val
        195                 200                 205

Pro Gln His Leu Ser Ser Ser Pro Pro Gln Pro Val Leu Ser Pro Pro
    210                 215                 220

Gly Glu Glu Gly Arg Asn Ser Ala Ala Phe Gly Lys Thr Val Ser Arg
225                 230                 235                 240

Leu Asn Thr Gly Gly Gly Glu Arg Gln Asp Ser Ser Glu Asp Gln Val
                245                 250                 255

Gly Gly Thr Gly Arg Gln Ser Asp Gln Ala Thr Lys Ala Asn Ser Gly
            260                 265                 270

Ser Thr Pro Ala Gly Cys Ala Gln Thr Ala Gly Leu Leu Thr Asp Val
        275                 280                 285

Gln Ser Ser Gly Thr Asn Val Glu His Gly Arg Glu His Phe Ser Thr
    290                 295                 300

Pro Gln Lys Pro Ala Asp Gly Ser Ala Arg Thr Cys Gly Phe Arg Glu
305                 310                 315                 320

Thr Arg Val Ser Pro Ser Asn Ser Ser Leu Pro Arg Thr Ala Cys Arg
                325                 330                 335

Ser Arg Leu Asp Ala Phe Leu Pro Gln Lys Ser Val Ser Pro Asp His
            340                 345                 350

Glu His Val Arg Gly Thr Gly Gly Ala Arg Ala Phe Val Gly Gly Asp
        355                 360                 365

Ser Pro Phe Pro Glu Lys Pro Asp Thr Leu Pro Ala Thr Val Thr Ala
    370                 375                 380

Glu Leu Ala Thr Glu Ala Pro Pro Ala Ser Arg Asp Pro Pro Val Glu
385                 390                 395                 400

Glu Phe Pro Gly Ala His Glu Leu Glu Ser Leu Pro Pro Pro His Val
                405                 410                 415

Asn Ser Gly Arg Pro Pro Ile Gly Glu Lys Asp Gly Ala Ala Ala Ser
            420                 425                 430

Pro Gly Val Ser Arg Leu Pro Ser Gln Glu Arg Val His Thr Leu Leu
        435                 440                 445

Tyr Pro Asn Glu Lys Asp Ala Ser Ser Leu Ser Arg Cys Cys Pro Ser
    450                 455                 460

Ser Met Gln Pro Pro Pro Ala Gly Pro Arg Gln Glu Glu Ala Arg Ser
465                 470                 475                 480

Phe Ser Val Ser Ala Ala Ser Ala Pro Gly Ala Pro Pro Gly Ile Val
                485                 490                 495

Tyr Gln Ala Ser Ala Cys Ala Ser Pro Ala Thr Val Ala Ser Phe Ala
            500                 505                 510

Thr Pro Leu Thr Thr Pro Val Gly Ala Ser Ala Gln Ser Glu Pro Ala
        515                 520                 525

Ala Leu His Ala His Ser Arg Ser Arg Thr Gly Ala His Pro Glu Ala
    530                 535                 540

Leu Pro Pro Gly Val Pro Gly Val Thr Ser Gln Leu Gly Arg Gly Ala
545                 550                 555                 560

Arg Gly Asp Arg Glu Thr Leu Ala Gly Gly Ala Arg Pro Gly Gln Asp
                565                 570                 575

Gly Val Cys Glu Arg Arg Gly Asp Val Ala Arg Gly Arg Leu Gly Gly
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           580                 585                 590

Val Ser Val Ala Gly Asp Glu Ala Ala Glu Gly Thr Ser His Lys Ala
        595                 600                 605

Ala Leu Glu Gly Ala Tyr Val Gln Asp Gly Cys Ser Pro Gln Pro Leu
    610                 615                 620

Asn Pro His Ala Pro Ser Gly Ile Ser Ala Pro Thr Asn Gly Ser Ser
625                 630                 635                 640

Glu Leu Ala Ser Ser Ala Ile Pro Ala Ser Thr Cys His Asp Ala Phe
                645                 650                 655

Val Arg Ser Pro Val Ser Gly Ser Asp Cys Met Ser Val Ala Asn Pro
            660                 665                 670

Gly Gly Pro Pro Gly Ala Leu Gly Gly Leu Phe Pro Ser Pro Arg Gly
        675                 680                 685

Pro Ser Gly Pro Arg Pro Thr Pro His Pro Ala Gln Met Ala Phe Ala
    690                 695                 700

Phe Val Gly Gln Gln Pro Val Phe Pro Gly Phe Asp Ala Ser Gln Pro
705                 710                 715                 720

Ala Gly Ser Thr Phe Gln Tyr Pro Pro Ile Arg Gly Ala Val Ser Gly
                725                 730                 735

Val Ser Pro Pro Pro Pro Met His Pro Ser Ser Phe Ala Gln Pro Val
            740                 745                 750

Trp Ser Pro Thr Ser Val Pro Ser Ser Ser Val Ser Ser Ser Val Ser
        755                 760                 765

Ser Ser Gly Val Ser Ser Ser Ala Pro Pro Pro Leu Ala Val Gly Phe
    770                 775                 780

Gln Asn Pro Cys Pro Trp Arg Pro Thr Ala Pro Arg Asp Arg Ser Glu
785                 790                 795                 800

Gly Gly Ala Gly Ser Pro Gly Val Ser Cys Gly Ser Ala Pro Pro Ala
                805                 810                 815

Pro Thr His Pro Thr Gly Lys Gly Gly Ala Ala Gly Arg Ala Gly Lys
            820                 825                 830

Gln Leu Gly Gln Ala Thr Arg Phe Leu Ser Ser Val Ser Gly Val Val
        835                 840                 845

Tyr Asp Lys Gly Gly Glu Lys Trp Ile Ala Arg Trp Ser Glu Asn Gly
    850                 855                 860

Lys Pro Phe Lys Lys Thr Phe Ala Val Gly Lys His Gly Phe Asp Ala
865                 870                 875                 880

Ala Arg Lys Met Ala Glu Asp Cys Arg Leu Gln Ala Leu Tyr Ala Lys
                885                 890                 895

Arg Trp Asn Ser Ala Ser Gly Leu Pro Ala Ser Phe Ser Lys Ser Asn
            900                 905                 910

Ser Leu Gly Arg Ser Thr Pro Gly Asp Arg Gly Lys Thr Glu Ser Thr
        915                 920                 925

Asn Ser Ala Lys Cys Lys Arg Asp Thr Ser Gly Glu Ser Gly Cys Thr
    930                 935                 940

Asp Thr Gly Leu Arg Ser Leu His Met Gly Gly Ala Gly Asp Leu Ser
945                 950                 955                 960

Ser Leu Gly His Pro Gly Thr Pro Pro Arg Asp Gln Glu Gly Ala Pro
                965                 970                 975

Ala Ser Phe Leu Leu Glu Gly Thr Gly Val Val Arg Ser Ser Gln Val
            980                 985                 990

Gln Thr Pro Phe Arg Leu Tyr Asp  Ser Val Pro Ser Pro  Leu Arg Ser
        995                 1000                1005
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Gly Asp  Ala Leu Gly Ala Gln  Arg Gly Leu Val Pro  Gln Leu Leu
    1010                1015                1020

Asn Asn  Ala Leu Val Gly Val  Pro Phe Ala Pro Pro  Pro Gly Ala
    1025                1030                1035

Ser His  Ser Gly Cys Ser Ala  Ala Leu Pro Pro Gly  Pro Gly Ala
    1040                1045                1050

Pro Val  Gln Val Ser Ser Pro  His Thr Gly Phe Val  Ala Pro Ala
    1055                1060                1065

Asp Val  Asp Ala Pro Pro Arg  Asp Gly Leu Glu Gly  Leu Gly Gly
    1070                1075                1080

Ala Ala  Glu Val Ser Pro Gln  Ile Ala Val Gln Asp  Gly Gly Lys
    1085                1090                1095

Lys Gly  Glu Gly Leu Leu Gly  Ser Ala Ser Leu Ser  Val Arg Arg
    1100                1105                1110

Arg Arg  Lys Arg Glu Pro Asp  Glu Lys Phe Ser Pro  Gly Glu Ser
    1115                1120                1125

Asn Ala  Ala Val Lys Lys Thr  Pro Arg Pro Gly Ser  Phe His Pro
    1130                1135                1140

His Ser  Cys Pro Gly Ser Glu  Gly Phe Arg Ser His  Asp Gly Pro
    1145                1150                1155

Gly Asp  Ser Thr Glu Ala Arg  Cys Ala Gly Leu Pro  Ala Phe Gln
    1160                1165                1170

His Ala  Thr Ala Pro Ser Ser  Val Cys Trp Pro Ser  Thr Ala Ser
    1175                1180                1185

Leu Pro  Ser Leu Asp Lys Ala  Gly Gln Arg Ala Glu  His Ala Gly
    1190                1195                1200

Pro Ser  Ala Phe Ser Ser Phe  Ser Ser Val Gln Gln  Ser Pro Gly
    1205                1210                1215

Ser Val  Glu Thr Trp Arg Pro  Glu Gly Asp Gly Gly  Pro Ala Ser
    1220                1225                1230

Pro Ala  Arg Asp Ala Gly Arg  Arg Gly Ala Glu Ser  Glu Glu Arg
    1235                1240                1245

Glu Thr  Gly Glu Leu Ala Gly  Pro Phe Ala Gly Val  Ser Ala Ser
    1250                1255                1260

Ala Gly  Ser Ala Ser Arg Lys  Gly Gln Gln Lys Gln  Leu Thr Arg
    1265                1270                1275

Gln Ile  Gln Arg Gln Gln Gln  Leu Tyr Arg Gln Gln  Glu Ala Leu
    1280                1285                1290

Leu Gln  Asn Gln Glu Glu Leu  Phe Ser Arg Leu Leu  Arg Arg Arg
    1295                1300                1305

Ser Arg  Gln Glu Arg Ser Asp  Val Arg Arg Arg Met  Gln Arg Asp
    1310                1315                1320

Val Ser  Ser Leu Arg Arg Leu  Pro Ala Met Leu Leu  Ser Pro Leu
    1325                1330                1335

Arg Asp  Thr Leu Val Ala Ser  Ala Ala Arg Leu Pro  Leu Ala Thr
    1340                1345                1350

Arg Gly  Thr Lys Arg Glu Ser  Gln Lys Glu Arg Arg  Asp Cys Gly
    1355                1360                1365

Ala Gly  Ile Gly Gly Glu Thr  Ala Ser Glu Lys Lys  Glu Met Ala
    1370                1375                1380

Glu Pro  Val Arg Val His Arg  Arg Asp Arg Gly Gly  Ala Arg Asp
    1385                1390                1395
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Glu Glu  Lys Pro Ser Thr Glu  Gly Val Arg Gln Ala  Asp Pro Lys
    1400                1405                1410

Gly Arg  Lys Ala Glu Gly Phe  Pro Thr Trp Val Ile  Pro Pro Asn
    1415                1420                1425

Glu Glu  Leu Lys Ala Ala Gln  Val Leu Arg Ala Leu  Arg Val Gln
    1430                1435                1440

Arg Arg  Ala Ala Ala Arg Glu  Gly Lys Leu Leu Glu  Ser Leu Leu
    1445                1450                1455

Val His  Arg Gly Glu Gly Glu  Gly Thr Phe Ser Glu  Glu Thr Glu
    1460                1465                1470

Gly Asn  Thr Gly Ile Glu Asp  Ala Gly Thr Glu Ser  Asp Ala Thr
    1475                1480                1485

Val Thr  Gln Glu Thr Ala Glu  Lys Val Val Glu Asn  Val Gln Lys
    1490                1495                1500

Met Glu  Glu Leu Glu Ser Lys  Val Glu Lys Glu Asn  Glu Arg Arg
    1505                1510                1515

Arg Glu  Ala Glu Asp Glu Thr  Pro Lys Gln Ser Ser  Glu Glu Ala
    1520                1525                1530

Pro Gly  Val Gln Gln Ser Pro  His Lys Leu Ser Thr  Asn Asn Glu
    1535                1540                1545

Asn Asp  Ala Ser Pro Gln Lys  Leu Thr Lys Ser Val  Arg Phe Ala
    1550                1555                1560

Glu Ser  Val Ala Gly Ser Ser  Ser Ala Val Gln Thr  Ala Gly Ala
    1565                1570                1575

Ala Asp  Glu Glu Pro Leu Ala  Thr Glu Thr Leu Glu  Gly Arg Arg
    1580                1585                1590

Val Gly  Gly Ile Pro Val Pro  Ala Thr Ser Ser Pro  Ala Pro Val
    1595                1600                1605

Phe Pro  Cys Thr Ala Ala Gln  Leu Gly Asp Leu Cys  Met Asp Thr
    1610                1615                1620

Leu Tyr  Ala Leu Gly Thr Val  Arg Pro Gln Trp Arg  Arg Gln Asp
    1625                1630                1635

His Arg  Arg Ala Phe Gly Trp  His Leu Ser Gln Ile  Lys Pro Asp
    1640                1645                1650

Leu Ile  Leu Pro Ser Leu His  Ala Ser Arg Val Leu  Arg Arg Leu
    1655                1660                1665

Ser Pro  Arg Pro Ser Asn Ala  Val Glu Phe Pro Arg  Glu Glu Leu
    1670                1675                1680

Ala Ala  Ala Ser Ser Ala Ala  Gly Leu Val Tyr Gly  Glu Gly Leu
    1685                1690                1695

Ser Ser  His His Thr Leu Arg  Ser Tyr Val Asp Ala  Phe Arg Pro
    1700                1705                1710

Leu Phe  Ser Ser Pro Ser Ser  Pro Pro Leu Glu Phe  Leu His Leu
    1715                1720                1725

Ser Ser  Gly Asp Leu Leu Met  Ser Leu Trp Gln Leu  Glu Glu Gly
    1730                1735                1740

Gly Arg  Ala Ala Val Ile Asp  Asn Val Leu Leu Ala  Leu Asp Ala
    1745                1750                1755

Leu Tyr  Glu Arg His Thr Gly  Arg Arg Leu Arg Gly  Thr Ala Pro
    1760                1765                1770

Pro Pro  Phe Ala Val Ser Ser  Pro Ser Ser Ala Pro  Ser Ser Leu
    1775                1780                1785

Phe Ala  Leu Ala His Leu Gln  Gly Gly Ala Thr Ser  Thr Thr Pro
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    1790                1795                1800

Leu Pro  Ala Thr Ala Leu Pro  Ser Pro Pro Phe Pro  Arg Val Ser
    1805                1810                1815

Ser Ala  Pro Asp Ser Pro Val  Phe Ala Pro Asp Ala  Ser His Gly
    1820                1825                1830

Pro Ser  Gln Arg Arg Gln Val  Ser Pro His Val Thr  Phe Glu Thr
    1835                1840                1845

Pro Pro  Thr His Pro Arg Asp  Arg Asp Ser Glu Thr  Ser Val Glu
    1850                1855                1860

Arg Asn  Ala Ser Pro Glu Ala  Ser Pro Gln Ala Ala  Thr Leu Ala
    1865                1870                1875

Ala Pro  Ala Pro Cys Asp Gly  Asp Arg Glu Glu Asn  Phe Val Leu
    1880                1885                1890

Ala Tyr  Asn Pro Glu Ala Lys  Ala Leu Arg Gln Val  Asn Phe Leu
    1895                1900                1905

Ala Val  Gly Val Arg Val Phe  Leu His Leu Glu Val  Val Glu Glu
    1910                1915                1920

Met Leu  His Leu Gln Ala Lys  Met Gln Arg Thr Pro  Gly Arg Asp
    1925                1930                1935

Asp Arg  Ala Thr Ala Ser Ser  Gly Pro Ser Val Asp  Asp Gly Ser
    1940                1945                1950

Gly Leu  Met Thr Ser Leu Pro  Ser Thr Cys Ser Gly  Val Ser Gly
    1955                1960                1965

Lys Lys  Asp Pro Met His Trp  Ser Ala Leu Phe Val  Thr Val Pro
    1970                1975                1980

Ala Pro  Ser Val Ser Thr Ala  Ala Ser Lys Pro Leu  Phe Val Val
    1985                1990                1995

Ala Glu  Met Val Asp Arg Arg  Leu Gln Val Pro Cys  Gly Glu Gln
    2000                2005                2010

Leu Leu  Phe Arg Pro Leu Pro  Leu Ser Pro Ala Ala  Pro Ser Ala
    2015                2020                2025

Leu Leu  Ala Phe Ala Pro Ala  Arg Val Cys Gln Leu  Leu Arg Ala
    2030                2035                2040

Gly Ala  Met Cys Leu Thr Arg  Phe Thr Glu Lys Glu  Gly Gly Lys
    2045                2050                2055

Arg Pro  Arg Gly Ser Ala Gln  Arg Cys Ser Ala Ala  Ser Ser Phe
    2060                2065                2070

Phe Tyr  Ser Pro Pro Pro Leu  Asp Leu Ser His Leu  Ala Ser Phe
    2075                2080                2085

Ala Pro  Ala Ala Ser Thr Leu  Thr Pro Pro Ser Ser  Pro Ala Ser
    2090                2095                2100

Ser Pro  Ser Ala Ser Ala Ser  Gln Thr Gly Pro Gly  Arg Ala Lys
    2105                2110                2115

Ser Arg  Gly Thr Ser Pro Val  Gly Pro Glu Ser Pro  Glu Ala Ala
    2120                2125                2130

Ser Thr  Thr Ser Asp Gly Leu  Ala Val Pro Gly Ser  Ala Ser Ala
    2135                2140                2145

Val Ser  Thr Pro Gly Val Pro  Ala Gly Ala Ser Gly  Ala Ser Leu
    2150                2155                2160

Gly Ala  Pro Ala Pro Ser Pro  Met Ala Ser Pro Gly  Gly Ser Pro
    2165                2170                2175

Gly Arg  Pro Pro Lys Pro Val  Cys Cys Pro Ala Ala  Pro Gly Ile
    2180                2185                2190
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Glu Thr  Ala Trp Arg Cys Lys  Cys Ser His Arg Arg  His Glu Leu
    2195                 2200                 2205

Gln Leu  Glu Ile Lys Gln Lys  Leu Arg Gln Asp Lys  Lys Arg Cys
    2210                 2215                 2220

Leu Ala  Leu Ile Arg Glu Tyr  Pro Asp Leu Ser Leu  Leu Val Gly
    2225                 2230                 2235

Ala Pro  Pro Ala Thr Pro Arg  Glu Lys Glu Thr Gly  Ala Lys Arg
    2240                 2245                 2250

Gln Ala  Pro Glu Gly Arg Arg  Thr Ala Thr Pro Ser  Gly Ser Gly
    2255                 2260                 2265

Thr Leu  Thr Ala Lys Gly Gly  Asp Leu Gln Gly Ser  Thr Pro Ser
    2270                 2275                 2280

Gly Ala  Gly Leu Leu Ser Leu  Ala Arg Thr Ser Gln  Leu Glu Met
    2285                 2290                 2295

Leu Ala  Tyr Leu Val Glu Val  Asp Pro Trp Lys Tyr  Ala Lys Asn
    2300                 2305                 2310

Arg Gln  Asp Ala Pro Lys Pro  Glu Glu Ile Pro Gly  Leu Leu Ala
    2315                 2320                 2325

Lys Tyr  Lys Ala Ala Val Arg  Thr Ala Glu Tyr Gly  Arg Met Leu
    2330                 2335                 2340

Gln Lys  Trp Arg Ala Gly Gln  Ser Arg Glu Asp Glu  Gly Arg Gly
    2345                 2350                 2355

Gly Ala  Asp Gly Arg Lys Glu  Gly Asp Gly Leu Leu  Ser Pro Thr
    2360                 2365                 2370

Ala Ser  Pro Pro Ser Arg Arg  Lys Gln Gly Lys Asp  Ser Ser Pro
    2375                 2380                 2385

Asn Ser  Ala Ser Ser Gln Ala  Ser Gly Pro Ala Pro  Ser Pro Ser
    2390                 2395                 2400

Leu Ser  Pro Gly Ala Gly Ala  Ala Ala Val Leu Glu  Ala Glu Lys
    2405                 2410                 2415

Pro Glu  Pro Gln Ser Pro Gln  Glu Ser Pro Cys Pro  Leu Glu Pro
    2420                 2425                 2430

Ala Ala  Gly Gln Glu Pro Arg  Ala Thr Ser Ser Ala  Leu Pro Ala
    2435                 2440                 2445

Gly Ser  Pro Pro Trp Ala Leu  Pro Leu Val Pro Pro  Gly Gly Ser
    2450                 2455                 2460

Pro Arg  Ala Ser Val Ser Pro  Ser Val Leu Glu Glu  Leu Leu Arg
    2465                 2470                 2475

Ile Gln  Thr Ala Met Ser Gln  Leu Ala Ile Gly Thr  Ala Ile Cys
    2480                 2485                 2490

Val Arg  Val Lys Ala Leu Leu  Gly Leu Pro Ala Gly  Ala Glu Gln
    2495                 2500                 2505

His Ile  Arg Gly Val Val Thr  Arg Asn Ala Leu Lys  Phe Pro Trp
    2510                 2515                 2520

Glu Lys  Pro Ala Ala Pro Gln  Val Gln Ala Ala Gly  Pro Ser Val
    2525                 2530                 2535

Gly Ala  Ser Arg Thr Ser Pro  Ser Arg Arg Leu Ser  Gly Gly Val
    2540                 2545                 2550

Leu Pro  Gly Asp Glu Ala Gly  Glu Arg Arg Glu Lys  Gly Gly Ala
    2555                 2560                 2565

Arg Arg  Gly Val Ala Glu Gly  Asp Thr Glu Lys Lys  Glu Asp Glu
    2570                 2575                 2580
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Gly Thr  Ala Leu Cys Ala Gly  Ser Arg Glu Thr Glu  Ala Asp Gly
    2585                2590                2595

Ala Gly  Tyr Leu Thr Leu Ser  Leu Asn Asn Arg Lys  Glu Glu Phe
    2600                2605                2610

Ile Leu  Ser Phe Arg Glu Val  Gln Cys Leu Val Ala  Gln Asp Asp
    2615                2620                2625

Leu Arg  Leu Val Arg Thr Arg  Ala Arg Gln Trp Val  Ser Ser Phe
    2630                2635                2640

Gly Pro  Gln Pro Ser Ala Asp  Arg Lys Gly Glu Arg  Glu Glu Glu
    2645                2650                2655

Lys Glu  Thr Gly Gly Arg Thr  Arg Lys Phe Val Val  Asp Glu Asp
    2660                2665                2670

Phe


<210> SEQ ID NO 10
<211> LENGTH: 2282
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 10

Met Asp Ser Gly Arg Asp Val Lys Asp Gly Thr Ala Gly Ser Arg Pro
1               5                  10                  15

Lys Asp Gly Gly Asp Gly Val Gly Glu Thr Arg Ala Ser Gly Arg Glu
            20                  25                  30

Ser Glu Glu Arg Ser Leu Gln Leu Glu Ala Asn Glu Cys Pro Gly Ala
        35                  40                  45

Val Met Ser Arg Arg Glu Asp Glu Ala Glu Pro Gln Ser Ser Pro Ser
    50                  55                  60

Ser Ser Pro Pro Arg Glu Glu Gly Pro Gln Asn Val Asp Asp Ala Asp
65                  70                  75                  80

Thr Ala Asn Gly Ser Gly Glu Ala Gly Leu Gln Arg Pro Pro Gln Lys
                85                  90                  95

Arg Arg Leu Glu Gln Gly Leu Glu Ala Glu Ala Gly Val Gly Ser Ser
            100                 105                 110

Arg Val Glu Glu Val Glu Ala Val Cys Arg Lys Arg Pro Ala Phe Ser
        115                 120                 125

Gly Val Ala Asp Ala Phe Leu Glu Arg Pro Val Thr Leu Lys Asn Ser
    130                 135                 140

Ser Glu Glu Asp Ala Ala Arg Leu Ser Gly Asp Asp Ala Ala Gly Ala
145                 150                 155                 160

Ser Leu Leu Ser Val Arg Ser Ala Gly Ala Leu Thr Gly Asp Phe Pro
                165                 170                 175

Ser Ser Ser Ser Arg Leu Pro Ala Met Leu Ser Gly Ala Arg Gly Glu
            180                 185                 190

Asn Ala Glu Glu Ser Val Arg Asp Ala His Thr Pro Ala Gln Asp Ser
        195                 200                 205

Arg Asp Ser Ala Leu Ala Ser Phe Ala Pro Thr Leu Ala Pro Gly Gln
    210                 215                 220

Glu Ser Glu Tyr Thr Arg Ala Lys Leu Trp Ser Ile Glu Lys Ala Phe
225                 230                 235                 240

Asp Ala Phe Leu Ala Asp Gln Lys Ala Asn Gly Arg Arg Gln Gly Ser
                245                 250                 255

Arg Ser Met Arg Glu Pro Ser His Ala His Pro Gly Leu Ser Ala Glu
            260                 265                 270
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Arg Glu Thr Ser Ser Gly Ala Ser Ala Ala Thr Ser Asp Leu Ser Arg
        275                 280                 285

Glu Asp Val Glu Glu Leu Phe Arg Gln His Gly Val Ser Pro Arg Glu
    290                 295                 300

Leu Val Arg Met Leu Ser Gly Arg Arg Asp Gly Pro Gly Thr Ser Pro
305                 310                 315                 320

Glu Glu Leu Arg Ala Ala Val Ala Trp Ala Arg Gln Leu Phe Pro Ala
                325                 330                 335

Ala Pro Arg Ser Pro Ser Glu Leu Arg Met Tyr Leu Gln Arg Ala Val
            340                 345                 350

Leu Asp Arg Gln Lys Arg Leu Arg Glu Arg Trp Gly Ala Glu Ala Asn
        355                 360                 365

Pro Cys Gly Asp Ala Ser Val Tyr Gly Asp Glu Lys Leu Arg Glu His
    370                 375                 380

Leu Ser Asp Leu Ser Ala Phe Met Pro His Leu Asp Ala Gly Arg Glu
385                 390                 395                 400

Val Tyr Met Gln Trp Gln Arg Ser Arg Gly Arg Arg Asp Phe Asp Ala
                405                 410                 415

Phe Val Arg Pro Pro Gly Leu Thr Pro Phe Arg Asp Ser Ser Ser Arg
            420                 425                 430

Gln Gly Asp Phe Ala Ala Ser Pro Leu Tyr Ser Phe Ser Ser Arg Thr
        435                 440                 445

Pro Trp Ala Ser Ala Cys Lys Glu Ala Ser Thr Pro Pro Ala Ala Lys
    450                 455                 460

Gln Gln Ala Pro Pro Pro Ser Leu Trp Asn Leu Pro Asn Arg Pro Gln
465                 470                 475                 480

Pro Tyr Thr Leu Ala Asp Val Gln Glu Ala Met Glu Gly Pro Glu Gly
                485                 490                 495

Val Leu Arg Val Ala Arg Pro Leu Thr Gly Phe Gly Glu Asp Ala Glu
            500                 505                 510

Ser Leu Ser Phe Ala Ser Leu Pro Lys Gly Ala Glu Thr Leu Phe Trp
        515                 520                 525

Ser Ser Gly Arg Gly Leu Tyr Phe Leu Arg His Leu Glu Arg Thr Lys
    530                 535                 540

Ala Gly Glu His Asp Val Val Gly Glu Ala Gly Val Trp Val Ala Ala
545                 550                 555                 560

Ser Glu Glu Glu Phe Gly Gly Phe Ile Ile His Arg Lys Phe Ser Val
                565                 570                 575

Ala Lys Phe Gly Phe Glu Arg Ala Lys Met Leu Ala Cys Arg Trp Tyr
            580                 585                 590

Asn Asp Arg Gln Glu Ala Arg Arg Gly Gln His Ala Leu Pro His Arg
        595                 600                 605

Glu Lys Pro Lys Gly Ile Met Ser Ser Asp Arg Pro Leu Ser Arg Glu
    610                 615                 620

Ala Ala Pro Glu Ala Ser Arg Phe Ser Ala Ser Arg Ala Gly Glu Leu
625                 630                 635                 640

Ser Gly Lys Ala Gln Glu Ala Pro Lys Ser Thr Gly Gly Thr Ala Ala
                645                 650                 655

Glu His Pro Arg Ala Ser Gln Lys Cys Arg Val Met Asp Thr Thr Cys
            660                 665                 670

Pro Val Pro Gly Val Arg Tyr Asp Ser Arg Asp Arg Ala Trp Leu Ala
        675                 680                 685

Thr Trp His Asp Gly Val Arg Gln Tyr Lys Arg Cys Phe Ser Ile Lys
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    690                 695                 700

Lys Tyr Gly Phe Ala Lys Ala Lys Glu Cys Ala Ile Arg Met Lys Met
705                 710                 715                 720

Ser Leu Val Gly Gln Pro Gly Val Ser Gln Ser Gly Arg Gln Ala Pro
                725                 730                 735

Phe Pro Val Arg Pro Phe Thr Ser Arg Ala Cys Ser Pro Leu Gln Asp
            740                 745                 750

Phe Phe Arg Glu Gly Asp Arg Arg Val Ala Ala Ser Ser Phe Ser Leu
        755                 760                 765

Leu Pro Ser Gly Arg Gly Glu Pro Arg Gly Ser Leu Gly Ser Ser Gln
    770                 775                 780

Gly Ala Asp Asp Glu Arg Ser Lys Pro Gln Ser Cys Arg Gly Leu Val
785                 790                 795                 800

Glu Gln Leu Leu Ala Arg Phe Gln Asp Ser Glu Gly Phe Thr Arg Gly
                805                 810                 815

Leu Pro Gly Asp Asp Glu Asn Arg Gly Lys Arg Leu Ser Lys Gln Ala
            820                 825                 830

Gln Asp Asp Phe Gln Ser Trp Arg Pro Pro Pro Gly Ala Arg Phe Gly
        835                 840                 845

Ser Ala Ala Gln Ala Ser Arg His Ser Thr Asp Glu Val Gly Gly Phe
    850                 855                 860

Ala Gly Phe Pro Gly Phe Ala Ala Ser His Cys Gly Glu Lys Pro Gly
865                 870                 875                 880

Gly Glu Gly Pro Ser Phe Leu Gln Lys Ser Gly Phe Val Gln Glu Asn
                885                 890                 895

Ala Phe Ser Pro Pro Ser Glu Arg Phe Glu Thr Gly Val His Arg Arg
            900                 905                 910

Val Pro Ser Leu Ser Ser Glu Leu Ala Asn Pro Gln Val Thr Glu Glu
        915                 920                 925

Val Glu Glu Phe Leu Phe Ser Leu Ser Thr Arg Ala Arg Gln Ser Leu
    930                 935                 940

Leu Ala Ser Leu Arg Arg Gly Ala Glu Asp Ser Arg Arg Ser Ala Trp
945                 950                 955                 960

Pro Gly Ala Ser Arg Asp Cys His Thr Gly Ala Gly Thr Pro Gly Gly
                965                 970                 975

Thr Asp Val Ala Asp Arg Arg Ala Thr Arg Glu Thr Arg Arg Asp Arg
            980                 985                 990

Glu Gly Glu Glu Ser Thr Ser Glu  Asp Gly Thr Val Arg  Arg Glu Thr
        995                 1000                1005

Asp Ala  Gly Ala Val Ser Pro  Asp Glu Val Ser Arg  Ala Asn Ala
    1010                1015                1020

Glu Glu  Arg Ala Ala Gly Glu  Lys Thr Arg Ser Ser  Glu Arg Ile
    1025                1030                1035

Trp Thr  Gly Glu Gly Glu Arg  Ser Ala Gly Asp Arg  Asp Asp Lys
    1040                1045                1050

Gly Glu  Gly Glu Gly Gly Gly  Gly Val Val Glu Gly  Arg Thr Glu
    1055                1060                1065

Lys Gly  Gly Asp Asp Asp Lys  Lys Pro Gly Glu Glu  Glu Ser Ala
    1070                1075                1080

Glu Arg  Glu Glu Glu Leu Lys  Asn Asp Ala Tyr Ala  Tyr Phe Thr
    1085                1090                1095

His Leu  Thr Asn Arg Glu Trp  Asp Leu Leu Asp Tyr  Leu Asp Thr
    1100                1105                1110
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Leu Asp  Phe Glu Thr Val Asp  Leu Asp Ala Val Met  Pro Phe Ile
    1115                 1120                 1125

Asn Gln  Val Pro Lys Val Arg  Gly Val Cys Phe Asp  Arg Lys Gly
    1130                 1135                 1140

Leu Tyr  Trp Ile Ser Gln Trp  His Ser Gln Gln Lys  Lys His Arg
    1145                 1150                 1155

Glu Trp  Phe Gly Val Lys Arg  Leu Gly Phe Arg Lys  Ala Trp Ala
    1160                 1165                 1170

Leu Ala  Val Cys Val Arg Arg  Asp Ala Glu Lys Val  Glu Asp Glu
    1175                 1180                 1185

Pro Val  Asp Tyr Pro Lys Leu  Pro Asp Tyr Glu Glu  Val Leu Gly
    1190                 1195                 1200

Val Thr  Tyr Ala Arg Phe Ala  Ser Gly Arg Tyr Trp  Val Ala His
    1205                 1210                 1215

Tyr Met  Arg Pro Ala Ala Pro  Ser Ser Gly Cys Leu  Gly Ser Val
    1220                 1225                 1230

Gly Arg  Lys Leu Phe Pro Val  Ser Glu Ser Ser Phe  Glu Glu Ala
    1235                 1240                 1245

Arg Ser  Gln Ala Val Ala Val  Ala Thr Ala Phe Pro  Leu Pro Leu
    1250                 1255                 1260

Ala Phe  Phe Val Asp Pro Glu  Arg Arg Ala Thr Ser  Ala Phe Glu
    1265                 1270                 1275

Ser Ala  Arg Ala Glu Asn Leu  Gln Gly Asp Lys Gln  Val Leu Leu
    1280                 1285                 1290

Ser Lys  Asn Cys Leu Phe Asn  Val Phe Thr Trp Leu  Asn Gly Gly
    1295                 1300                 1305

Ala Ser  Trp Thr Asn Val Arg  Arg Trp Ala His Ala  Lys Arg Met
    1310                 1315                 1320

Gln Leu  Ala Glu Asp Asp Trp  Pro Gln Gln Phe Phe  Ser Leu Pro
    1325                 1330                 1335

Ser Pro  Ala Lys Gly Asp Ser  Phe Ala Glu Ala Glu  Lys Glu Arg
    1340                 1345                 1350

Ala Glu  Glu Arg Thr Gly Gly  Glu Glu Val Lys Ala  Asn Ser Ala
    1355                 1360                 1365

Ser Arg  Ala Ala Ala Lys Ser  Glu Trp Pro Val Ala  Ser Thr Thr
    1370                 1375                 1380

Ser Pro  Ala Glu Asp Leu Ala  Ser Ser Gly Ser Pro  Arg Asp Leu
    1385                 1390                 1395

Gln Lys  Leu Ser Pro Leu Leu  Ala Asp Ser Ser Leu  Thr Lys Glu
    1400                 1405                 1410

Leu Leu  Gly Gly Asp Arg Glu  Leu Gly Asn Ala Met  Asp Gly Ala
    1415                 1420                 1425

Arg Gly  Pro Arg Gly Leu Asp  Thr Ala Lys Gly Arg  Ala Lys Asp
    1430                 1435                 1440

Glu Glu  Arg Leu Thr Ala Lys  Asp Ala Glu Ser Arg  Gln Ala Thr
    1445                 1450                 1455

Leu Pro  Gly Gly Arg Ala Ala  His Gly Gly Gly Val  Gly Gly Ser
    1460                 1465                 1470

Leu Gly  Thr Ala Cys Glu Glu  Glu Leu Asp Glu Pro  Leu Ser Pro
    1475                 1480                 1485

Leu Asp  Ile Glu Ser Ile Val  Ala Asp Ala Tyr Glu  Ser Phe Ser
    1490                 1495                 1500
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Asp Glu  Asp Ala Glu Gly Glu  Gly Asp Gly Gly Lys  Pro Gly Lys
    1505                1510                1515

Arg Ile  Arg Leu Pro Lys Ile  Gly Gly Val Tyr Tyr  Lys Arg Asp
    1520                1525                1530

Gly Asn  Tyr Lys Ala Trp Ala  Ala Ser Trp His Ile  Gln Gly Lys
    1535                1540                1545

Arg Thr  Arg Arg Tyr Phe Thr  Val Lys Lys His Gly  Phe Arg Asn
    1550                1555                1560

Ala Tyr  Leu Lys Ala Val Arg  Ala Arg Arg Glu Ala  Glu Arg His
    1565                1570                1575

Glu Gly  Ile Ser Val Lys His  Arg His His Ala Leu  Val Pro Gly
    1580                1585                1590

His Pro  Gly Asn Met Leu Gly  Ala Ser Lys Val Cys  Ala Glu Ser
    1595                1600                1605

His Glu  Val Ser Gly Phe Pro  His Gly Asp Glu Asp  Ser Arg Leu
    1610                1615                1620

Thr Arg  Gly Gly Ala Ser His  Ala Ala Val Ala Pro  Gly Arg Val
    1625                1630                1635

Asn Arg  Glu Arg Ser Val Ala  Leu Val Asp Arg Ala  Thr Lys Asp
    1640                1645                1650

Asp Glu  Asp Asp Glu Arg Asp  Leu Gln Arg Glu Lys  Thr Gly Ala
    1655                1660                1665

Gly Gly  Gly Glu Ala Cys Ser  Gly Glu Ser Val Lys  Val Ala Leu
    1670                1675                1680

Gly Thr  Arg His Asp Ser Phe  Ser Asp Gly Ser Cys  Arg Thr Leu
    1685                1690                1695

Asp Lys  Leu Ser Thr Gln Phe  Glu Gln Lys Pro Arg  Gly Gly Ala
    1700                1705                1710

Gly Glu  Glu Ala Glu His Pro  Thr Arg Lys Gln Gly  Gln Glu Thr
    1715                1720                1725

Gly Gly  Val Asp Glu Pro Leu  Ser Arg Ala Ala Ser  Ile Val Gly
    1730                1735                1740

Gly Arg  Glu Val Arg Leu Thr  Ser Gly Val Ser Val  His Leu Thr
    1745                1750                1755

Pro Leu  Glu Arg Val Ala Lys  Ala Val Asp Val Asp  Leu Lys Glu
    1760                1765                1770

Leu Thr  Asp Arg Val Ser Arg  Ala Ala Phe Arg Gly  Gly Asp Ser
    1775                1780                1785

Arg Leu  Phe His Arg Thr Val  Asp Asn Cys Glu Gly  Glu Ala Asp
    1790                1795                1800

Glu Val  Ala Gln Gly Leu Asp  Thr His Arg Glu Asp  Val Asp Val
    1805                1810                1815

Thr Arg  Asn Leu Glu Phe Ala  Met Ala Arg Glu Thr  Leu Asp Val
    1820                1825                1830

Leu Leu  Ser Asp Leu Tyr Ser  Val Val Ala Lys Leu  Ser Gly Ala
    1835                1840                1845

Gly Arg  Trp Thr Ser Leu Val  Ser Pro Thr Ala Ala  Glu Ala Glu
    1850                1855                1860

Pro Leu  Val Ser Ala Trp Asp  Arg Ser Ala Arg Glu  Glu Arg Arg
    1865                1870                1875

Glu Lys  Phe Glu Asp Thr Asn  Ala Ala Ser Asp Glu  Pro Gly Tyr
    1880                1885                1890

Pro Thr  Ser Ser Ala Gln Ile  His Val Ala Ile Gln  Leu Val Val
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    1895                1900                1905

Ile Lys  His Tyr Leu Ala Thr  Val Arg Thr Ala Asn  Arg Val Glu
    1910                1915                1920

Gln Ile  Ala Pro Leu Leu Ala  Leu Phe Glu Pro Cys  Ile Lys Gln
    1925                1930                1935

Gly Met  Leu Pro His Glu Cys  Ala Leu Pro Arg Leu  Arg Trp Leu
    1940                1945                1950

Val Cys  Gln Leu Cys Arg Ala  Ser Leu Pro Trp Leu  Asp Glu Ser
    1955                1960                1965

Asp Val  Leu Thr Asp Ala Leu  Leu Tyr Arg His Leu  Glu Glu Leu
    1970                1975                1980

Val Glu  Thr Glu Glu Ala Glu  Ala Pro Gln Glu Gly  Val Pro Pro
    1985                1990                1995

Gly Gly  Gln Ile Val Phe Ser  Ala Gly Phe Ala Glu  Gly Asn Ser
    2000                2005                2010

Thr Val  Ala Ser Arg Asn Val  Phe Thr Gly Glu Ser  Arg Val Ala
    2015                2020                2025

Gly Gly  Phe Arg Thr Asp Ser  Glu Lys Glu Ser Gly  Ile Asp Asp
    2030                2035                2040

Arg Asp  Glu Ala Ser Leu Ala  Ala Leu Ile Cys Leu  Pro Gly Lys
    2045                2050                2055

Gly Lys  Lys Leu Arg Glu Glu  Ala Asp Val Glu Lys  Asp Asp Thr
    2060                2065                2070

Ser Ala  Ser Leu Asn Cys Glu  Ser Gly Lys Lys Thr  Glu Ala Glu
    2075                2080                2085

Ser Gln  His Ser Arg Ser Pro  Thr Glu Val Ala Ala  Ser Ser Val
    2090                2095                2100

Ser Gly  Ser Glu Gly Lys Asp  Gly Ser Ser Asp Asn  Glu Arg Ser
    2105                2110                2115

Gly Asp  Ala Asp Asp Ala Thr  Glu Gly Ser Glu Lys  Cys Glu Lys
    2120                2125                2130

Thr Arg  Gly Gly Asp Gln Arg  Arg Ala Ala Pro Arg  Thr Ser Ser
    2135                2140                2145

Ala Ser  Thr Ala Ser Gly Glu  Thr Pro Glu Lys Ser  Lys Asn Arg
    2150                2155                2160

Gly Ser  Asp Ala Leu Lys Gly  Lys Asn Glu Gly Gly  Ala Thr Gly
    2165                2170                2175

Thr Ser  Gly Glu Gln Arg Asp  Asp Glu Asp Arg Asp  Leu Glu Asn
    2180                2185                2190

Val Glu  Ile Ser Lys Asp Thr  Arg Ala Gly Ser Gly  Gly Arg Arg
    2195                2200                2205

Arg Thr  Gly Glu Arg Arg Gly  Gln Arg Phe Cys Ala  Ser Gly Gly
    2210                2215                2220

Glu Leu  Arg Val Ser Glu Glu  Ser Pro Asp Arg Ala  Lys Thr Glu
    2225                2230                2235

Lys Ser  Lys Gly Glu Pro Val  Arg Asp Ser Leu Ser  Pro Asp Ala
    2240                2245                2250

Ser Ser  Arg Leu Pro Ser Arg  Cys Gly Thr Pro Pro  Pro Ala Ala
    2255                2260                2265

Ala Thr  Gly Ser Cys Ala Thr  Val Glu Ser Asp Val  Pro Ala
    2270                2275                2280
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<210> SEQ ID NO 11

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<211> LENGTH: 3236
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 11

Met Thr Thr Val Ser Arg Ala His Ala Ser Arg Ser Arg Arg Lys Ser
1 5 10 15

Arg Asp Glu Asp Ser Glu Gly Ser Ser Leu Pro Ala Val Gly Ile His
20 25 30

Glu Thr Gln Ser Pro Val Phe Ser Arg Glu Gly His Glu Gly Asp Arg
35 40 45

Ala Ala Gln Pro Glu Asp Val Val Ala Ala Glu Ser His Ser Asn Pro
50 55 60

Gln Trp Pro Thr Pro Leu Asp Thr Gly Phe Asp Lys Gly Ala Pro Pro
65 70 75 80

Leu Gly Cys Ser Arg Ser Glu Glu Leu Arg Ser Pro Pro Met Ala Ser
85 90 95

Gly Ser Phe His Gly Ser Gly Thr Gly Gly Asp Gly Gly Cys Leu Leu
100 105 110

Ser Leu Glu Ala His Ala Val Ser Lys Asp Ser Glu Arg Gln Val Asn
115 120 125

Ser Gly Leu Pro Gly Gly Gly Asp Glu Ile Ser Gly Arg Leu Ser Pro
130 135 140

Ser Cys Ala Ser Leu Pro Leu Val Ala Ala Ala Leu Ser Pro Val Glu
145 150 155 160

Asp Thr Arg Leu Glu Arg Asp Ser Ser Ile Pro Val Leu Lys Pro Ser
165 170 175

Leu Ser Ile Pro Asn Leu Leu Val Thr Ser Pro Ser Leu Thr Ser Val
180 185 190

Ser Tyr Val Cys Glu Ala Asp Arg Ser Ala Glu Gly Lys Ala Pro Ser
195 200 205

Met Asp Ala Leu Pro Pro Ser His Ser Ala Ala Pro Glu Ser Gly Leu
210 215 220

Trp Arg Glu Cys Asp Glu Arg Gly Lys Asn Ser Phe Phe Ser Ser Gly
225 230 235 240

Leu Pro Ala His Pro Glu Gly Asn Gly Glu Arg Ala Gly Glu Gly Gln
245 250 255

Asp Pro Arg Ser Gly Asp Phe Glu Thr Pro Glu Glu Ala Ala Phe Ser
260 265 270

Val Gln Arg Ile Leu Gln Glu Ser Glu Glu Leu Phe Leu Leu Ser Gly
275 280 285

Cys Ala Arg Glu Asp Arg Gly Gly Glu Ser Asp Ala Ala Phe Lys Thr
290 295 300

Met Thr Arg Ser Glu Gly Ala Phe Ser Arg Glu Pro Ala His Arg His
305 310 315 320

Ala Phe Ala Lys Pro Gly Asn Gly Gly Glu Ser Glu Pro Phe Met Ser
325 330 335

Ile Asp Glu Glu Arg Ala Ala Ser Pro Ser Ala Ser Gly Pro Ser Ser
340 345 350

Tyr Ala Phe Leu Ser Phe Glu Glu Thr Ser Ala Gly Ser Arg Arg Ser
355 360 365

Pro Asp Ala Gln Ser Pro Pro Leu Ser Gly His Leu Ser Asp Gly Asp
370 375 380

Arg Thr Gln Arg Lys Ala Gly Glu Arg Phe Leu Glu Asp Pro Arg Gly

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385                 390                 395                 400

Asn Leu Lys Arg Ser Arg Ser Pro Leu Ile Ala Arg Asp Cys Asn Arg
                405                 410                 415

Ser Leu Gly Thr Cys Asp Ser Ser Leu Thr Thr Arg Gly Pro Val Ala
            420                 425                 430

Ser Asp Thr Ser Pro Arg Arg Gly Tyr Thr Asp Gln Trp His Ser His
        435                 440                 445

Arg Lys Ala Gln Ser Pro Gly Arg Phe Arg Arg Thr Asn Thr Glu Gly
    450                 455                 460

Asn Ala Thr Pro Val Asp Ser Gln Ser Ser Pro Pro Ser Lys Lys Arg
465                 470                 475                 480

Cys Cys Leu Ala Glu Arg Phe Ala Phe Glu Arg Arg Arg Gln Pro Pro
                485                 490                 495

Val Pro Leu Pro Ser Val Ala Ser Ala Val Ala Ala Ala Leu Ala Gln
            500                 505                 510

Phe Pro Pro Gly Ala Cys Thr Ala Ala Val Glu Arg Ala Asp Asp Val
        515                 520                 525

Pro Pro Glu Gly Ser Gly Asn Gly Val Leu Pro Gly Gly Glu Val Ser
    530                 535                 540

Asp Leu Ser Leu Ser Asp Arg Lys Ser Gly Ala Ser Pro Arg Gln Thr
545                 550                 555                 560

Leu Asp Thr Phe Leu Pro Ala Lys Gly Ala Ser Ala Ala Leu Lys Gln
                565                 570                 575

Glu Gly Ser Ser Ser Glu Ile Gly Glu Gly Cys Pro Ala Ser Asp Asp
            580                 585                 590

Ala Ala Val Ala Ala Thr Leu Ser Gly Trp Lys Arg Gly Arg Gly Pro
        595                 600                 605

Arg Ser Gly Cys Val Ser Gly Ser Ser Arg Ala Ser Ser Leu Glu His
    610                 615                 620

Ala Gly Ala Arg Arg Arg Ser Gly Val Glu Arg Lys Arg Arg Glu Lys
625                 630                 635                 640

Arg Lys Ala Ala Leu Ala Thr Ala Ala Val Val Ser Ala Ser Val Arg
                645                 650                 655

Arg Leu Ala Leu Val Ala Ala Pro Cys Leu Val Glu Asn Thr Leu Arg
            660                 665                 670

Gln Trp Trp Arg Leu Gln Glu Gln Val Gly Asp Glu Leu Asp Asn Gly
        675                 680                 685

Gly Val Ser Glu Glu Gln Thr Thr Gly Arg Ser Gly Arg Arg Thr Gly
    690                 695                 700

Lys Asn Pro Ile Val Gly Gly Arg Val Lys Glu Gly Glu Gln Ser Val
705                 710                 715                 720

Arg Leu Glu Ile Glu Arg Ser Gly Asp Ser Pro Arg Asn Thr Ala Lys
                725                 730                 735

Thr Glu Pro Gly Asp Gln Gly Ala Ala Gln Gly Gln Gly Gly Pro Glu
            740                 745                 750

Gln Ile Ala Glu Asn Glu Ser Gly Thr Glu Arg Met Glu Thr Ser Gln
        755                 760                 765

Thr Lys Gln Glu Ala Gln Asp Leu Pro Leu His Arg Glu Ala Ala Ser
    770                 775                 780

Ala Ser Ala Thr Pro Phe Ile Pro Glu Gly Arg Thr Gln Glu Arg Asp
785                 790                 795                 800

Ser Tyr Leu Arg Val Thr Leu Phe Ala Ala Ser Gln Val Leu Asn Ser
                805                 810                 815
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Gly Gln Phe Arg Gln Ala Ile Arg Met Phe Pro Gly Ala Asp Ser Pro
            820                 825                 830

Arg Gly Asp Gln Arg Ser Arg Cys Val Val Arg Val Tyr Lys Gln Ser
        835                 840                 845

Leu Arg Lys Arg Arg Leu Ser Gly Asp Arg Asn Arg Arg Val Gly Glu
    850                 855                 860

Asp Gly Asn Leu Val Ser Val Ser Leu Arg Asp Leu Leu Ala Asn Ser
865                 870                 875                 880

Gly Glu Asp Trp Glu Pro Ser Ser Pro Ser Ala Ala Thr Arg Pro Leu
                885                 890                 895

Pro Leu Ala Pro Ser Ala Leu Ser Ala His Gln Ala Arg Ser Ser Phe
            900                 905                 910

Gly Ser Phe Ser Arg Arg Ser Ala Glu Gly Val Pro Gly Pro Ser Pro
        915                 920                 925

Trp Gly Gly Asp Cys Ser Pro Thr Ala Ala Arg Ser Pro Leu Pro Leu
    930                 935                 940

Ser Thr Ser His Asp Leu Arg Arg Arg Arg Val Cys Pro Pro Arg Arg
945                 950                 955                 960

Arg Tyr Ser Pro Ser Glu Ser Val His Ser Ser Asp Gly Arg Gly Gly
                965                 970                 975

Ala Cys Ala Ile Ala Ala Lys Lys Pro Lys Gly Arg Arg Ser Arg Gly
            980                 985                 990

Arg Glu Glu Gln Thr Arg Glu Glu  Val Ser Glu Ser Arg  Cys Ser Thr
        995                 1000                1005

Pro Arg  Ser Cys Ser Ser Val  Arg Tyr Ala Val Ser  Asp Gly Ser
    1010                1015                1020

Pro Ala  Ser Ser Arg Ala His  Leu Gly Arg Pro Asp  Asp Glu Gly
    1025                1030                1035

Asp Glu  Arg Met Thr Gly Gly  Gln Arg Thr Pro Arg  Gly Thr Pro
    1040                1045                1050

Gln Glu  Gly Glu Asp Ser Asp  Phe Leu Pro Ala Gly  Met Ser Gly
    1055                1060                1065

Leu Arg  Gly Gly Thr Leu Pro  Leu Asp Gln Leu Gly  Glu Arg Ser
    1070                1075                1080

Arg Ser  Ala Glu Arg Trp Met  Pro Ala Pro Ser Val  Ala Val Val
    1085                1090                1095

Pro Phe  Ala Pro Asn Ile Leu  Ala Lys Lys His Ala  Glu Asp Val
    1100                1105                1110

Glu Asn  Gln Leu Asp Gly Gly  Lys Met Ser Leu Asp  Gly Val Gly
    1115                1120                1125

Gln Lys  Glu Cys Gly Leu Val  Glu Thr Gly Asp Thr  Gly Glu Gln
    1130                1135                1140

Glu Ala  Ala Val Ala Ala Ser  Glu Lys Arg Arg Pro  Leu Glu Ala
    1145                1150                1155

Gln Thr  Pro Gly Arg His Gly  Thr Thr Val Leu Met  Lys Gly Glu
    1160                1165                1170

Gly Leu  Leu Ala Gly Arg Thr  Ser Glu Val Asp Gly  Asp Arg Thr
    1175                1180                1185

Gly Glu  Lys Thr Thr Gln Ile  Ser Pro Phe Ser Glu  Ala Thr Gly
    1190                1195                1200

Ile Cys  Ile Leu Arg Arg Ser  Pro Arg Arg Val Gln  Ser Asn Ser
    1205                1210                1215
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Ser Glu  Ala Ser Arg Thr Ala  Val Arg Ser Thr Glu  Gly Leu Glu
    1220                1225                1230

Thr Ser  Asp Lys Leu Gly Val  Asp Val Gly Thr Thr  Asn Lys Glu
    1235                1240                1245

Ala Asp  Ser Phe Ser Ala Ser  Cys Asp Ser Pro Arg  Asp Ser Leu
    1250                1255                1260

Glu Arg  Asn Val Gly Glu Ile  Val Ala Ile Trp Ala  Arg Ala Arg
    1265                1270                1275

Asp Ala  Lys Gln Gly Gly Arg  Ile Arg Arg Arg Val  Trp Leu Pro
    1280                1285                1290

Pro Gly  Met Ala Thr His Gly  Gly His Glu Gly Asn  Glu Gln Asn
    1295                1300                1305

Asn Glu  Ala Ile Cys Gly Gly  Gly Ala Thr Pro Met  Met Lys Thr
    1310                1315                1320

Glu Arg  Ala Met Glu Glu Gly  Arg Gly Asp Ala Lys  Thr His Pro
    1325                1330                1335

Val Gly  Gly Thr Tyr Ala Glu  Thr Glu Lys Lys Val  Val Asp Glu
    1340                1345                1350

Met Lys  Ala Trp Trp Ser Lys  Leu Thr Cys Ala Ser  Val Glu Ala
    1355                1360                1365

Val Pro  Val Gln Thr Leu Thr  Leu Asp Asp Phe Ala  Arg Ala Phe
    1370                1375                1380

Ser Thr  Val Ala Asn Arg Ala  Val Asp Leu Leu Cys  Leu Ala Phe
    1385                1390                1395

Arg Ala  Arg Gly Ala Gly Pro  Val Phe Arg Pro Val  Leu Ser Ser
    1400                1405                1410

Ser Pro  Lys Gln Gln Gly Asn  Ser Pro Gln Pro Glu  Ser Glu Asp
    1415                1420                1425

Val Glu  Thr Arg Ile Glu Thr  Tyr Arg Gln Gln Val  Arg Arg Leu
    1430                1435                1440

Tyr Arg  Arg Arg Gln Gln Leu  His Glu Ala Thr Gly  Asn Ser Pro
    1445                1450                1455

Phe Ser  Ser Ser Arg Val Gly  Gly Ala Leu Gln Arg  Arg Ile Gly
    1460                1465                1470

Glu Leu  Gln Arg Leu Arg Glu  Ala His Gly Arg Val  Asp Ile Pro
    1475                1480                1485

Asn Glu  Gly Pro Arg Arg Glu  Glu Asp Ser Glu Lys  Cys Pro Ala
    1490                1495                1500

Ser Leu  Trp Asp Val Pro Leu  Arg Gln Arg Lys Gln  Gly Arg Lys
    1505                1510                1515

Arg Val  Ser Pro Trp Tyr Ser  Val Gly Val Arg Trp  Leu Ala Asp
    1520                1525                1530

Phe Ser  Ala Phe Glu Tyr Phe  Val Val Lys Asn Tyr  Arg Lys Glu
    1535                1540                1545

Asp Leu  Gly Ala Thr Val Ser  Leu Ser Asn Arg Gly  Glu Ala Ser
    1550                1555                1560

Asp Thr  Thr Trp Ser Val Asp  Gly Thr Gly Ser Gln  Arg Ala Pro
    1565                1570                1575

Val Pro  Cys Leu Ser Arg Ala  Gly Thr Pro Arg Thr  Val Ser Pro
    1580                1585                1590

Ser Pro  Pro Ser Ala Asp Ala  Met Asn Leu Trp Ala  Gln Ala Tyr
    1595                1600                1605

Ala Pro  Ser Leu Asn Gln Pro  Arg Gly Met Ser Pro  Ala Thr Thr
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    1610                1615                1620

Pro Pro  Leu Ser Glu Ser Ala  Thr Pro Arg Gly Asn  Val Ser Pro
    1625                1630                1635

Pro Phe  Ser Glu Ala Ser Ser  Ser Gly Gln Arg Gly  Lys Lys Val
    1640                1645                1650

Ala Pro  Gly Pro Ser Ala Asp  Glu Lys Lys Asp Glu  Asp Tyr Gln
    1655                1660                1665

Ser Ala  Gly Ser Leu Arg Trp  Glu Val Asp Gly Gly  Gln Arg Arg
    1670                1675                1680

Gln Val  Gln Val Arg Ser Arg  Leu Leu Leu His Glu  Leu Leu Gln
    1685                1690                1695

Pro Pro  Ser Leu Asp Ala Thr  Gly Leu Arg Thr Ala  Leu Val Leu
    1700                1705                1710

Ile Val  Leu Arg Leu Gln Arg  Phe Leu Arg Leu Lys  Glu Gly Gly
    1715                1720                1725

Val Asn  Asn Arg Gly Arg Gly  Gln Arg Ser Glu Arg  Lys Arg Arg
    1730                1735                1740

Cys Arg  Ala Met Pro Pro Ile  Phe Ile Phe Arg Asp  Asp Ser Asn
    1745                1750                1755

Ala Phe  Gln Glu Ala Leu Leu  Ala Lys Lys Leu Asp  Ile Arg Leu
    1760                1765                1770

Asp Ser  Asp Ser Pro His Thr  Asp Val Pro Ser Arg  Arg Ser Leu
    1775                1780                1785

Asp Gly  Glu Val Gly Asp Glu  Arg Arg Arg Leu Arg  Ser Val Lys
    1790                1795                1800

Pro Thr  Asn Ser Asn Asp Leu  Ser Asp Glu Arg Gly  Pro Pro Pro
    1805                1810                1815

Pro Ser  Thr Met Ser Pro His  Ser Leu Gly Ser Gly  Pro Cys Asp
    1820                1825                1830

Thr Gln  Glu Gly Val Gln Asn  Leu Gln Gln Asp Ala  Ser Leu Phe
    1835                1840                1845

Ser Pro  Ala Leu Ala Gln Gly  Gln Ala His Ala His  Thr Asp Ala
    1850                1855                1860

Val Pro  Gly Ala Arg His Asp  Asp Val Leu Pro Arg  Ser Pro Arg
    1865                1870                1875

Phe Pro  Val Val Asp Ala Gly  Pro Glu Glu Thr Pro  Arg Pro Glu
    1880                1885                1890

Val Glu  Ser Met Leu Asp Ser  Glu Ser Gly Asp Pro  Thr Gly Leu
    1895                1900                1905

Gly Gln  Ala Ser Arg Arg Arg  Trp Arg Gly Arg Gly  Ser Arg Thr
    1910                1915                1920

Ser Val  Gln Arg Thr Val Ser  Thr Cys Leu His Glu  Asp His Ser
    1925                1930                1935

Gly Asp  Lys Thr Pro Arg Glu  Glu Thr Phe Gly Gly  Asp Ala Ala
    1940                1945                1950

Ser Leu  Leu Arg Val Ala Ser  Ser Val Pro Pro Ser  Thr Cys Ser
    1955                1960                1965

Ser Pro  Gln Ser Ser Ser Gly  Gly Arg Arg Glu Arg  Gly Arg Arg
    1970                1975                1980

Gly Val  Arg Gly Arg Arg Gly  Arg Gly Arg Leu Ile  Ser Gln Gln
    1985                1990                1995

Gly Ser  Ser Leu Leu Gly Gln  Thr Val Ser Ala Gly  Ala Leu Ser
    2000                2005                2010
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Ser Gly  Asp Thr Ala Gly Ala  Ile Ser Thr Glu Gly  Glu Asn Arg
    2015                 2020                 2025

Arg Asn  Ala Val Arg Pro Gly  Ala Leu Glu His Ser  Asp Glu Asp
    2030                 2035                 2040

Lys Glu  Asp Leu Ser Ala Ser  Ser Pro Pro Ser Asp  Asp Gly Ile
    2045                 2050                 2055

Ser Gln  Arg Ser Ser Gly Ser  Gln Gly Asp Ser Ser  Ser Ser Gly
    2060                 2065                 2070

Gly Pro  Ser Ser Glu Ala Cys  Arg Lys Thr Thr Ser  His Val Ala
    2075                 2080                 2085

Ala Lys  Ala Asp Ser Ala Ser  Pro Arg Ala Leu His  Pro Ser Ala
    2090                 2095                 2100

Arg Pro  Gln Pro Arg Gly Thr  Ala Ser Trp Thr Pro  Gly Gly Glu
    2105                 2110                 2115

Pro Ala  Val Ser Gly Val Gln  His Pro Ser Ala Leu  Thr Pro Ser
    2120                 2125                 2130

Pro Ser  Arg Gly Arg Phe Ser  Glu Asp Asn Val Ala  Ser Arg Val
    2135                 2140                 2145

Ser Arg  Val Ser Ser Val Gly  Ala Leu Leu Arg Ser  Arg Cys Val
    2150                 2155                 2160

Val Gly  Glu Glu Gln Lys Glu  Thr Gln Asn Ser Cys  Ser Leu Trp
    2165                 2170                 2175

Val Val  Glu Lys Gly Ala Leu  Glu Pro Phe Trp Trp  Arg Thr Ala
    2180                 2185                 2190

Ser Ala  Val Gly Cys Val Ser  Ala Gly Arg Arg Asp  His Ser Asp
    2195                 2200                 2205

Lys Asp  Ala Asn Arg Leu Phe  Leu Ala Asp Lys Glu  Ala Gly Thr
    2210                 2215                 2220

Gly Pro  Leu Gln Asp Phe Val  Leu Pro Asp Phe Ser  Gly Ser Ala
    2225                 2230                 2235

Arg Glu  Ile His Gly Asp Glu  Arg Gly Ser Asp Ser  Asp Ala Ser
    2240                 2245                 2250

Cys Lys  Ser Ala Ala Leu Ser  Thr Thr Ser Asp Ser  Ser Gly Ile
    2255                 2260                 2265

Ser Glu  Val Ser Leu Asp Leu  Glu Ser Thr Val Gln  Glu Val Ala
    2270                 2275                 2280

Leu Gly  Thr Ile Leu Ser Ser  Ala Leu Ser Ala Leu  His Gly Lys
    2285                 2290                 2295

Thr Gly  Asp Gly Asp Thr Gln  Glu Ser Asp Ala Glu  Arg Glu Ala
    2300                 2305                 2310

Asn Ala  Asp Asp Gly Ser Ala  Thr Gly Val Asn Glu  Lys Asp Leu
    2315                 2320                 2325

Arg Gly  Glu Ser Arg Pro Glu  Leu Pro Ser Pro Ile  Pro Gly Lys
    2330                 2335                 2340

Asp Glu  Leu Gly Ser Gln Glu  Glu Gly Lys Thr Ala  Ser Ser Leu
    2345                 2350                 2355

Pro Ser  Val Lys Ala Glu Gln  Gly Gly Ser Glu Arg  Gly Gly Ala
    2360                 2365                 2370

Asp Glu  Ile Val Lys Lys Ala  Thr Ser Val Leu Arg  Ala Cys Lys
    2375                 2380                 2385

Asp Pro  Asp Glu Ala Thr Ser  Thr Ser Leu Val Pro  Glu Gly Glu
    2390                 2395                 2400
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Asp Glu  Asn Asp Ala Cys Gly  Ala Leu Glu Pro Asp  Ser Leu Val
    2405                 2410                 2415

Ser Val  Ser Ala Leu Gly Glu  Ser Ser Glu Glu Leu  Phe Thr Glu
    2420                 2425                 2430

Val Pro  Gln Asn Glu Lys Glu  Leu Lys Lys Thr Leu  Gln His Val
    2435                 2440                 2445

Asp Pro  Arg Leu Cys Gln Gln  Met Leu His Gly Gly  Leu Cys Phe
    2450                 2455                 2460

Ile Arg  Thr Tyr Val Asp Leu  Glu Thr Lys Lys Glu  Ser Leu Gln
    2465                 2470                 2475

Ala Gly  Pro Phe Ala Ala Lys  Arg Arg Arg Val Ala  Gln Leu Leu
    2480                 2485                 2490

Arg Gly  Leu Gln Gly Leu Phe  Asp Ala Leu Glu Ser  Val Arg Glu
    2495                 2500                 2505

Arg Glu  Gly Asp Asp Leu Ser  Gly Glu His Glu Gly  Asp Ser Ala
    2510                 2515                 2520

Ser Gly  Gly Leu Phe Thr Ala  Glu Gln Glu Lys Glu  Gly Ala Asp
    2525                 2530                 2535

Lys Val  Ser Gly Asp Arg Glu  Asn Ala Gly Glu Arg  Gly Gln Lys
    2540                 2545                 2550

Thr Ala  Ala Glu Thr Gly Asp  Gln Lys Ala Ser Ile  Glu Asp Ala
    2555                 2560                 2565

Val Ala  Ala Ala Phe Cys Arg  Arg Val Gly Ala Ala  Ile Ala Thr
    2570                 2575                 2580

Glu Thr  Cys Gly Ser Ile Gln  Thr Val Phe Pro Glu  Ile Gly Glu
    2585                 2590                 2595

Ala Tyr  Asp Val Glu Asp Ser  Val Ala Arg Leu Gly  Ala Pro Pro
    2600                 2605                 2610

Arg Ala  Pro Val Arg Thr Arg  Arg Glu Cys Thr Gly  Thr Gly Phe
    2615                 2620                 2625

Thr Ser  Thr Ala Ala Leu Pro  Glu Pro Arg Gly Glu  Asp Gly Arg
    2630                 2635                 2640

Lys Gln  Glu Thr Ser Glu Pro  Leu Gly Val Glu Ala  Ala Asp Lys
    2645                 2650                 2655

Thr Asp  Ile Gln Gly Glu Tyr  Ala Gln Glu Ser Glu  His Thr Trp
    2660                 2665                 2670

Thr Gln  Glu Met Gly Arg Lys  Ala Ser Leu Phe Leu  Ser Gly Thr
    2675                 2680                 2685

Leu Glu  Leu Ala Gln Leu Lys  Glu Glu Gln Gln Val  Glu Glu Leu
    2690                 2695                 2700

Gln Gly  Glu Gly Asp Pro Leu  Thr Ser Phe Leu Leu  Pro Ser Asp
    2705                 2710                 2715

Gln Ser  Asp Ser Thr Lys Lys  Ala Asn Glu Glu Cys  Met Gly Gly
    2720                 2725                 2730

Arg Thr  Ala Arg Glu Leu Tyr  Ala Glu Arg Glu Glu  Asp Val Lys
    2735                 2740                 2745

Thr Leu  Gly Arg Arg Arg Glu  Ala Gln Thr Glu Ser  Arg Ala Arg
    2750                 2755                 2760

Gly Pro  His Val Asp Ser Ser  Ala Glu Ala Ala Ser  Val Ala Gln
    2765                 2770                 2775

Gly Asp  Glu Gly Gly Glu Glu  Ala Arg Lys Arg Lys  Lys Asp Glu
    2780                 2785                 2790

Lys Arg  Glu Lys Arg Ser Gly  Asn Ala Phe Leu Asp  Ala Leu Leu
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    2795                2800                2805

Glu Pro  Ala Leu Arg Glu Asp  Val Gly Arg Ala Phe  Leu Thr Asp
    2810                2815                2820

Phe Gly  Ser Gln Ala Pro Gln  Asn Ser Thr Asp Ala  Gly Lys Pro
    2825                2830                2835

Ile Phe  Leu Ser Pro Cys Val  Phe Gly Val Arg Gly  Gly Ala Arg
    2840                2845                2850

Trp Lys  Lys Leu Gly Leu Phe  Asn Asp Glu Ala Gln  Arg Glu Gly
    2855                2860                2865

Thr Glu  Ser Ser Pro Trp Arg  Asn Asp Cys Ser Asp  Pro Met Ser
    2870                2875                2880

Tyr Arg  Ala Asp Ala Pro His  Thr Trp Arg Arg His  Glu Gly Leu
    2885                2890                2895

Leu Trp  Gly Gly Ser Arg His  Ala Ala Ser Ala Leu  Arg His His
    2900                2905                2910

Gly Arg  Lys Ser Pro Ala Phe  Leu Ser Pro Gln Trp  Glu Asp Asp
    2915                2920                2925

Glu Arg  Leu Ser Leu Ser Ser  Ser Ala Asp Glu Arg  Gly Tyr Thr
    2930                2935                2940

Ser Ser  Gly Ser Glu Arg Phe  Leu Ser Ile Pro Thr  Arg Arg Lys
    2945                2950                2955

Tyr Gly  Leu Arg Phe Gln Arg  Arg Ser Thr Lys Thr  Gly Arg Ala
    2960                2965                2970

Pro Ser  Pro Thr Ala Gly Arg  Ser Ser Val Asn Arg  Ser Gly Trp
    2975                2980                2985

Arg Glu  Thr Leu Arg Pro Ser  Ser Gly Phe Ser Gly  Glu Glu Thr
    2990                2995                3000

Pro Arg  Ser Leu Ser Ser Arg  Arg Arg Arg Gly Gly  Leu Gly Gly
    3005                3010                3015

Ser Ser  Pro Thr Ala Phe Arg  Pro Pro Met Thr Arg  Ala Ala Thr
    3020                3025                3030

Gly Lys  Ala Ala Ala Cys Val  Arg His Gly Asp Gly  Asp Glu Cys
    3035                3040                3045

Ala Glu  Pro Asp Ser Gln Phe  Gly Ala Phe Gly Ser  Ala Asp Leu
    3050                3055                3060

Gly Leu  Ser Asp Arg Arg Gly  Glu Ala Gly Glu Ala  Asp Thr Arg
    3065                3070                3075

Glu Glu  Lys Ala Gly Gly Ser  Ala Arg His Gly Lys  Arg Gly Ser
    3080                3085                3090

Gly Val  Arg Ser Gly Gly Ala  Arg Glu Ala Gly Ser  Asp Ala Gly
    3095                3100                3105

Thr Asp  Thr Leu Trp Val Ala  Pro Gly Ser Gly Pro  Asn Thr Cys
    3110                3115                3120

Arg Ser  Gly Arg Lys Ser Pro  Ala Ala Ala Ala Leu  Ser Ser Leu
    3125                3130                3135

Pro Thr  Gly Val Tyr Phe Asp  Ala Ser Arg Lys Leu  Trp Arg Cys
    3140                3145                3150

Gln Trp  Arg Glu Asn Gly Arg  Phe Lys Thr Lys Gly  Phe Ser Leu
    3155                3160                3165

Asn Val  Tyr Lys Thr Leu Lys  Glu Ala Arg Arg Ala  Cys Val Val
    3170                3175                3180

Tyr Arg  Cys Leu Met Gly Gly  Trp Glu Val Asp Pro  Arg Trp Leu
    3185                3190                3195
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Gly Pro  Asp Asp Asp Glu Gln  Asp Asn Ser Gly Gly  Ala Asp Glu
    3200                3205                3210

Val Gly  Arg Pro Val Pro Ser  Asp Gly Ile Ser Asp  Val Val Gly
    3215                3220                3225

Glu Ala  Arg Arg Lys Gly Glu  Tyr
    3230                3235


<210> SEQ ID NO 12
<211> LENGTH: 1670
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 12

Met Ile Asn Leu His Gln Leu Phe Arg Val Phe Ser Arg Val Ser Ser
1               5                   10                  15

Ser Ala Ser Asp Pro Ser Ala Ser Asn Pro Ser Pro Ala Ser Leu Val
            20                  25                  30

Ser Val Pro Ala Leu Gln Thr Leu Ser Phe Pro Ala Leu Gln Gln Gln
        35                  40                  45

Asp Leu Leu Ala Ser Leu Ala Ala Ala Ser Leu Pro Gly Pro Asp Ser
    50                  55                  60

Val Thr Met Ser Ser Ser Pro Thr Ser Val Leu Asn Ser Ser Phe Cys
65                  70                  75                  80

Ser Leu Pro Ser Ser Arg Lys Pro Ala Ala Leu Pro Phe Pro Ala Thr
                85                  90                  95

Ser Pro Lys Thr Pro His Leu Ser Asp Ser Phe Pro Ala Ser Ala Ile
            100                 105                 110

Ser Gly Pro Ser Ser Pro Gly Leu Gln Glu Leu Leu Ala Ser Pro Glu
        115                 120                 125

Leu Ala Ala Ala Ala Leu Ala Ser Leu Gln Lys Gln Gln Leu Arg Leu
    130                 135                 140

Ala Leu Gly Thr Glu Arg Gly Gly Cys Gly Ala Arg Gly Asp Glu His
145                 150                 155                 160

Leu His Ser Ile Leu Leu Gln His Lys Ala Thr Ser Glu Asn Ala Met
                165                 170                 175

Arg Trp Ser Trp His Ala Gly Arg Asp Gly Ala Gln Glu Leu Asp Thr
            180                 185                 190

Val Pro Glu Thr Phe Asp Leu Pro Leu Ser Leu Ser Ser Phe Leu Gly
        195                 200                 205

Val Ala Pro Gln Gln Pro Ser Ser Leu Pro Arg Ser Ser Leu Leu Pro
    210                 215                 220

Pro Thr Asp Phe Ser Leu Thr Asp Gly Thr Leu Arg Val Ser Ser Ser
225                 230                 235                 240

Met Leu Pro Ala Leu Ala Thr Gly Ser Glu Ser Gly Ser Ser Arg Gly
                245                 250                 255

Leu Asn Ser Ala Gln Ala Ser Pro Ser Phe Ser Ser Leu Arg Gly Pro
            260                 265                 270

Pro Val Ser Val Pro Glu Glu Glu Val Ser Gly Ser Leu Glu Gly Ser
        275                 280                 285

Pro Gly Pro Phe Ser Ser Gly His Pro Pro Ala Ala Pro Ser His Pro
    290                 295                 300

Cys Ser Thr Val Ser Gly Ala Asp Thr Gln Glu Ala Glu Pro Pro Leu
305                 310                 315                 320

Leu Thr Leu Val Ala Val Asn Thr Pro Asp Ala Gln Asp Pro Ala Val
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                325                 330                 335

Asp Gly Ala Ser Leu Cys Ala Ser Lys Glu Gly Met Arg Thr Ser Ser
            340                 345                 350

Ala Asp Leu Gly Asp Ser Leu Leu Ala Pro Pro Gly His Gly Ser Ala
        355                 360                 365

Ala Pro Leu Pro Gly Arg His Leu Gly Ser Asp Ala Thr Arg Thr Thr
    370                 375                 380

Thr Thr Thr Gly Ser Gly Ala Pro Glu Ser Pro Ser Leu Pro Leu Ala
385                 390                 395                 400

Arg Gly Asp Cys Glu Gly Ala Glu Arg Gly Leu Ala Leu Leu Glu Ala
                405                 410                 415

Pro Val Asn Gly Phe Asn Leu Ala Ala Ser Gln Ser Val Leu Gly Gly
            420                 425                 430

Phe Ala Ala Asp Thr Arg Gly Glu Ala Gly Glu Lys Gly Ile Ala Pro
        435                 440                 445

Gln Ser Arg Lys Ala Arg Lys Pro Gly Thr Ala Val Glu Thr Ala Gly
    450                 455                 460

Ala Pro Glu Ala Val Arg Arg Gly Arg Ala Ala Cys Asn Gly Glu Ala
465                 470                 475                 480

Glu Thr Thr Gly Leu Glu Thr Ala Pro Gln Gln Val Ser Thr Ser Glu
                485                 490                 495

Glu Thr Ala Lys Ser Gly Arg Glu Leu Ala Cys Ala Arg Ala Gly Met
            500                 505                 510

Asp Glu Glu Glu Asp Ala Ala Phe Pro Ser His Val Val Ser Glu Phe
        515                 520                 525

Arg Gly Pro Pro Glu Ile Ser Asn Val Phe Asn Asp Leu Asp Cys Ser
    530                 535                 540

Ser Ala Val Glu Arg Pro Gln Gly Cys Leu Gln His Ala Ala Val Gln
545                 550                 555                 560

Pro Phe Leu Pro Ala Val Ala Pro Glu Val Arg Pro Ser Ala Thr Thr
                565                 570                 575

Ala Gly Arg Thr Pro Met Gly Leu Trp Ser Glu Ala Gly Arg Val Ser
            580                 585                 590

Ser Leu Glu Thr Asp Thr Ala Glu Ile Gly Arg Arg Leu Asp Gly Glu
        595                 600                 605

Ser Ser Gly Ser Pro Asp Arg Trp Gly Asp Ala Arg Leu Ser Ser Pro
    610                 615                 620

Asp Ser Val Pro Ser Ser Ala Asp Val Pro Val Pro Ser Arg Pro Gln
625                 630                 635                 640

Cys Gln Glu Gln Val Pro Gln Val Asp Pro Asp Ser Ser His Pro Leu
                645                 650                 655

Phe Ala Ser Cys Ser Ala Gly Ser Ser Ser Thr Ala Gly Ser Ala Ser
            660                 665                 670

Ala Leu Ala Gly Leu Ala Ser Pro Phe Pro Pro Pro Lys Ser Pro Lys
        675                 680                 685

Thr Gly Ala Asn Asp Pro Arg Met Thr Pro Ser Glu Gly Glu Met Arg
    690                 695                 700

Ala Val Ser Gly Ala Pro Pro Ser Leu His Met Ser Pro Pro Ile Pro
705                 710                 715                 720

Pro Leu Ala Leu Gln Asp Ser Phe Gly Glu Cys Thr Ala Ser Ser Leu
                725                 730                 735

Ala Gly Val Asp Ala Pro Glu Ala Thr Ala Gly Gly Leu Ala Glu Gly
            740                 745                 750
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Val Ala Thr Gly Gly Gly Ser Asp Ser Val Gly Glu Gly Arg Leu Pro
        755                 760                 765

Gly Ala Ala Ser Leu Glu Val Pro Ser Ser Pro Ser Ala Leu Leu Ser
    770                 775                 780

Gly Ala Pro Ala Ser Leu Leu Leu Leu Leu Arg Asn Gly Gln Ser Gly
785                 790                 795                 800

Ala Ala Ala Leu Val Ala Ala Met Gln Gln His Gln Ala Leu Ser Gly
                805                 810                 815

Asp Ala Glu Glu Ala Leu Glu Ala Val Leu Ala Gly Gly Ser Asn Val
            820                 825                 830

Gly Asp Met Ala Asn Ser Ser Arg Gly Leu Glu Thr Val Gly Asp Gly
        835                 840                 845

Thr Arg Gly Ser Ala His Thr Thr His Ala Ala His Ser Ser Gly Arg
    850                 855                 860

Asn Ala Val Gly Ala Cys Pro Ala Pro Asp Arg Glu Gly Glu Thr Val
865                 870                 875                 880

Ala Val Pro Thr Ser Val Leu Thr Asn Asn Pro Ala Ser Thr Ser Lys
                885                 890                 895

Thr Met Pro Ser Val Tyr Ser Thr Pro Ala Ser Ala Gly Leu Ser Leu
            900                 905                 910

Thr Ser Ser Ser Thr Pro Pro Val Leu Pro Thr Pro Asn Pro Gly Ala
        915                 920                 925

Gly Met Pro Pro Leu Ala Ser Ala His Ala Ala Ser Pro Ala Val Pro
    930                 935                 940

Gly Asp Ala Asn Leu Gln Ser Leu Phe Phe Trp Ala Pro Gln Ala Cys
945                 950                 955                 960

Pro Leu Gln Pro Gly Ala Leu Ala Val Asp Ala Ser Ala Ser Ser Cys
                965                 970                 975

Gly Gly Val Gly Ser Cys Asn Gly Gly Pro Ala Pro Pro Gly Pro Ser
            980                 985                 990

Pro Val Ala Glu Leu Leu Asp Ala  Ser Gly Ser Gly Pro  Phe Gly Ala
        995                 1000                1005

Ala Gly  Ser Gly Ala Gln Leu  Ala Ala Gly Pro Phe  Gly Ala Ala
    1010                1015                1020

Thr Pro  Ala Ser Ala Thr Phe  Gln Gln Gln Leu Leu  Leu Leu Ser
    1025                1030                1035

Ala Ala  Phe Asp Gln Ile Gly  Ser Ser Ser Phe Pro  Val Val Gly
    1040                1045                1050

Gly Glu  Asn Phe Ile Gly Tyr  Ser Ala Leu Ser Ala  Ala Arg Pro
    1055                1060                1065

Asp Ala  Ser Asp Leu Ser Ala  Ser Gly Gly Pro Pro  Ala Ser Leu
    1070                1075                1080

Pro Val  Leu Leu Ala Ala Ala  Asn Ala Gly Val Gly  Pro Gly Ala
    1085                1090                1095

Ala Gly  Val Gly Asp Gln Pro  Asp Phe Leu Ala Leu  Leu Gly Gly
    1100                1105                1110

Gly Ser  Ala Ser Arg Glu Gly  Ala Arg Asp Pro Val  Gly Gly Glu
    1115                1120                1125

Leu Gly  Gly Ala Gly Asn Ser  Ala Thr Ser Met Lys  Gly Val Lys
    1130                1135                1140

Arg Gln  Phe Val Gln Asn Gly  His Gly Thr Ala Ser  Gln Thr His
    1145                1150                1155
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Pro Glu  Glu Asn Thr Gln Gly  Pro Gly Arg Ser Ala  Ala Val Val
    1160                1165                1170

Gly Arg  Ala Thr Lys Lys Gln  Arg Arg Gly Pro Pro  His Ser Gly
    1175                1180                1185

Ala Ala  Val Ser Ser Gly Ala  Pro Ser Gly Val Leu  Ala Val Pro
    1190                1195                1200

Gly Cys  Leu Gly Pro Pro Ser  Val Ala Lys Gly Pro  Gly Ser Asp
    1205                1210                1215

Glu Phe  Asn Leu Gln Gln Leu  Gln Gln Ser Arg Asp  Ser Arg His
    1220                1225                1230

Ser Ala  Asp Asn Ala Ser Gly  Ile Pro Asn Trp Pro  Pro Val Phe
    1235                1240                1245

Ser Asn  Gly Asn His Thr Leu  Gly Val Gly Thr Arg  Ser Pro Ser
    1250                1255                1260

Pro Ser  Val Cys Ser Ile Ser  His Asp Ala Gly Phe  Phe Gly Ala
    1265                1270                1275

Ser Gly  Ser Asn His Ala Gly  Ser Leu Ser Thr Pro  Val Cys Leu
    1280                1285                1290

Pro Gln  Leu Pro Gly Ala Ala  Ser Ala Ser Glu Gly  Pro Cys Glu
    1295                1300                1305

Ala Gln  Gln Thr Pro Pro Gly  Ser Ile Pro Glu Ala  Thr Thr Leu
    1310                1315                1320

Gly Gly  Leu Ser Ala Ala Ser  Gly Asn Pro Asn Ser  Thr Phe Ser
    1325                1330                1335

Val Ser  Ala Gly Gly Gly Val  Ala Pro Ala Ile Leu  Asn Leu Ser
    1340                1345                1350

Ser Ala  Ser Arg Thr Ser Ser  Gln Thr Ser Pro Cys  Cys Pro Thr
    1355                1360                1365

Ala Pro  Gly Ser Leu Leu Ser  Gly Gly Ser Gly Pro  Ala Leu Phe
    1370                1375                1380

Phe Ala  Gly Pro Pro Ser Pro  Leu Gln Lys Ala Pro  Val Tyr Ala
    1385                1390                1395

Gly Gly  Ser Gly Ser Val Cys  Ala Ser Ser Gly Asp  Ala Ile Ala
    1400                1405                1410

Ala Ala  Ala Leu Leu His Leu  Arg Thr Leu Gln Gln  Leu Gln Glu
    1415                1420                1425

Leu Gln  Arg His Phe Gln Arg  Pro Gly Ser Leu Pro  Pro Ala Val
    1430                1435                1440

Thr Pro  Ala Cys Leu Pro Ser  Gly Val Ala Gly Cys  Ser Pro Ala
    1445                1450                1455

Gly Leu  Gly Ala Ser Thr Pro  Gly Thr His Ser Val  Val Cys Asn
    1460                1465                1470

Ser Ser  Ala Ser Pro Val Pro  Gly Ala Ser Arg Val  Pro Arg Arg
    1475                1480                1485

Pro Asp  Gly Arg Gly Thr Gly  Gly Ala Gly Gly Asp  Pro Gly Pro
    1490                1495                1500

Ser Lys  Arg Gly Ser Val Ser  Val Ser Pro Ser Ala  Gln Gln Phe
    1505                1510                1515

Val Leu  Leu Gln Leu Lys Gln  Gln Gln Pro Gly Ser  His Gly Asn
    1520                1525                1530

Ala Leu  Ser Leu Gly Thr Gln  Gly Asn Ser Ser Asn  Pro Ala Pro
    1535                1540                1545

Ala Gly  Gly Ala Gly Ala Pro  Gln Gln His His Pro  Gly Val Cys
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    1550                1555                1560

Tyr Ser  Pro Pro Lys Asp Val  Trp Arg Ala Arg Ile  Thr Val Asp
    1565                1570                1575

Gly Arg  Gln His Glu Gln Gln  Phe Ser Val Lys Arg  His Gly Phe
    1580                1585                1590

Glu Glu  Ala Arg Leu Leu Ala  Val Gln Trp Arg Ala  His Met Glu
    1595                1600                1605

Asn Leu  Arg Leu Gly Gly Ala  Ala Lys Gly Lys Gly  Asn Ala Ser
    1610                1615                1620

Ala Ser  Ser Ala Ser Ala Ala  Thr Ala Thr Ser Gln  Gly Ser Ser
    1625                1630                1635

Gln Ser  Ser His Gln Pro Pro  Leu Gly Ser Leu Met  Val Ser Gly
    1640                1645                1650

Asn Ser  Gly Met Ser Gly Pro  Gly Ala Gly Pro Leu  Ala Asn Arg
    1655                1660                1665

Gly Leu
    1670


<210> SEQ ID NO 13
<211> LENGTH: 2330
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 13

Met Ser Asp Tyr Ala Pro Ser Arg Phe Ala Ser Pro Pro Gly Asn Ala
1               5                   10                  15

His Pro Lys Ser Pro Leu Phe Ala Arg Pro His Ser Cys Arg Glu Met
            20                  25                  30

Glu Thr Arg Ala Ser Val Gly Thr Ser Arg Gly Ser Arg Gln Pro Leu
        35                  40                  45

Cys Leu Arg Gly Ser Pro His Gly Cys Leu Ser Pro Gln Lys Gly Gln
    50                  55                  60

Asp Arg Leu Pro Ser Phe Ser Pro Leu Arg Thr Gln Pro Thr Leu Leu
65                  70                  75                  80

Ser Pro Pro Phe Pro Ser Lys Gly Cys Phe Ser Ser Cys Leu Pro Ser
                85                  90                  95

Ser Gln Ala Phe Thr Ser His Arg Ala Arg Gly Pro Ser Pro Glu Val
            100                 105                 110

His Ala Val Ser Ala Asp Ala Ser Thr Ser Ser Ser Pro Ile Ser Pro
        115                 120                 125

Ala Ser Arg Ser Ala Ser Glu Gln Gln Pro Arg Arg Glu Met Cys Ser
    130                 135                 140

Pro Pro Gly Ala Ser Ser Asp Ser Thr Ser Pro Thr Gly Ser Ser Ser
145                 150                 155                 160

Cys Ser Ala Glu Gln Asp Asp Val Leu Cys Phe Arg Gln Arg Phe His
                165                 170                 175

Leu Pro Pro Leu Leu His Leu Ser Thr Ser Arg Lys Arg Leu Arg Glu
            180                 185                 190

Glu Asp Ala Ser Ala Ser Ala Cys Ile Ser Ser Leu Gly Asn Leu Pro
        195                 200                 205

Leu Asp Val Asp Thr Lys Arg Arg Arg Gln Glu Tyr Asp Arg Leu Ser
    210                 215                 220

Thr Ala Ser Leu Ser Ser Phe Arg Ser Pro Lys Thr Pro Arg Leu Pro
225                 230                 235                 240
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Ser Cys Leu Ala Arg Arg Asp Pro Glu Glu Ser His Ala Asp Leu Ser
                245                 250                 255

Glu Ser Arg Thr Phe Leu Gln Arg Leu Glu Ala Ala Gly Gln Ser Arg
            260                 265                 270

Lys Gly Asp Thr Ser Arg Glu Thr Ile Glu Ala Asp Glu Lys Lys Val
        275                 280                 285

Leu Ser Thr His Ser Thr Asp Thr Ser Val Gln Arg Ser Pro Ser Glu
    290                 295                 300

Ser Ala Glu Arg Arg Ser Phe Gly Lys Arg Ser Asp Pro Asn Asn Gly
305                 310                 315                 320

Leu Pro Met Ala His Ser Pro Thr Pro Phe Thr Ser Lys Arg Thr Asp
                325                 330                 335

Leu Gly His Ala Leu Asp Asn Ala Leu Ser Met Arg Ala Ala Ser Arg
            340                 345                 350

Cys Gly Phe Pro Gly Pro Ala Glu Ala Thr Val Ala Pro Ala Ala Ser
        355                 360                 365

Gly Ala Ser Arg Thr Ala Ser Pro Leu Pro Phe Thr Val Pro Val Val
    370                 375                 380

Leu Ala Ala Ser Pro Pro Thr Met Pro Ser Ala Cys Ser Pro Asp Leu
385                 390                 395                 400

Cys Arg Ala Ser Thr Ser Pro Leu Ser Cys Ala Gly Val Ser Ser Leu
                405                 410                 415

Asp Ala Pro Gln Ala Val Gly Arg Arg Ser Glu Val Ala Ala Cys Val
            420                 425                 430

Ser Pro Ala Ala Ser Glu Glu Thr Val Gly Asp Thr Arg Glu His Ala
        435                 440                 445

Asp Leu Ser Ser Pro Val Ala Trp Pro Val Ala Cys Leu Ala Ser Ser
    450                 455                 460

Pro Gly Val Ala Lys Lys Pro Leu Asp Leu Gln Ile Asp Pro Glu Gln
465                 470                 475                 480

Pro Arg Gly Asn Asp Lys Leu Val Glu Pro Glu Phe Pro Gly Gly Thr
                485                 490                 495

Ala Ala Val Ser Glu Ser Ala Pro Val Ala Gly Ala Asp Ala Pro Arg
            500                 505                 510

Leu Cys Asp Tyr Gly Leu Ser Glu Ala Gly Val Leu Pro Ala Ser Gly
        515                 520                 525

Pro Trp Leu Arg Lys Pro Asn Pro Met Leu Thr Pro Asp Thr Glu Trp
    530                 535                 540

Ala Ala Pro Ser Ser Gln Glu Asp Arg Ala Cys Thr Gln Lys Glu Thr
545                 550                 555                 560

Ser Ala Ala Arg Leu Ala Pro Asn Leu Leu Tyr Arg Gln Ala Asp Ala
                565                 570                 575

Ala Ala Asp Asn Val Thr Lys Gly His Glu Asp Asp Ser Gln Phe Pro
            580                 585                 590

Leu Arg Ser Gly Ser Phe Thr Ser Ser Ala Val Ala Cys Pro Ser Pro
        595                 600                 605

Pro Asp Val Gln Ala Asp Ser Glu Ala Ala Cys Thr Trp Gly Thr Pro
    610                 615                 620

Gly Asn Gly Asp Thr Cys Glu Ser Thr Gly Gly Trp Arg Gly Ala Thr
625                 630                 635                 640

Asn Val Glu Ala His Thr Cys Leu Thr Gly Glu Asp Gly Ser Arg Tyr
                645                 650                 655

Gly Leu Gln Gly Pro Leu Ser Gln Asp Ser Pro Phe Gln Pro Pro Leu
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           660                 665                 670

Pro Ser Met Arg Pro Val His Phe Gly Gly Phe Glu Ala Trp Gly Gly
        675                 680                 685

Asn Ser Glu Ala Ser Gln Gly Asp Ala Gln Gly Leu Gln Phe Pro Arg
    690                 695                 700

Val Glu Arg Phe Ser Ser Arg Arg Thr Glu His Gly Ser Glu Gly Gly
705                 710                 715                 720

Phe Cys Gly Gln Leu Ala Gly Glu Leu Leu Pro Thr Ser Thr Ser Gly
                725                 730                 735

Gln Pro His Ser Gln His Val Ala Asp Leu Glu Ser His Thr Gly Ala
            740                 745                 750

Val Phe Ala Ser Cys Asp Pro Ala Met His Ala His Ala Ser Leu Tyr
        755                 760                 765

Gly Tyr Pro Gly Ala Phe Tyr Asn Ser Phe Gly Thr Ala Ser Ser Ile
    770                 775                 780

Phe Asp Leu Thr Gln Pro Pro Gln Ala Phe Leu Tyr Gly Gly Thr Tyr
785                 790                 795                 800

Gly Asn Asn Gly Pro Asp Asp Tyr Cys Val His Arg Thr Asn Ser Thr
                805                 810                 815

Ser Cys Gln Gly Phe Thr Ala Pro Asp Asn Ile Ser Thr Gly Thr Leu
            820                 825                 830

Asn Thr Ala Asp Ala Gln Gln Glu Trp Thr Thr Pro Ala Pro Val Ser
        835                 840                 845

Asp Ala Ala Val Gly His Trp Glu Thr Ser Asp Phe Gly Pro Gln His
    850                 855                 860

Leu Asn Gly Arg Ala Ser Ser Ser Val Pro Asp His Gly Gly Gly Leu
865                 870                 875                 880

Pro Phe Gly Gly Asn Gly Asn Ser Trp Gly Thr Ser Gly Asn Gly Asp
                885                 890                 895

Ala Trp Gly Thr Pro Gly Asn Gly Asp Thr Cys Glu Ser Thr Gly Gly
            900                 905                 910

Trp Arg Gly Thr Thr Asn Val Glu Gly His Thr Cys Leu Thr Gly Glu
        915                 920                 925

Asp Gly Ser Arg Tyr Gly Leu Gln Ala Pro Leu Ser Gln Asp Ser Pro
    930                 935                 940

Tyr Gln Pro Pro Leu Pro Pro Met Gln Pro Val His Phe Ala Asn Phe
945                 950                 955                 960

Tyr Ser Ala Cys Phe Pro Pro Leu Pro Pro Pro Pro Val Phe Pro Gly
                965                 970                 975

Ile Gly Cys Val Ser Ala Ser Tyr Pro Asp Ile Leu Leu Pro Gln Ala
            980                 985                 990

Arg Phe Leu Ser Gln Ser Cys Pro  Gly Pro Pro Ser Val  Leu Arg Cys
        995                 1000                1005

Pro Pro  Pro Ala Ala Leu Leu  Arg Gly Ser Ser Pro  Leu Asp Cys
    1010                1015                1020

Trp Ser  Leu Pro Ala Leu Pro  Ser Leu Pro Arg Ile  Pro Ser Asp
    1025                1030                1035

Phe Ala  Ser Asp Pro Ala Ser  Val Pro Leu Pro Ala  Ala Val Gln
    1040                1045                1050

Asn Leu  Pro Glu Asp Ser Pro  Arg Leu Arg Leu Pro  Cys Gln Gly
    1055                1060                1065

Ala Ser  Thr Arg Asp Gln Ser  Pro Leu Gln Tyr Glu  Gly Asn Phe
    1070                1075                1080
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Gly Gly  Ser Asp Glu Val Leu  Arg Pro Gln Val Glu  Val Ala Glu
    1085                 1090                 1095

Asn Arg  Gly Thr Pro Asn Phe  Leu Ala Ala Ser Tyr  Ser Leu Leu
    1100                 1105                 1110

Gly Ala  Phe Ser Cys Glu Gly  Asp Asn Arg Asp Asn  Glu Tyr Glu
    1115                 1120                 1125

Thr Gln  Leu Trp Gln Gln Leu  Asn Glu Ser Gly Glu  Leu Gly Val
    1130                 1135                 1140

Ser Gly  Leu Pro Gln Pro Tyr  Ser Val Glu Glu Gly  Arg Arg Gln
    1145                 1150                 1155

Glu Leu  Gln Ser Pro Tyr Pro  Ala Pro Tyr Glu Asn  Ile Pro Tyr
    1160                 1165                 1170

Ser Thr  Pro Ser Tyr Asn Ser  Val Ser Tyr Thr Ala  Ala Ser Lys
    1175                 1180                 1185

Asp Arg  Leu Val Gly Asp Asn  Thr Ala Tyr Asn Gly  Ala Ala Tyr
    1190                 1195                 1200

Cys Pro  Phe Tyr Gly Gly Ser  Gly Met Tyr Glu Thr  Pro Gln Arg
    1205                 1210                 1215

Ser Glu  Glu Asn Ser Leu Tyr  Ser Ala Asp Pro Gln  Val His Phe
    1220                 1225                 1230

Ser Glu  Ser Glu Lys Thr Gly  Ser Ser Asp Ser Phe  Pro Tyr Ser
    1235                 1240                 1245

Phe Phe  Ser Leu Gly Thr Pro  Ala Leu Tyr Pro Gly  Gly Ser Leu
    1250                 1255                 1260

Gln Thr  Gly Ala His Leu Glu  Glu Val Pro Gly Ser  Gly Asp Ala
    1265                 1270                 1275

Glu Gly  Ser Ala Trp Ser Pro  Ser Leu Glu Ser Arg  Arg Leu Arg
    1280                 1285                 1290

Gly Arg  Thr Arg Ser Pro His  Ala Gln Ser Pro Asn  Ser Asn Arg
    1295                 1300                 1305

Ser Arg  Gly Ala Ala Trp Thr  Phe Ser Pro Ala Ser  Leu Pro Phe
    1310                 1315                 1320

Glu Val  Pro Ala Ala Ala Lys  Ala Ser Gly Arg Lys  Arg Arg Ala
    1325                 1330                 1335

Pro Gly  Ser Leu Pro Ala Gln  Thr Asp Arg Gly His  Lys Asp Phe
    1340                 1345                 1350

Leu Leu  Glu Leu Leu Ala Ser  Arg Leu Glu Pro Val  Lys Gly Val
    1355                 1360                 1365

His Met  Asp Arg Leu Arg Lys  Thr Trp Val Ala Ser  Trp Leu Val
    1370                 1375                 1380

Gly Lys  Arg Arg Ile Thr Arg  Ile Phe Ser Phe Gln  Lys Phe Gly
    1385                 1390                 1395

Phe Phe  Gly Ala Arg Glu Gln  Ala Ile Arg His Arg  Arg Glu Ala
    1400                 1405                 1410

Leu Leu  Asn Pro Glu Leu Asp  Asn Ser Glu Arg Arg  Glu Ala Leu
    1415                 1420                 1425

Ala Asn  Val Glu Arg Ala Thr  Asp Asp Glu Leu Gln  Gln Ala Ala
    1430                 1435                 1440

Asp Ala  Leu Pro Phe Val Val  Gly Val Thr Tyr His  Arg Ala Ser
    1445                 1450                 1455

Arg Cys  Trp Val Ala Asn His  Arg Lys Pro Met Gly  Lys Ile Val
    1460                 1465                 1470
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Gln Arg  Lys Lys Phe Ala Val  Ala Glu Leu Gly Phe  Leu Glu Ala
    1475                1480                1485

Arg Tyr  His Ala Ala Val Met  Met Phe Cys Trp Asn  Lys Gln Gly
    1490                1495                1500

Arg Thr  Gln Glu Pro Glu Asp  Tyr Asp Gln Gly Ala  Thr Glu Ala
    1505                1510                1515

Phe Asn  Ser Arg Gln Val Pro  Gln Arg Pro Gly Asp  Asp Arg Ala
    1520                1525                1530

Phe Glu  Phe Ser His Pro Thr  Cys Ser Glu Asn Glu  Pro Leu Tyr
    1535                1540                1545

Thr Leu  Lys Ala Leu Asp Ser  Gly Thr Cys Asp Asp  Ala Met Val
    1550                1555                1560

Leu Leu  Ala Phe Ile Cys Gly  Ser Pro Trp Arg Lys  Ile Cys Arg
    1565                1570                1575

Gly Gln  Gln Cys Gly Asp Asp  Pro Thr Leu Leu Glu  Ala Ala Ser
    1580                1585                1590

Thr Ile  Gln Thr Glu Lys Ser  Leu Trp Arg Thr Arg  Val Lys Ser
    1595                1600                1605

Ala Ala  Asp Glu Val Arg Glu  Gly Pro Gln Arg Arg  Leu Glu Gly
    1610                1615                1620

Thr Asp  Ala Gly Asp Ser Gly  Ala Phe Pro Arg Gly  Gln Ser Pro
    1625                1630                1635

Glu Lys  Gly Arg Pro Arg Arg  Arg Arg Lys Thr Ala  Thr Leu Arg
    1640                1645                1650

Glu Gln  Glu Asp Val Thr Glu  Asp Lys Thr Glu Asp  Gly Arg Glu
    1655                1660                1665

Asp Lys  Thr Glu Asp Gly Gly  Glu Asp Lys Thr Glu  Asp Gly Gly
    1670                1675                1680

Glu Asp  Gly Arg Glu Asp Glu  Gly Glu Asp Glu Gly  Glu Asp Pro
    1685                1690                1695

Gly His  Gly Trp Gly Glu Arg  Arg Arg Cys Arg Lys  Ser Asp Arg
    1700                1705                1710

Glu Asn  Ala Gly Glu Ala Glu  Arg Gly Gln Lys Arg  Glu Lys Arg
    1715                1720                1725

Gln Gln  Ser Glu Gly Arg Cys  Val Val Ala Glu Val  Asp Leu Arg
    1730                1735                1740

Asp Ala  Lys Asp Thr Val Val  Arg Arg Asn Arg Val  Ala Arg Arg
    1745                1750                1755

Glu Gly  Leu Glu Thr Gly Phe  Gly Lys Lys Asn Ala  Lys Ser Gly
    1760                1765                1770

Ala Glu  Ser Cys Leu Ser Gln  Thr Pro Ala Leu Gly  Pro Ser Ser
    1775                1780                1785

Pro Pro  Phe Pro Val Ser Phe  Lys Lys Arg Arg Lys  Ser Ser Ser
    1790                1795                1800

Arg Glu  Ala Asp Leu Arg Gln  Ser Arg Pro Arg Arg  His Arg Asn
    1805                1810                1815

Asp Thr  Glu Glu Ala Arg Ser  Ile Cys Glu Asp Ser  Pro Ser Ser
    1820                1825                1830

Glu Val  Ala Pro Thr Pro Ala  Ser Ser Ser Phe Ser  Pro Ala Ala
    1835                1840                1845

Ser Leu  Ser Ser Asp Gly Ser  Arg Leu Gly Ser His  Asn His Asp
    1850                1855                1860

Leu Thr  Asp Ser Gly Arg Ser  Ala Ser Val Ser Arg  Gly Arg Ser
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    1865                1870                1875

Thr Asp  Phe Ser Met Phe Ala  Gly Leu Pro Tyr Leu  Lys Ser Leu
    1880                1885                1890

Glu Ser  Asn Thr Arg Phe Val  Pro Pro Ser Arg Pro  Gly Glu Ser
    1895                1900                1905

Gly Leu  Pro Asn Val Tyr Ala  Ser Tyr Asn Ser Gly  Leu Ala Phe
    1910                1915                1920

Glu Ala  Asn Arg Pro Cys Pro  Leu Ala Phe Asp Ser  Arg Ala Asp
    1925                1930                1935

Pro Thr  Gly Trp Pro His Thr  Phe Pro His Pro Ala  Glu Ala Tyr
    1940                1945                1950

Gly Ser  Gly Ile Ala Ser Trp  Thr Pro Asn Ala Asn  Gly Phe Phe
    1955                1960                1965

Glu Ser  Leu Ala Tyr Thr Gly  Asn Leu Glu Glu Leu  Arg Asp Leu
    1970                1975                1980

Cys Gly  Arg Thr Pro Asp Ala  Arg Asp Ser Ser Asp  His Trp Gln
    1985                1990                1995

Glu Ala  Ala Ala Ala Ala Ser  Ser Arg Leu Pro Leu  Arg Pro Pro
    2000                2005                2010

Ala Val  His Ser Trp Gln Asp  Ala Pro Cys Ala Lys  Asp Pro Ala
    2015                2020                2025

Pro Cys  Val Glu Leu Ala Arg  Asp Glu Cys Leu Ala  Gly Gly Asp
    2030                2035                2040

Arg Gln  Thr Cys Arg Phe His  Ser Ala Phe Asp Ser  Ala Asp Ala
    2045                2050                2055

Gly Asp  Tyr Lys Phe Asn Thr  Asp Ala Arg Cys Leu  Arg Gly Pro
    2060                2065                2070

Thr Phe  Asn Pro His Ser Asn  Ala Val Ala Thr Leu  Arg Arg Glu
    2075                2080                2085

Gln Glu  Ala Ala Arg Gly Ala  Ser Gly Gln Thr Pro  Ser Phe Phe
    2090                2095                2100

Phe Pro  Arg Leu Val Pro Val  Ala Gln Thr Asp Trp  Glu Ala Asp
    2105                2110                2115

Pro Gly  Arg Gly Ser Gly Asp  Ser Leu Ser Ala Pro  His Glu Ala
    2120                2125                2130

Gly Glu  Ala Val Gly Val Glu  Gly Ser Glu Gly Ala  Pro Cys Glu
    2135                2140                2145

Trp Asn  Phe Glu Arg Asp Ala  His Pro Val Ile Leu  Pro Thr Ser
    2150                2155                2160

Asn Cys  Ser His Gly His Glu  Arg Leu Ala Ser Ser  Asn Ala Phe
    2165                2170                2175

Thr Glu  Ala Lys Gln Arg Asn  Ala Leu Arg Cys Thr  Pro Gln Glu
    2180                2185                2190

Thr Val  Gly Gly Val Asn Glu  Asn Gly Ser Pro Leu  Phe Ser Thr
    2195                2200                2205

His Arg  Asp Ala Pro Glu Ala  Met Ser Ala Leu Thr  Glu Val Ser
    2210                2215                2220

Asp Arg  Glu Thr Gln Arg Gly  Pro Ala Val Leu Gln  Ser Gly Asn
    2225                2230                2235

Thr Glu  Ala Leu Leu Gln Asp  Ser Thr Ser Asn Ser  Ala Ser Pro
    2240                2245                2250

Thr Gln  Arg Arg Ala His Gly  Leu Asp Pro Glu Pro  Asp Glu Ser
    2255                2260                2265
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Lys Ala  Arg Gly Glu Arg Ser  Lys Glu Glu Asp Arg  Glu Thr Leu
    2270                2275                2280

Arg Thr  Glu Ala Pro Ser Lys  Gly Arg Lys Gln Ile  Leu Ser Pro
    2285                2290                2295

Pro Thr  Glu Arg Asn Ser Met  Tyr Gly Glu Ala Met  Ser Ile Asp
    2300                2305                2310

Arg Gln  Val Ser Ala Leu Pro  Thr Leu Leu Ser His  Gly Thr Ala
    2315                2320                2325

Phe Pro
    2330


<210> SEQ ID NO 14
<211> LENGTH: 1438
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 14

Met Ala Ala Ala Ser Pro Pro Ala Gln Pro Leu Gly Ala Thr Ser Pro
1               5                  10                  15

Cys Thr Phe Ser Pro Pro Cys Ser Phe Ser Pro Ser Asp Thr Cys Ser
            20                  25                  30

Val Phe Phe Ala Thr Pro Ser Arg Ala Val Ser Ala Val Pro Glu Leu
        35                  40                  45

Pro Ala Thr Ser Ser Ala Gln Leu Pro Glu Arg Thr Arg Leu Arg Asn
    50                  55                  60

Arg Ser Ile Gln Ser Ala Ser Thr Thr Glu Ala Ser Pro Phe Val Asp
65                  70                  75                  80

Ser Ala Ser Leu Phe Pro Glu Ser Leu Ser Glu Ala Pro Lys Ala Val
                85                  90                  95

Ser Val Asp Gly Glu Ser Arg Arg Thr Arg Glu Arg Arg Arg Lys Ser
            100                 105                 110

Arg Ser Leu Leu Ala Ala Ala Glu Glu Thr Pro Glu Ala Thr Ala Ala
        115                 120                 125

Ser Pro Asn Gly Ser Ser Ser Glu Ile Ser Asp Glu Ala Ser Thr Phe
    130                 135                 140

Val Leu Thr Pro Ala Thr Ala Ser Leu Ala Pro Ala Ala Leu Pro Pro
145                 150                 155                 160

Phe Met Thr Glu Arg Ser Asp Pro Thr Glu Lys Lys Tyr Glu Ala Gln
                165                 170                 175

Asn Met Gln Val Thr Ala Val Glu Pro Val Gly Leu Ala Pro Arg Ser
            180                 185                 190

Ala Ser Arg Ser Glu Leu Gly Asp Ala Glu Ser Leu Ser Ala Gly Lys
        195                 200                 205

Ser Gly Leu Gln Gly Glu Ser Ala Ala Pro Thr Ala Ala Leu Glu Ala
    210                 215                 220

Asp Ala Gly Glu Thr Arg Leu Glu Thr Gly Leu Ala Gly Glu Pro Val
225                 230                 235                 240

Asn Ser Ser Ser Glu Gly Val Gly Tyr Gly Gly Asp Glu Gln Thr Val
                245                 250                 255

Ala Gly Glu Thr Arg Glu Pro Gly Thr Ala Glu Glu Lys Leu Gly Asp
            260                 265                 270

Leu Lys Pro Glu Val Arg Pro Arg Phe His Ala Tyr Ala Glu Gln Asp
        275                 280                 285

Val Cys Ala Trp Ala Thr Ser Met Leu Ala Arg Lys Glu Leu Arg Lys
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    290                 295                 300

Leu Lys Ala Ala Ala Val Lys Leu Glu Gly Asp Arg Met Pro Arg Ser
305                 310                 315                 320

Asp Val Val Gly Leu Tyr Phe Lys Lys His Arg Pro Cys Trp Ser Val
                325                 330                 335

Asp Tyr His Thr Arg Gln Gly Lys Arg Lys Thr Val Glu Phe Phe Val
            340                 345                 350

Pro Asp Leu Ser Arg Glu Thr Ile Glu Leu Val Leu Val His Ala Ile
        355                 360                 365

Glu Cys Arg Lys Tyr Met Pro Arg Arg Phe Asp Gln Ala Pro Ala Phe
    370                 375                 380

Val Pro Glu Pro Asp Asp Thr Thr Ser Gly Met Pro Tyr Arg Tyr Gly
385                 390                 395                 400

Ala Arg Leu Leu Ser Pro Lys Val Leu Ala Trp Ile Val Glu Asn Thr
                405                 410                 415

Asn Gly Ser Gly Arg Thr Ser Gln His Gly His Gly Gln Ser Arg Arg
            420                 425                 430

Leu Glu Gly Asp Lys Val Gly Glu Gly Asp Ala Gly Ala Gln Leu Leu
        435                 440                 445

Ser Gly Pro Ala Gly Ile Asp Ala Phe Gly Ser Arg Ser Pro Arg Ala
    450                 455                 460

Gly Phe Ala Arg Gln Arg Asp Asn Asn Ser Arg Arg Gln Gly Lys Ala
465                 470                 475                 480

Ser Gly Cys Arg Pro Gly Ala Asp Leu Ala Thr Ser Ser Glu Glu Lys
                485                 490                 495

Ala Thr Arg Glu Gly Glu Thr Glu Leu Pro Gly Gly Ser Ala Gly Pro
            500                 505                 510

Gly Ser Val Pro Ala Gly Thr Ala Tyr Gly Asp Tyr Ala Arg Gln Leu
        515                 520                 525

Pro Ser Glu Gly Tyr Gln Thr Pro Pro Thr Met Glu Gly Arg Met Thr
    530                 535                 540

Pro Ala Gly Leu Leu Ser Gly Gln Glu Phe Gly His Gly Gln Gly Met
545                 550                 555                 560

Gln Gly Ala Gly Val Met Trp Arg Asp Asp Pro Arg Gln Ala Leu Gln
                565                 570                 575

Ala Met Pro Gln Pro Leu Asn Leu Ala Pro His Ala Thr Pro Phe Met
            580                 585                 590

Ser Arg Ala Gly Gly Leu Tyr Asp Gln Arg Glu Ala Ser Val Glu Pro
        595                 600                 605

Gly Arg Asp Val Tyr Pro Val His Tyr Pro Thr Pro Tyr Ala Tyr Gly
    610                 615                 620

Pro Gly Ile Pro Ala Asp Ala Gly Ala Pro Ser Ala Gly Pro Gly Pro
625                 630                 635                 640

Tyr Pro His Gln Phe Pro Ser Gly Gly Ala Gly Tyr Val Val Asn Gly
                645                 650                 655

Arg Val Pro Asp Ser Ala Asp His Glu Ala His Ser Pro Arg Ser Pro
            660                 665                 670

Glu Ser Tyr Trp Gly Pro Gln Ala Gly Ser Gln Gly Ala Glu Asp Lys
        675                 680                 685

Asp Cys Gln Val Val Gly Cys Met Leu Pro Asn Gly Ser Glu Met Ala
    690                 695                 700

Met Arg Arg Met Glu Ser Tyr Val Gly Asp Arg Asp Asn Leu Arg Gly
705                 710                 715                 720
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Ser Ala Ala Phe Ala Gly Asp Gly Arg Thr Gln Ala Glu Gly Leu Ser
                725                 730                 735

Pro Gln Cys Glu Pro Asn Ala Lys Arg Arg Arg Leu Gln Ala Gly Gly
            740                 745                 750

Asp Gly Ser Asn Gly Gly Leu Glu Ala Ser Gly Pro Glu Arg Pro Phe
        755                 760                 765

Pro Gly Ser Gln Met Leu Gln Pro Ser Asp Glu Trp Ala Arg Asn Gly
    770                 775                 780

Gln Arg Ala Phe Ala Val Gln Pro Gly Thr Gly Gly Arg Thr Phe Met
785                 790                 795                 800

Asn Gly Gly Phe Arg Gln Pro Gly Pro Glu Asp Ala Arg Gln Pro Leu
                805                 810                 815

Leu Leu Ser Ser Ala Pro Tyr Ser Pro Pro Ser Val Phe Pro Ala Ala
            820                 825                 830

Pro Pro His Leu Ser His Ala Val Arg Leu Pro Pro Gly Ser Ser Asp
        835                 840                 845

Ala Ala His Arg Thr Pro Met Ser Gly Ala Ala Gly Cys Ala Ser Pro
    850                 855                 860

Val Ala Ser Ala Phe Arg Lys Glu Ala Glu Ala Ser Glu Trp Pro Ser
865                 870                 875                 880

Asn Glu Val Tyr Gly Ser Pro Gln Ala Phe Pro Asp Lys Ala Asn Ala
                885                 890                 895

Phe Ala Lys Gly Val Thr Leu Pro Arg Arg Gln Ser Phe Ala Phe Ser
            900                 905                 910

Asp Ala Gly Leu Pro Thr Pro Thr Thr Ser Pro His His Gly Ser Tyr
        915                 920                 925

Cys Ala Ser Thr Ile Ala Ser Ser Ser Pro Lys Ser Ala Ser Pro Val
    930                 935                 940

Ser Gln Ser Gly Cys Phe Pro Cys Asp Phe Tyr Pro Ala Thr Ala His
945                 950                 955                 960

Tyr Ser Gly Pro Gly Val Glu Thr Pro Ser Asp Val Ser Ser Phe Val
                965                 970                 975

Pro Ala Pro Ala Glu Thr Ala Glu Gln Gln Ile His Gly Ala Gly Gln
            980                 985                 990

Ala Ala Val Lys Thr Pro Glu Ser  Gly Leu His Met Pro  Ser Ser Gly
        995                 1000                1005

Trp Pro  Gln Gln Ala Ser Val  Pro Gly Ala His Gly  Ala Glu Phe
    1010                1015                1020

Tyr Ala  Ser Arg Ala Phe Ala  Asn Gly Ala His Ala  Pro Ser Leu
    1025                1030                1035

Ser Leu  Arg Pro Ser Trp Arg  Tyr Pro Gly Gly Glu  Arg Ser Glu
    1040                1045                1050

Gly Asp  Leu Thr Thr Gln Glu  Gln Asn Ala Pro Ala  Gly Ala Ser
    1055                1060                1065

Pro Ser  Ser Pro Val Trp Ser  Gly Asn Thr Gly Val  Cys Thr Thr
    1070                1075                1080

Glu Gly  Cys Gly Val Trp Leu  Glu Asn Arg Gln Ala  Ala Gly Ser
    1085                1090                1095

Val Glu  Gly Ala Ala Asp Pro  Gly Val Gln Gly Ser  Ala Cys Met
    1100                1105                1110

Gln Gly  Lys Pro Gln Glu Gly  Gly Arg Cys Ser Pro  Glu Pro Ala
    1115                1120                1125
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Leu Gly  Val Arg Arg Pro Ala  Glu Phe Ala Gly Ala  Pro Val Gly
    1130                1135                1140

Ala Cys  Arg Ala Val Glu Asp  Arg Thr Met Thr Gly  Glu Arg Gly
    1145                1150                1155

Ala Trp  Gly Asn Glu Ala Arg  Arg Glu Thr Val Thr  Gly Asp Gln
    1160                1165                1170

Glu Cys  Cys Gly Asp Gln Ala  Arg Asp Pro Met Val  Phe Ser His
    1175                1180                1185

Met Gly  Ser Arg Ala Glu Leu  Ser Gly Phe Asp Asp  Gly Ser Glu
    1190                1195                1200

Leu Pro  Pro Ala Ser Pro Leu  Asn Glu Cys Met His  Pro Leu Gly
    1205                1210                1215

Lys Pro  Gly Ser Arg Ile Phe  Pro Glu Phe Gly Ala  Trp Pro Gly
    1220                1225                1230

Ser Pro  Pro His Glu Gly Ser  Phe Val Gln Glu Phe  Asp Ile Phe
    1235                1240                1245

Lys Glu  Asn Gly Glu Gly Ala  Ala Gly Ala Val Asp  Asp Ala Met
    1250                1255                1260

Ala Leu  Trp Pro Asn Gly Gly  Ala Phe Gly Gln Arg  Thr Asp Pro
    1265                1270                1275

Leu Ala  His Glu Glu Glu Lys  Glu Gly Glu Leu Trp  Lys Gly Gln
    1280                1285                1290

Pro Thr  Pro Phe Cys Ser Ser  Pro Ala Leu Trp Cys  Val Cys Pro
    1295                1300                1305

Val Glu  His Thr Arg Glu Phe  Asp Val Met Asp Met  Val Thr Leu
    1310                1315                1320

Pro Asp  Leu Ser His Thr Ala  Gly Pro Val Ser Arg  Pro Leu Pro
    1325                1330                1335

Asn Ala  Pro Leu Cys Gly Gly  Cys Val Val Ala Gly  Val Gly Glu
    1340                1345                1350

Ala Gln  Ala Gly Asp Gly Glu  Ser Lys Gln Gly Ala  Lys Leu Ala
    1355                1360                1365

Pro Asp  Ser Gln His Leu His  Gly Gly Ala Ala Asn  Pro Gly Ala
    1370                1375                1380

Val Gly  Lys Leu Val Thr Asp  Glu Thr Ala Gln Thr  Ser Gly Arg
    1385                1390                1395

Glu Gln  His Pro Gly Glu Gly  Asp Ser Thr Glu Gln  Arg Leu Ser
    1400                1405                1410

Gly Leu  Ala Ala Arg Ala Thr  Pro Gln Arg Glu Thr  Lys Arg Pro
    1415                1420                1425

Gly Pro  Ser Arg Arg Thr Glu  Gly Glu Leu
    1430                1435


<210> SEQ ID NO 15
<211> LENGTH: 3817
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 15

Met Asp Phe Glu Arg Gly Glu Ser Pro Gly Asp Ser Arg Gly Ser Val
1               5                  10                  15

Ala Phe Leu His His Thr Glu Lys Leu Glu Arg Leu Pro Gly Thr Gly
            20                  25                  30

Glu Thr Thr Ile Arg Gly Met Ser Phe Thr Pro Pro Tyr Ile Cys Met
        35                  40                  45
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Glu Arg Pro Pro Arg Ala Asn Cys Glu Ala Leu Arg Leu Lys Ser Pro
    50                  55                  60

Pro Glu Asn Arg Ala Phe Ser Ser Arg Ser Glu Ser Pro Ser Pro Thr
65                  70                  75                  80

Pro Phe Ala Arg Glu Cys Ala Ser Leu Gly Leu Val Trp Gly Asp Glu
                85                  90                  95

Gly Thr Arg Ala Gly Leu Leu Arg Thr Arg Leu Phe Thr Pro Pro Asp
            100                 105                 110

His Thr Pro His Leu Leu Ala Glu Thr Gly Ala Ser Cys Leu Glu Asp
        115                 120                 125

Leu Phe Pro Gly Thr Val Pro Gln Leu Leu Ser His Leu Pro Ser Pro
    130                 135                 140

Pro Pro Val Pro Gly Val Ala Arg Ala Ser Arg Ser Ser Ser Pro Leu
145                 150                 155                 160

Ala Gly Gly Ser Ser Leu Ala Cys Ala Ser Pro Trp Pro Arg Ser Ala
                165                 170                 175

Thr Pro Phe Phe Ala Ala Asn Met Pro Ala Leu Leu Pro Gly Arg Gly
            180                 185                 190

Pro Met Arg Val Thr Lys Trp Leu Asp Gly Gln Val Ser Asp Pro Gln
        195                 200                 205

Ser Asp Ser Cys Leu Arg Gly Gly Ala Ser Arg Val Glu Arg Ala Ala
    210                 215                 220

Ala Leu Leu Cys Gly Arg Ser Glu Glu Glu Gln Glu Arg Glu Arg Ser
225                 230                 235                 240

Val Asp Glu Arg Arg Leu Arg Lys Ala Ile Gly Val Thr Asp Glu Asp
                245                 250                 255

Glu Ser Glu Arg Glu Arg Glu Thr Glu Gly Gly Val His Glu Arg Leu
            260                 265                 270

Ser Arg Cys Ala Ala Ala Thr Ala Ala Asp Arg Ala Asn Asn Leu Leu
        275                 280                 285

Gly Leu Gly Val Glu Arg Gly Pro Glu Val Ala Gly Gly Arg Leu Gly
    290                 295                 300

Gly Tyr Trp Thr Thr Glu Ser Glu Val Tyr Pro Gln Arg Ile Gly Glu
305                 310                 315                 320

Leu Glu Gly Glu Gly Leu Gly Ser Pro Asp Pro Val Ala Ala Ser Ala
                325                 330                 335

Leu Val Thr Ala Val Gln Asp Ser Arg Glu Asn Leu Asn Cys Leu Thr
            340                 345                 350

Gly Val Leu Thr Thr Leu Arg Leu Ser Ser Arg Asp Ser Glu Gly Asp
        355                 360                 365

Phe Asp Leu Pro Leu Phe Val Gln Ser Arg Lys Trp Arg Ala Lys Tyr
    370                 375                 380

Asn Arg Arg Ser Leu Asp Leu Lys Gly Thr Val Ala Arg Ser Lys Ala
385                 390                 395                 400

Leu Gly Tyr Pro Ala Gly Leu Gln Ile Pro Glu Thr Tyr Arg Asp Leu
                405                 410                 415

Lys Asn Cys Met Gln Arg Pro Pro Ser Ile Asp Ala Ala Asp Ser Arg
            420                 425                 430

Ala Trp Arg Ser Ala Glu Ala Pro Arg Ala Ala Lys Lys Val Phe Ser
        435                 440                 445

Glu Gly Arg Arg Ala Thr Asp Arg Asp Glu Gln Val Ala Phe Val Glu
    450                 455                 460
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```
Asp Glu Val Thr Glu Gln Leu Leu Phe Asn Ala Asn Ala Ala Val Glu
465                 470                 475                 480

Gly Thr Thr Leu Tyr Asn Asn Leu Leu Cys Lys Tyr Gly Leu Glu Thr
                485                 490                 495

Arg Cys Phe Ser Thr Ser Ser Ala Pro Gly Asn Thr Ala Phe Glu Ser
            500                 505                 510

Arg Leu Ala Arg Ser Asp Ala Asp Pro Thr Ala Ser Ser Gln Ser Ala
        515                 520                 525

Ser Ala Leu Ser His Ala Ala Val Ser Pro Ser Leu Ala Ser Ala Leu
    530                 535                 540

Pro Val Ser Ser Leu Leu Leu Glu Asp Ala Ala Asp Ala Val Gly Asp
545                 550                 555                 560

Arg Ser Glu Leu Glu Thr Gly Ser Gln Ala Glu Ala Ala Ile Pro Thr
                565                 570                 575

Ser Glu Ala Ser Cys Met Arg Arg Glu Lys His Val Gly Glu Glu Ser
            580                 585                 590

Arg Ala Asp Lys Gly Ala Phe Leu Arg Ser Ala Ser Asp Ser Thr His
        595                 600                 605

Ala Glu Glu Asp Gly Leu Ser Gly Gly Lys Asp Ala Ser Ser Arg Glu
    610                 615                 620

Gly Gly Ser Glu Glu Arg Glu Glu Ala Ala His Glu Ala Ala Asp Ser
625                 630                 635                 640

Leu Trp Ser Leu Val Leu Asn Arg Asn Ile Ala Ala Leu Pro Gly Phe
                645                 650                 655

Met Thr Val Gly Arg Tyr Glu Cys Asp Leu Leu Pro Lys Arg Ser Ala
            660                 665                 670

Phe Ser Arg Lys Gln Leu Ala Gly Leu Val Ala Gly Ser Arg Pro Leu
        675                 680                 685

Pro Val Leu Pro Ser Ser Ser Asp Thr Pro Gly Ser Ala Ser Thr Glu
    690                 695                 700

Leu Leu Ala Glu Arg Val Ala Cys Ala Leu Thr Leu Asp Glu Gly Glu
705                 710                 715                 720

Ala Trp Asn Pro Ser Asp Ala Ser Asp Leu Asp Asp Phe Leu Glu Ser
                725                 730                 735

Ser Cys Ala Pro Asn Ala Leu Arg Arg Gly Arg Gln Ala Val Val Pro
            740                 745                 750

Val Arg Gly Ala Arg Arg Arg Arg Gly Ala Asp Leu Gly Leu Ser Pro
        755                 760                 765

Pro Pro Ser Ser Pro Ala Val Arg Cys Arg Ser Leu Val Arg Trp Ser
    770                 775                 780

Gln Gln Arg Pro Phe Phe Ser Asn Val Ser Ala Cys Ala Gly Ala Ala
785                 790                 795                 800

Asp Ser Arg Arg Glu Glu Trp Lys Asp Ala Gly Lys Val Ala Lys Pro
                805                 810                 815

Gly Ser Glu Ser Ala Leu Thr Ser Arg Asp Leu His Ala Ser Thr Gly
            820                 825                 830

Leu Val Asn Ala Ala Leu Asp Ser Ser Glu Gln Lys Ser Gly Glu Arg
        835                 840                 845

Glu Ser Ser Leu Ser Pro Gln Glu Arg Ile Leu Thr Gln Val Lys Lys
    850                 855                 860

Glu Leu Glu Asp Glu Arg Val Arg Glu Lys Gln Thr Ile Arg Asp Lys
865                 870                 875                 880

Asp Ser Glu Lys Gly Gln Gly Gly Glu Ser Asn His His Met Pro Gly
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```
              885                 890                 895

Thr Ala Asn Gly Gln Arg Thr Pro Asn Glu Gly Glu Ala Pro Met Glu
           900                 905                 910

Thr Glu Glu Ala Pro Thr Leu Glu Pro Ser Asn Gly Met His Arg Asp
       915                 920                 925

Gly Gln Asp Ala Gly Ala Arg Met His Ser Ser Ser Thr Arg Ala Leu
   930                 935                 940

Glu Gly Ala Val Glu Asp Glu Pro Lys Val Thr Leu Pro Asp Lys Asp
945                 950                 955                 960

Glu Pro His Ala Ser Ala Leu Cys Gly Glu Arg Glu Lys Gln Arg Gln
               965                 970                 975

Ser Phe Phe Ser Ser Val Ser Ser Arg Glu Asp Ala Gln Asp Glu Asp
           980                 985                 990

Ser Arg Trp Cys Val Ala Gly Gly  Met Tyr Asn Gly Trp  Lys Gly Thr
       995                 1000                1005

Tyr Asp  Val Trp Ile Tyr Arg  Arg Val Ser Ala Ala  Leu Arg Glu
   1010                1015                1020

Gly Lys  Gly Glu Glu Glu Lys  Arg Arg Glu Gly Glu  Lys Arg Lys
   1025                1030                1035

Thr Gly  Lys Gly Lys Gln Ser  Val His Thr Ala Ser  Leu Gly Ala
   1040                1045                1050

Gly Gly  Ala Gln Gly Leu Ser  Pro Gly Glu Thr Gln  Ala Ser Gly
   1055                1060                1065

Leu Ala  Pro Gly Ser Thr Pro  Leu Gly Ser Ala Gly  Thr Leu Ser
   1070                1075                1080

Ala Gly  Arg Asn Gly Glu Glu  Thr Arg Glu Ser Thr  Gly Ser Pro
   1085                1090                1095

Ala Gly  Ala Phe Ala Ser Ser  Ser Ser Leu Ala Ala  Lys Gly Gln
   1100                1105                1110

Asn Gly  His Ala Ser Val Glu  Asp Leu Lys Thr Gln  Lys Glu Glu
   1115                1120                1125

Ser Leu  Gly Cys Val Leu Ser  Ala Ser Ala Leu Pro  Leu Asn Pro
   1130                1135                1140

His Ser  Gly Glu Thr Arg Glu  Asp Ser Ala Gly Arg  Asp Glu Glu
   1145                1150                1155

Lys Gly  Glu Glu Arg Glu Arg  Asp Glu Asn Glu Pro  Pro Leu Tyr
   1160                1165                1170

Glu Trp  Arg Val Lys Arg Phe  Ser Ala Leu Ile His  Gly His Glu
   1175                1180                1185

Lys Ala  Ser Arg Leu Ala Cys  Lys Tyr Cys Val Tyr  Leu Glu Arg
   1190                1195                1200

Phe Gly  Arg Ile Arg Gly Arg  Leu Ser Ile Cys Ser  Thr Cys Cys
   1205                1210                1215

Arg Asp  Ala Cys Ser Gly Cys  Met Pro Ser Lys Lys  Arg Ala Ala
   1220                1225                1230

Gly Ala  Asp Phe Ser Pro His  Cys Arg Asn Gly Arg  Asp Ala Gly
   1235                1240                1245

Val Gly  Gly Ala Gly Arg Ala  Pro Lys Arg Arg Val  Gln Ala Lys
   1250                1255                1260

Lys Gly  Ala Ala Gly Ala Ala  Gly Val Cys Gly Asp  Arg Ala Arg
   1265                1270                1275

Lys Gly  Lys Gly Glu Asp Glu  Pro Glu Arg Asp Gly  Leu Asp Arg
   1280                1285                1290
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Arg Glu  Glu Gly Gly Thr Pro  Ser Ser Lys Gln Thr  Ala Glu Arg
    1295                 1300                 1305

Arg Gly  Ala Ala Lys Lys Glu  Gly Arg Glu Glu Asp  Asp Arg Val
    1310                 1315                 1320

Asp Gly  Lys Gly Thr Ser Leu  Ser Leu Glu Asn Asn  Ser Phe Glu
    1325                 1330                 1335

Ser Ser  Cys Pro Ala Met Arg  Ser Ser Leu Arg Ala  Ser Phe Glu
    1340                 1345                 1350

Val Lys  Gly Pro Leu Ser Pro  Ser Ser Ala Asp Asp  Arg Pro Asn
    1355                 1360                 1365

Glu Gly  Ala Ala Gly Arg Gly  Ala Pro Pro Gly Ser  Glu Gly Pro
    1370                 1375                 1380

Ser Arg  Asp Leu Ala Leu Arg  Ser His Ser Phe Ser  Ser Ala Ser
    1385                 1390                 1395

Ser Ser  Arg Lys Ser Ala Lys  Asn Ala Ala Glu Ser  Leu Arg Arg
    1400                 1405                 1410

Ile Ala  Gly Pro Leu Phe Arg  Ser Ser Gly Asp Leu  Thr Ala Ser
    1415                 1420                 1425

Gln Leu  Gly Ala Glu Thr Glu  Glu Ser Asp Val Leu  Gln Asp Val
    1430                 1435                 1440

Phe Glu  Leu Tyr Ser Glu Ala  Gly Glu Ala Trp Glu  Thr Cys Thr
    1445                 1450                 1455

Thr Pro  Val Ser Phe Ser Pro  Ser Leu Ser Val Ala  Ser Arg Asp
    1460                 1465                 1470

Thr Leu  Val Val Leu Gly Gly  Ser Gln Thr Thr Ala  Val Ala Arg
    1475                 1480                 1485

Leu Asp  Ser Gly Lys Met Ser  Glu Ala Val Arg Arg  Ser Ser Asn
    1490                 1495                 1500

Ala Leu  Ser Ala Ala Ala Ser  Ser Phe Pro Lys Gly  Lys Gly Phe
    1505                 1510                 1515

Gly Gly  Ala Ser Lys Lys Thr  Asp Ser Val Thr Leu  Ser Phe Leu
    1520                 1525                 1530

Ala Arg  Val Cys Arg Asn Leu  Arg Met Phe Leu Leu  Leu Cys Gln
    1535                 1540                 1545

His Asn  Thr Val Ala Gly Gly  Leu Pro Gly Asp Ser  Lys Cys Val
    1550                 1555                 1560

Cys Arg  Ala Gln Ser Gly Pro  Gly Gly Ala Gly Leu  Ala Gly Ala
    1565                 1570                 1575

Asp Gly  Arg Ala Pro Gly Asp  Leu Gly Asp Ser Lys  Gly Thr Ala
    1580                 1585                 1590

Ile Ala  Arg Gly Pro Gly Gly  Ala Ala Gly Arg Ala  His Gly Ser
    1595                 1600                 1605

Glu Pro  Trp Ala Ser Pro Asn  Tyr Thr Gly Gly Pro  Phe Phe Pro
    1610                 1615                 1620

Pro Ala  Gly Ser Ala Pro Ser  Gly Trp Pro Pro Val  Ala Gln Ala
    1625                 1630                 1635

Asn Ser  Arg Pro Glu Val Leu  Ser Ala Ile Gln Gly  Ala Gln Gly
    1640                 1645                 1650

Gln Gly  Pro His Val Ala His  Ser Leu Arg Leu Ala  Ala Ser Leu
    1655                 1660                 1665

Ser Pro  Ala Gln Thr Thr Ser  Glu Ser Phe Leu Ala  Pro Glu Ser
    1670                 1675                 1680
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Phe Ala  Ala Gly Val Arg Pro  Leu Leu Glu Gly Ser  Leu Ser Val
    1685                1690                1695

Leu Ile  Pro Glu Glu Pro Gln  Val Gly Leu Gly Pro  Ser Ala Gly
    1700                1705                1710

Gln Gln  Leu Ala Ser Ser Ser  Leu Ser Pro Gly Val  Ser Val Lys
    1715                1720                1725

Ala Glu  Pro Ser Ser Tyr Phe  Gln Ser Ala Gln Gly  Thr Cys Arg
    1730                1735                1740

Asp Val  Ser Ala Gly Ala Arg  Thr Ala Met Pro Ser  Ser Phe Leu
    1745                1750                1755

Glu Gln  Gly Arg Pro Gly Ala  Ala Pro Gly His Ala  Pro Ser Gly
    1760                1765                1770

Val Gly  Arg Cys Pro Pro Gln  Gly Arg Asp Ala Ser  Pro Gly Cys
    1775                1780                1785

Pro Gly  Phe Arg Thr Pro Pro  Ala Gly Phe Asp Gly  Pro Ser Ser
    1790                1795                1800

Ser Gly  Ala Gly Tyr Ser Leu  Ser Pro Tyr Gly Tyr  Pro Gly Thr
    1805                1810                1815

Glu Ile  Ser Pro His Leu Ala  Pro Phe Phe Pro Glu  Pro Tyr Arg
    1820                1825                1830

Arg Phe  Arg Glu Ser Arg Gly  Gly Pro Ala Trp Ile  His Ser Pro
    1835                1840                1845

Gly Ser  Val Asp Val Pro Ser  Ser Gly Leu Gln Ser  Pro Phe Thr
    1850                1855                1860

Gly Phe  His Ala Thr Ser Gly  Ser Ser Pro Pro Arg  Leu Gly Pro
    1865                1870                1875

Ser Glu  Gly Ala Ser Phe Ala  Glu Ala Ser Pro Arg  Ala Leu Ala
    1880                1885                1890

Gly Asp  Leu Gly Pro Ala Gly  Phe Leu Gly Ala Ser  Ala Gly Ala
    1895                1900                1905

Pro Ala  Ala Glu Gly Arg Gly  Pro Leu Phe Asp Pro  Ser Ala Glu
    1910                1915                1920

Gly Glu  Gly Lys Phe Ala Pro  Asp Ala Gly Ala Leu  Gly Thr Val
    1925                1930                1935

Glu Gly  Pro Ala Asp Cys Arg  Thr Gln Gly Glu Thr  Gly Arg Thr
    1940                1945                1950

Ala Asp  Glu Asp Glu Lys Lys  Lys Ala Lys Lys Ala  Lys Lys His
    1955                1960                1965

Gly Arg  Ile Thr Asp Ile Glu  Glu Arg Leu Ala Arg  Glu Glu Pro
    1970                1975                1980

Tyr Asp  Val Val Glu Glu Gly  Asp Asp Pro Glu Pro  Thr Arg Gln
    1985                1990                1995

Leu Gly  Leu Glu Ala Thr Glu  Lys Glu Gln Asp Val  Pro Arg Ser
    2000                2005                2010

Gly Asp  Ser Lys Ser Pro Asp  Gln Asp Ser Pro Gly  Gln Pro Ala
    2015                2020                2025

Asp Ile  Met His Gly Tyr Phe  Lys Ala Arg Val Arg  Asn Arg Arg
    2030                2035                2040

Val Lys  Asp Gly Leu Leu Leu  Arg Met Thr Ala Val  Leu Val Gly
    2045                2050                2055

Lys Gly  Phe Tyr Asp Leu Glu  Thr Val Glu Pro Gly  Ala Pro Arg
    2060                2065                2070

Arg Arg  Gly Gly Trp Gly Glu  Ser Gly Glu Glu Glu  Glu Glu Ser
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    2075                2080                2085

Glu Thr  Lys Tyr Leu Phe Ser  Asn Pro Ala Ser Gln  Lys Pro Cys
    2090                2095                2100

Asp Phe  Ile Leu Tyr Phe Asp  Thr Arg Glu Asn Arg  Asp Ala Ser
    2105                2110                2115

Val Ala  Ile Leu Asn Gln Ala  Leu Pro Ala Pro Pro  Pro Arg Leu
    2120                2125                2130

Pro Pro  Lys Asn Gly Glu Ser  Gln Ala Arg Arg Thr  Leu Arg Gln
    2135                2140                2145

Leu Tyr  Asp His Phe Leu Glu  Pro Lys Cys Gln Cys  Leu Glu Asp
    2150                2155                2160

Lys Thr  Leu Lys Val Lys His  Gly Val Ile Asn Leu  Leu Gly Phe
    2165                2170                2175

Pro Arg  Leu Tyr Val Lys Leu  His Cys Ser Met Ser  Trp Asp Glu
    2180                2185                2190

Arg Leu  Ser Leu Phe Ser Ser  Phe Leu His Trp Leu  Cys Arg Glu
    2195                2200                2205

Asp Asp  Ser Gln Pro Pro Pro  Trp Ser Ser Pro Glu  Leu His Pro
    2210                2215                2220

Glu Leu  Leu Ala Tyr Leu Val  Asp Leu Gly Arg Lys  Gly Phe Ala
    2225                2230                2235

Ser Gly  Gly Ala Ala Thr Thr  Ala Val Val Asn Ala  Pro Asp Leu
    2240                2245                2250

Pro Leu  Asp Asp Ser Ala Leu  Ser Lys Lys Asn Ala  Ala Leu Ile
    2255                2260                2265

Arg Ala  Tyr Met Gln Gln Asp  Thr Gly Ala Ser Gly  Pro Ser Gly
    2270                2275                2280

Ser Val  Gly Ala Thr Ser Ser  Asp Pro Glu Ala Pro  Arg Lys Asp
    2285                2290                2295

Asp Glu  Ala Glu Glu Gly Glu  Lys Asp Asp Ser Asn  Ala Ala Leu
    2300                2305                2310

Val Glu  Gly Pro Ala Pro Glu  Thr Ser Gly Asp Ser  Thr Gly Ala
    2315                2320                2325

Ala Gln  Pro Cys Gly Lys Gly  Arg Glu Glu Arg Glu  Ala Gly Asp
    2330                2335                2340

Lys Arg  Gly Pro Gly Asn Glu  Gly Cys Gly Lys Gly  Asp Gly Phe
    2345                2350                2355

Gly Ser  Pro Val Ala Val Ala  Gly Thr Thr Ala Ala  Pro Gly Glu
    2360                2365                2370

Thr Glu  Ser Val Ser Cys Pro  Ser Ser Thr Ser Gly  Gly Gly Ala
    2375                2380                2385

Ser Ser  Ala Leu Ser Ser Gly  Pro Ser Asp Ser Ala  Ala Ala Pro
    2390                2395                2400

Asp Gly  Cys Glu Ser Ser Pro  Val Ala Leu Glu Ser  Ala Ser Leu
    2405                2410                2415

Leu Ser  Phe Ser Pro Ser Ala  Ala Arg Ala Glu Val  Leu Thr Val
    2420                2425                2430

Pro Gly  Val Gly Leu Val Asn  Phe Ser Leu Pro Asp  Gly Val Lys
    2435                2440                2445

Phe Asp  Lys Ser Lys Leu Ala  Phe Arg Cys Tyr Trp  Arg Glu Gly
    2450                2455                2460

His Ala  Gly Val Val Thr Val  Gly Ala Gly Ala Ala  Val Ser Pro
    2465                2470                2475
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Ser Ser  Gly Ala Gly Thr Phe  Val Pro Ser Arg Pro  Thr Val Cys
    2480                 2485                 2490

Thr Ala  Gln Asn Lys Ser Arg  Thr Phe Ser Cys Arg  Lys Tyr Gly
    2495                 2500                 2505

Leu Tyr  Gln Ser Arg Val Leu  Ala Leu Gln Ala Arg  Leu Leu Ser
    2510                 2515                 2520

Glu Leu  Leu Trp Pro Gln Pro  Pro Ser Pro Ala Arg  Leu Arg Val
    2525                 2530                 2535

Ser Ala  Met Ala Ala Val Val  Tyr Gly Leu Ile Ala  Ala Pro Met
    2540                 2545                 2550

Pro Phe  Thr Asp Pro Trp Gln  Ala Val Cys Gly Val  Ser Val Ala
    2555                 2560                 2565

Glu Asp  Ala Leu Arg Gln Arg  Arg Glu Val Trp Lys  Asn Leu Leu
    2570                 2575                 2580

Asp Pro  Arg Gln Arg Arg Pro  Ala Pro Ala Pro Ile  Ser Gln Leu
    2585                 2590                 2595

Ser Leu  Pro Pro Val Ser Gly  Pro Pro His Ala Ser  Ser Ala Thr
    2600                 2605                 2610

Gln Glu  Leu Pro Asn Arg Pro  Gly Thr Pro Trp Pro  Gly Gln Glu
    2615                 2620                 2625

Thr Val  Cys Gly Ala Arg Gly  Pro Ala Pro Gly Leu  Ala Ser Ala
    2630                 2635                 2640

Trp Ala  Thr Tyr Gly Asn Pro  Gly Asp Arg Asp Ala  Ala Glu Pro
    2645                 2650                 2655

Gln Ser  Thr Tyr Val Gly Arg  Gly Pro Ala Gly Ala  Glu Gly Pro
    2660                 2665                 2670

Gly Gly  Gly Ile Ala Val His  Arg Glu Trp Ala Arg  Asn Ser Gly
    2675                 2680                 2685

Ser Glu  Ala Ala Gln Pro Cys  Gln Phe Gly Arg Ala  Val Glu Arg
    2690                 2695                 2700

Pro Val  Pro Gly Pro Gln Ser  Ser Leu Gly Pro Gly  Gly Asp Asn
    2705                 2710                 2715

Arg Gly  Asp His Met Ala Tyr  Asp Gln Ser Pro Ala  Gly Pro Ala
    2720                 2725                 2730

Ser Asn  Ala Pro Gly Pro Thr  Pro Pro Phe Val Gly  Pro Phe Ser
    2735                 2740                 2745

Pro Gly  Leu Val Leu Arg His  Gly Pro Pro Ala Phe  Ser Gln Asp
    2750                 2755                 2760

Pro Ser  Leu His Arg Pro Pro  Phe Ala Ala Gly Thr  Gly Pro Ala
    2765                 2770                 2775

Gly Gln  Arg Leu Ala Ser Asp  Ser Pro Tyr Pro Leu  Lys Asn Glu
    2780                 2785                 2790

Ala Ser  Pro Gln Leu Ala Met  Ala His Ala Pro Gly  Phe Glu Asn
    2795                 2800                 2805

Ser Asp  Gly Phe Gln Gly Glu  Gln Pro Leu Ala Lys  Gln Arg Lys
    2810                 2815                 2820

Ile Glu  Gly Ala Ser Asp Arg  Pro Val Pro Asp Glu  Gly Gln Val
    2825                 2830                 2835

Leu Gly  Thr Ile Ser His Gly  Lys Ser Pro Ala Ala  Arg Pro Val
    2840                 2845                 2850

Asp Gly  Asp Phe Ala Pro Asp  Gly Arg Ser Pro Leu  Phe Ser Gln
    2855                 2860                 2865
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Asp Ala  Ser Gly Val Gly Gly  Gly Arg Pro Ser Gly  Val Gly Gly
    2870                2875                2880

Gln Leu  Ala Ala Gly Gly Lys  Gly His Phe Ala Thr  Ala Pro Phe
    2885                2890                2895

Gly Ser  Gly Thr Leu Pro Thr  Thr Arg Gly Pro Ser  Gln Pro Gly
    2900                2905                2910

Gly Asp  Gly Leu Ser His Arg  Ser Gly Thr Glu Pro  Ala Ala Ala
    2915                2920                2925

Tyr Ser  Ser Pro Ala Gly Ala  Ala Tyr Pro Ser Ala  Ser Asn Ala
    2930                2935                2940

Ser Pro  Ile Tyr Gly Ala Ala  Pro Lys Arg Glu Gly  Asp Ser Pro
    2945                2950                2955

Phe Gly  Pro Ala Pro Pro Ser  Gly Tyr Cys Arg Pro  Gly Ser Pro
    2960                2965                2970

Ala Val  Asp Pro Lys Leu Pro  Gly Ser Val Pro Ser  Ser Gly Asn
    2975                2980                2985

Leu Asp  Ser Val Asn Tyr Gly  Ser Phe Phe Pro Gly  Gln Gln Ala
    2990                2995                3000

Pro Gln  Gly Asp Gly Arg Ile  Ala Pro Trp Gly Ser  Gly His Val
    3005                3010                3015

Gly Ala  Pro Arg Gly Glu Ala  Arg Gly Ser Glu Arg  Val Gly His
    3020                3025                3030

Ala Gly  Ala Ser Arg Gly Leu  Thr Gly His Glu Leu  Glu Glu Gly
    3035                3040                3045

Gln Gly  Gly Pro Gly Glu Glu  Gly Ala Gly Arg Glu  Arg Gln Arg
    3050                3055                3060

Lys Arg  Arg Lys Ser Ala Met  Ser Met Ser Ser Gln  Gly Glu Asn
    3065                3070                3075

Thr Pro  Leu Phe Ala Pro Thr  Ser Leu Pro Pro Val  Pro Phe Ala
    3080                3085                3090

Ser Gly  Asp Ser Leu Ala Asp  Gly Ser Gly Ser Asp  Phe Gly Gln
    3095                3100                3105

Gln Leu  Gly Pro Pro Phe Ser  His Gly Ser His Ala  Pro Pro Phe
    3110                3115                3120

Pro Glu  Ala Asn Ala Val Gly  Ser Gln His Phe Thr  Ala Asp Asn
    3125                3130                3135

Leu Glu  Thr Pro Gly Leu Pro  Ala Glu Leu Gly Gly  Gly Asp Gly
    3140                3145                3150

Arg Arg  Gln Ser Gly Ser Thr  His Glu Glu Val Ser  Gly Pro Arg
    3155                3160                3165

Ala Gly  Gly Glu Lys Gly Glu  Phe Ser Leu Glu Gly  Ala Pro Gln
    3170                3175                3180

Ala Ala  Ala Gln Gln Leu Ser  Ala Glu Thr Leu Thr  Phe Leu Leu
    3185                3190                3195

Gly Thr  Asn Val Val Trp Glu  Glu Asn Glu Lys Arg  Trp Arg Val
    3200                3205                3210

Gln Val  Arg Pro Pro Ser Pro  Arg Gly Cys Asp Gly  Glu Gly Ala
    3215                3220                3225

Asp Gly  Lys Leu Gly Gly Glu  Lys Lys Lys Arg Lys  Arg Asp Gly
    3230                3235                3240

Phe Ser  Ala Gly Gly Glu Arg  Arg Arg Ser Ser Thr  Gly Asn Glu
    3245                3250                3255

Pro Asp  Asp Gln His Lys Ala  Gly Thr Leu Glu Trp  Val Ser Met
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    3260                3265                3270

Ala Gln  Leu His Gln Ala Gln  Lys Leu Gln Asn Gln  Leu Val Gly
    3275                3280                3285

Lys Met  Glu Arg Gly Lys Gly  Glu Gly Gly Asp Glu  Glu Arg Leu
    3290                3295                3300

Gly Gly  Asp Gly Arg Gly Asn  Ile Phe Phe Asp Ala  Asn Gly Ser
    3305                3310                3315

Asp Glu  Asn Ala Lys Lys Ala  Ala Leu Leu Lys Ala  Arg Arg Trp
    3320                3325                3330

Leu Arg  Arg Arg Ile Val Gln  Gly Gln Ile Leu Val  Thr Gly Leu
    3335                3340                3345

Ser Arg  Asp Gly Leu Phe Ser  Ser Arg Pro Asp Glu  Pro Glu Arg
    3350                3355                3360

Ser Ser  Ser Val Ser Thr Gly  Ala Phe Thr Gly Ser  Ser Pro Asn
    3365                3370                3375

Asp Lys  Pro Thr Asp Leu Asn  Ala Ala Val Pro Pro  Leu Ser Pro
    3380                3385                3390

Phe Phe  Ser Pro Ile Pro Phe  Gly Ala Thr Thr Ala  Pro His Arg
    3395                3400                3405

Pro Ser  Pro Gly Phe Tyr Pro  Pro Ala Pro Ala His  Pro Thr Glu
    3410                3415                3420

Asp Gly  Cys Arg Pro Pro Met  Pro Ala Pro Val Pro  Met His Ala
    3425                3430                3435

Pro Gln  Gly Pro Val Asp Ser  Arg Thr Tyr Arg Gly  Ala Arg Pro
    3440                3445                3450

Val Tyr  Pro Gly Ser Asp Val  Thr Pro Gln Thr Cys  His Gly Val
    3455                3460                3465

Arg Pro  Glu Ser Met Gln Glu  Glu Gly Arg Ala Ala  Leu Leu Ala
    3470                3475                3480

Glu Gln  Gly Ser Ala Phe Phe  Val Ser Gly Asp Gly  Lys Gly Asp
    3485                3490                3495

Asn Arg  Gly Ala Thr Val Gly  Gln Ile Arg Gln Gly  Thr Val Arg
    3500                3505                3510

Val Met  Gln Ser Gln Thr Ala  Ser Gln Ser Leu Asp  Gln Gly Phe
    3515                3520                3525

Asp Leu  Pro His Pro Pro Ala  Pro Gly Pro Ala Tyr  Arg Gly Val
    3530                3535                3540

Pro Val  Gly His Gly Pro Ser  Gly Pro Tyr Tyr Leu  Asn Gly Gly
    3545                3550                3555

Cys Val  Ala Gln Arg Pro Tyr  Ala Thr Phe Ser Asn  Leu Ala Gly
    3560                3565                3570

Pro Val  Gln Gly Ser Phe Pro  Pro Leu Glu Phe Ser  Asn Gly Gly
    3575                3580                3585

Leu Pro  Thr Thr Ala Leu Gly  Arg Arg Gly Ser Asp  Ser Gly Pro
    3590                3595                3600

Gln Gly  Ala Gly Arg Asn Ala  Ser Gln Met Gln Pro  Gly Phe Ala
    3605                3610                3615

Ser Arg  Pro His Gly Pro Glu  Arg Leu Gly Arg Glu  Ser Ala Pro
    3620                3625                3630

Gln Ser  Gly Ala Pro Pro Gly  Phe Ser Pro His Ala  His Gly Arg
    3635                3640                3645

Gly Glu  Arg Asp Arg Pro Ser  Phe Ser Gly Ala Thr  Thr Met Pro
    3650                3655                3660
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Leu Ala  Ser Leu Thr Ala Phe  Ser His Pro Ala Ala  Gly Pro Met
    3665                 3670                 3675

Phe Val  Gly Thr Glu Gly Arg  Gly Gln Gln Gly Asp  Ile His Pro
    3680                 3685                 3690

Asn Leu  Cys Gly Val Ala Pro  Val Gly Gly Pro Arg  Gly Pro Ala
    3695                 3700                 3705

His Ala  Pro Met Pro Ala Tyr  Gly Pro Gly Gly Ala  Ala Gly Pro
    3710                 3715                 3720

Pro Arg  Asp Asp Arg Arg Ala  Glu Gly Gly Ala Pro  Gly Val Ser
    3725                 3730                 3735

His Ser  Asp Ile Phe Leu Ala  Asn Asp Arg Arg Leu  His Pro Glu
    3740                 3745                 3750

Met Cys  Leu His Ser Ala Pro  Ser Trp Gly Pro Ala  Gly Thr Phe
    3755                 3760                 3765

Ala Ser  Pro Asp Asn Arg Gln  Asn Ala Glu Pro Trp  Pro Ala Ala
    3770                 3775                 3780

His Ala  Ser Ser Asn Asn Phe  Phe Asp Tyr Thr Gly  Val Asn Met
    3785                 3790                 3795

Pro Ala  Ala Gly Pro Pro Ile  Gln Leu Asp Trp Ser  Lys Val Arg
    3800                 3805                 3810

Gly Ala  Gly Gly
    3815


<210> SEQ ID NO 16
<211> LENGTH: 432
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 16

Met Asp Arg Ala Gly Leu Leu Phe Leu Arg Gly Ala Ala Gly Pro Gly
1               5                  10                  15

Pro Leu Lys Cys Phe Gly Pro Arg Val Glu Ala Phe Ser Gly Ser Ile
            20                  25                  30

Ser Leu Leu Ser Leu Asp Ser Arg Gly Pro Thr Pro Phe Arg Thr Pro
        35                  40                  45

Phe His Thr Thr Ser Ala Leu Ser Lys Ser Arg Gln Pro Pro Lys Glu
    50                  55                  60

Ser Pro Glu Ser Ala Ala Ala Cys Thr Phe Ser Pro Leu Phe Pro Ser
65                  70                  75                  80

Pro Val Arg Ala Ser Pro His Arg Asn Leu Leu Gly Ala Arg Val Ser
                85                  90                  95

Val Pro Cys Lys Pro Leu Ala Cys Val Gly Ala Pro Lys Arg Arg His
            100                 105                 110

Gly Glu Thr Ser Asp Gly Phe Ser Ser Arg Ala Ala Val Ala Ala Glu
        115                 120                 125

Ala Leu Pro Pro Trp Pro Ser Asp Phe Leu Gln Ser Glu Glu Ile Ala
    130                 135                 140

Val Asp Ser Pro Gln Lys Pro Thr Gly Phe Ser Arg Pro Ser Asn Ala
145                 150                 155                 160

Arg Val Ser Pro Ala Pro Asn Ala Trp Glu Ala Ala Ala Val Phe Arg
                165                 170                 175

Arg Leu His Ala Phe Asp Ser Gly Leu Arg Gly Asp Ala Ser Gly Ala
            180                 185                 190

Phe Ala Ala Ser Ala Thr Cys Gly Cys Leu Ala Ala Ala Ser Arg Arg
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        195                 200                 205

Asn Pro Cys Leu Pro Ala Tyr Gln Leu Ser Trp Asn Leu Leu Gln Ala
    210                 215                 220

Arg Met Phe Gly Gly Arg Ala Gly Gly Leu Lys Arg Arg Lys Pro Arg
225                 230                 235                 240

Arg Asp Pro Gly Arg Val Ile Gln Ser Gly Met Gly Arg Arg Gln Glu
                245                 250                 255

Phe Phe Trp Pro Glu Lys Ala Arg Arg Thr Arg Val Pro Leu Tyr Gln
            260                 265                 270

Asn Ser Arg Pro Asn Leu Val Tyr Asp Gln Arg Phe Arg Arg Phe Met
        275                 280                 285

Cys Met Trp Tyr Ala Asn Gly Val Gln Val Phe Arg Pro Phe Ser Cys
    290                 295                 300

Arg Gly Arg Arg Gly Gly Arg Gly Lys Glu Gly Leu Pro Asp Gly Leu
305                 310                 315                 320

Gly Ile Gly Arg Gly Ser Gly Thr Trp Glu Arg Ala Arg Ala Lys Ala
                325                 330                 335

Val Val Leu Leu Lys Gln Leu Gln Arg Gln Gly His Leu Asp Arg Leu
            340                 345                 350

Ala Lys Pro Asp Val Thr Arg Ser Gly Val Arg Gly Val Tyr Phe Asp
        355                 360                 365

Thr Glu Glu Lys Leu Trp Val Ala Thr Trp Asn Glu His Gly Leu Arg
    370                 375                 380

Arg Phe Lys Ala Phe Pro Thr Met Glu Met Gly Phe Asp Ala Ala Tyr
385                 390                 395                 400

Gln Ala Ala Val Ala Val Arg Arg Gln Lys Leu Arg Glu Asn Tyr Ile
                405                 410                 415

Phe Ser Met Gln Arg Asn Arg Lys Lys Ser Gly Arg Pro Pro Phe Lys
            420                 425                 430


<210> SEQ ID NO 17
<211> LENGTH: 1919
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 17

Met Glu Leu Pro Asp Gln Ala Ala Tyr Gly Arg Gln Leu Ala Lys Arg
1               5                   10                  15

Arg Arg Leu Ser Ala Ser Glu Glu Glu Ala Thr Leu Ala Ser Lys Asn
            20                  25                  30

Asp Gly Lys Glu Gly Leu Gln Glu Pro Ala Gly Ala Thr Ala Gly His
        35                  40                  45

Leu Leu Pro Ala Glu Glu Pro Gly Gln Tyr Thr Pro Glu His Thr Glu
    50                  55                  60

Gly Arg Arg Glu Phe Arg Arg His Pro Val Val Leu Pro Gly Gly Gly
65                  70                  75                  80

Arg Lys Ala Ala Ser Tyr Gly Leu Ile Ala Val Gly Gly Gly Asp Ser
                85                  90                  95

Leu Arg Ala Ser Arg Arg Thr Arg Ser Ser Ala Ser Ile Glu Thr Ser
            100                 105                 110

Ala Glu Glu Lys Glu Thr Tyr Thr Ser Pro Glu Leu Gly Pro Gly Ala
        115                 120                 125

Ser Trp Ser Val Ser Thr Val Lys Gly Ser Lys Gly Arg Ser Asp Glu
    130                 135                 140
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Glu Gly Arg Ala Gly Lys Arg Ala Gly Ala Cys Ala Ala Arg Asp Ser
145                 150                 155                 160

Ala Ala Gly Ser Arg Thr Leu Ala Gln Ala His Arg Gly Ala Asp Glu
                165                 170                 175

Glu Arg Met Pro Pro His Ser Pro Ala Arg Asp Gly Ser His Gln Val
            180                 185                 190

Cys Trp Asp Ser Asp Ala Leu Pro Ser Pro Tyr Asp Leu Gly Leu Ser
        195                 200                 205

Glu Ala Asp Asp Glu Glu Val Ser Pro Gln Lys Glu Ala Leu Ala Glu
    210                 215                 220

Lys Asn Gly Leu Val Leu Phe Ala Glu Gln Ser Leu Gln Gly Val Gly
225                 230                 235                 240

Leu Ala Thr Leu Thr Val Pro Ser Gly Ala Thr Ser Ser Lys Gly Ala
                245                 250                 255

Phe Ser Ala Gly Ser Pro Phe Leu Pro Gly Ser Gly Thr Thr Ala Ser
            260                 265                 270

Pro Arg Ser Pro Val Pro Arg Gly Asp Lys Ser Leu Gly Asp Gly Ser
        275                 280                 285

Val Gly Ser Asp Asp Val Ser Ala Lys Ala Ser Pro His Ala His His
    290                 295                 300

Asn Val Thr Ala Gly Ala Asp Ala Ser Gln Ser Ser Glu Asp Ala Phe
305                 310                 315                 320

Phe Pro Ala Ala Pro Pro Gly Gly Val Pro Gly Thr Leu Thr Val Glu
                325                 330                 335

Asp Leu Leu Thr Met Pro Glu Arg Arg Gln Asp Pro Glu Ala Glu Lys
            340                 345                 350

Ala Ala Lys Thr Asp Phe Asp Cys Leu Ala Ala Leu Ile Gln Asp Ala
        355                 360                 365

Leu Gly Glu Ala Gly Gly Ala Ala Gly Arg Val Ala Pro Lys Arg Arg
    370                 375                 380

Cys Arg Thr Leu Gly Ala Gly Phe Asn Pro Thr His Ser Ala Arg Pro
385                 390                 395                 400

Gly Ser Gly Ser Val Ala Gly Leu Glu Ala Pro Gly Ala Leu Gly Arg
                405                 410                 415

Glu Leu Asp Ala Leu Val Ala Gly Gly Ser Pro Glu Glu Ser Arg Ala
            420                 425                 430

Asp Leu Glu Pro Asp Gly Gln Ala Ala Gly Ala Ser Trp Gln Glu Ser
        435                 440                 445

Leu His Leu Gln Tyr Asp Thr Arg Asn Gly Gly Thr Tyr Glu Glu Asp
    450                 455                 460

Leu Asp Ser Ala Ser Leu Ser Phe Leu Leu Gly His Ser Glu Gly Ser
465                 470                 475                 480

Glu Lys Gly Pro Ala Leu Ser Ala Ser Ala Gly Ala Ser Thr Ala Ala
                485                 490                 495

Ser Val Ser Ser Phe Phe Pro Ser Glu Ala Ala Cys Gly Val Tyr Ala
            500                 505                 510

Pro Gly Gln His Gly Arg Pro Thr Gln Ser Gln Asp Pro Ala Lys Glu
        515                 520                 525

Arg Gln Arg Arg Leu Ala Arg Asp Arg Glu Thr Leu Asn Leu Ser Ala
    530                 535                 540

Gln Ile Ala Ala Arg Phe Lys Ser Cys Arg Thr Glu Asp Val Met Arg
545                 550                 555                 560

Leu Phe Arg Arg Tyr Leu Ala Val Ser Ser Arg Gln Val Arg Asp Pro
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               565                 570                 575

Ala Thr Leu Glu Arg Val Val Ala Ala Cys Cys Tyr Ile Ser Ser Arg
            580                 585                 590

Gln Ala Met Asp Gly Leu Ser Leu Ser Asp Ile Cys His Glu Met Asp
        595                 600                 605

Ala Ser Asn Gly Gln Asp Leu Phe Ala Ala Gly Cys Glu Ala Arg Gly
    610                 615                 620

Lys Lys Leu Ala Asp Gly Glu Gly Met Gly Arg Gly Glu Ser Asp Arg
625                 630                 635                 640

Glu Arg Ala Gly Gly Ala His Ala Ser Met Arg Cys Lys Ser Leu Gly
                645                 650                 655

Lys Trp Val Val Arg Ile Cys Arg Lys Leu Gln Leu Gln Ala Leu Pro
            660                 665                 670

Asp Lys Asp Asp Asp Pro Glu Glu Arg Ala Asn Arg Val Leu Ala Arg
        675                 680                 685

Val Lys Gln Leu Leu Ile Ala Lys Met Glu Glu Glu Glu Gln Arg Arg
    690                 695                 700

Pro Gln Leu Val Asn Ala Phe Val Arg Ala Thr Gln Ser Ala Val Glu
705                 710                 715                 720

Lys Gln Arg Leu Lys Ala Glu Gly Asp Arg Ser Glu Ala Asp Ser Leu
                725                 730                 735

Ala Ser Leu Glu Ser Leu Leu Gly Asp Glu Ala Arg Arg Ala Asp Ala
            740                 745                 750

Arg Ala Asp Ala Glu Ala Arg Arg Gln Thr Pro Ala Glu Ala Gln Leu
        755                 760                 765

Gly Asp Phe Leu Asp Gly His Gln Gly Gly Glu Lys Thr Gly Arg Val
    770                 775                 780

Ser Ser Ala Arg Ile Asn Gly Arg Ala Ala Glu Ala Ser Pro Ala Pro
785                 790                 795                 800

Pro Ala Pro Gln Gly Ser Thr Ala Pro Ala Asp Ser Thr Pro Ala Ala
                805                 810                 815

Gly Ser Glu Glu Arg Gln Ala Leu Asn Ala Ile Glu Glu Leu Leu Ala
            820                 825                 830

Gln Val Thr Gly Gly Ser Asn Leu Asp Cys Phe Gly Ser Ala Thr Leu
        835                 840                 845

Ala Ala Val Asp Ser Asp Leu Ala Ser Gly Thr Ser His Val Asp Arg
    850                 855                 860

Glu Ser Cys Ala Arg Leu Arg Arg Leu Asp Lys Ser Gly Arg Asp Ala
865                 870                 875                 880

Phe Ala Ala Asp Ala Asp Gly Pro Glu Arg Pro Thr Glu Asn Glu Val
                885                 890                 895

Glu Pro Glu Ala Arg Pro Gly Ala Ala Gly Val Leu Ala Glu Asp Val
            900                 905                 910

Asp Glu Ala Ser Met Ala Leu Thr Pro Asn Pro Asp Asp Arg Ser Ser
        915                 920                 925

Ser Ala Ser Gly Asp Ala Ala Glu Pro Val Ala Ser Ile Arg Leu Glu
    930                 935                 940

Gln Arg Glu Glu Lys Asn Gly Asp Ala Ser Gly Leu Ser Leu Asp Ile
945                 950                 955                 960

Cys Pro Ser Leu Phe Asp Pro Val Asp Met Pro Ala Leu Ser Ala Ser
                965                 970                 975

Ser Glu Gly Asp Ser Gly Asp Ser Ser Pro Phe Ser Pro Ile Leu Thr
            980                 985                 990
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Ser Leu Leu Ser Ala Ser Leu Pro  Pro Ser Glu Thr Leu  Ala Gln Ala
        995                 1000                1005

Lys Asp  Met Gln Pro Ala Ala  Arg Leu Gln Leu Gln  Arg Phe Thr
    1010                1015                1020

Trp Leu  Gln Lys Met Arg Ala  Glu Ala Leu Glu Lys  Leu Lys Lys
    1025                1030                1035

Glu Lys  Glu Ala Val Phe Arg  Gly Leu Val Leu Leu  Gln Arg Ile
    1040                1045                1050

Leu Gln  Leu Phe Tyr Asp Val  Lys Gln Gly Glu Ser  Asp Gly Glu
    1055                1060                1065

Arg Glu  Asp Gly Glu Asp Gly  Glu Gly Lys Lys Arg  Gln Lys Gly
    1070                1075                1080

Trp Thr  Glu Glu Asp Pro Leu  Asp Lys Arg Trp Arg  Ala Arg Gly
    1085                1090                1095

Arg Cys  Asp Ala Val Ser Leu  Ala Ser Leu Ile Ile  Ile Val Phe
    1100                1105                1110

Lys Trp  Met Gln Ile Pro Ile  Pro Gln Arg Ile Ala  Leu Asp Ala
    1115                1120                1125

Leu His  Val Asp Arg Lys Ser  Val Tyr Lys Arg Arg  Leu Glu Gln
    1130                1135                1140

Met His  Ile Leu Lys Thr Leu  Phe Gly His Leu Arg  Gly Met Val
    1145                1150                1155

Glu Lys  Lys Asp Gly Ser Ser  Ser Ser Ala Leu Ala  Glu Glu Leu
    1160                1165                1170

Lys Ala  Ser Leu Pro Pro His  Leu Ala Ser Leu Leu  Gln Gln Val
    1175                1180                1185

Val Gly  Asn Pro Ala Thr Met  Gln Arg Leu Leu Ala  Leu Ala Asp
    1190                1195                1200

Glu Glu  Glu Glu Leu Gly Asn  Phe Ile Ser Ser Gln  Ser Leu Gly
    1205                1210                1215

Ser Asp  Gly Glu Ser Gly Lys  Ala Ser Ala Gly Leu  Gly Gly Val
    1220                1225                1230

Pro Pro  Pro Ala Ala Ala Ala  Ser Pro Thr Pro Val  Lys Thr Ser
    1235                1240                1245

Gln Ala  Ser Phe His Pro Gln  Ala Pro Ala Ala Ser  Ser Pro Glu
    1250                1255                1260

Ser Ser  Ala Pro Ser Val Ala  Val Glu Pro Glu Gln  Asp Ala Ala
    1265                1270                1275

Ser Ser  Ser Phe Leu Ala Ala  Ile Leu Ala Glu Val  Ala Ala Glu
    1280                1285                1290

Arg Glu  Val Gly Ala Val Lys  Thr Arg Gly Pro Gly  Asp Ala Glu
    1295                1300                1305

Arg Thr  Ala Ala Glu Leu Gly  Phe Gln Thr Arg Lys  Lys Arg Arg
    1310                1315                1320

Val Ser  Glu Leu Asn Ala Gln  Arg Ser Pro Asp Asn  Gly Leu Gly
    1325                1330                1335

Ser Asp  Leu Tyr Asp Glu Asp  Arg Glu Ala Ser Ser  Ala Val Pro
    1340                1345                1350

Val Ala  Ser Pro Leu Ala Asn  Leu Cys Ser Ser Leu  Ser Ser Ser
    1355                1360                1365

Ser His  Arg Asn Pro Ser Glu  Met Ala Ala Val Ala  Ser Val Ala
    1370                1375                1380
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Pro Ser  Pro Arg Ala Ala Arg  His Pro Arg Ala Pro  Asp Glu Met
    1385                1390                1395

Thr Leu  Gln Gly Leu Ala Val  Gly Lys Asp Ala Gly  Thr Pro Arg
    1400                1405                1410

Gln Ala  Gly Gly Tyr Ala Gly  Thr Phe Leu Pro Gly  Asp Gly Asp
    1415                1420                1425

Arg Val  Ser Glu Gly Glu Asp  Gly Arg Ser Glu Arg  Val Arg Ala
    1430                1435                1440

Arg Phe  Leu Ala Glu Arg Gly  Ser Met Asp Ala Ser  Ser Ser Phe
    1445                1450                1455

Ala Leu  Gly Phe Ser Leu Ala  Glu Ala Leu Leu Arg  His Gly Phe
    1460                1465                1470

Cys Leu  Pro Ser Pro Ser Asp  Pro Pro Ala Gly Leu  Ala Asp Ala
    1475                1480                1485

Gln Phe  Ala Thr Gly Asp Leu  Leu Arg Asp Gly Gly  Ser Ser Ser
    1490                1495                1500

Gly Glu  Arg Ala Leu Arg Met  Gln Pro Glu Gly Phe  Ser Ala Thr
    1505                1510                1515

Arg Gly  Ser Arg Pro Ala Val  Ala Pro Gly Pro Ala  Gly Phe Gly
    1520                1525                1530

Ile Gln  Ala Glu Ala Glu His  Glu Gly Arg Gly Asp  Val Asn Ser
    1535                1540                1545

Thr Asp  Val Ile Phe Ser Asn  Arg Ala Thr Arg Asp  Ile Ile Ala
    1550                1555                1560

Ser Phe  Leu Ala Ser Ala Ser  Thr Glu Gly His Pro  Gly Thr Ala
    1565                1570                1575

Ser Leu  Thr Gly Arg Gly Leu  Glu Asp Gly Arg Ser  Pro Arg Leu
    1580                1585                1590

Arg Gly  Pro Leu Ala Ala Val  Pro Lys Ala Val Ser  Gln Ala Asp
    1595                1600                1605

Arg Gly  Pro Gly Arg Phe Asn  Arg Gly Ala Ser Gly  Ser Cys Arg
    1610                1615                1620

Gln Pro  Ser Ser Arg Ser Pro  Pro Leu Pro Val Ser  Pro Tyr Arg
    1625                1630                1635

Gly Arg  Thr Gly Asp Ser Ser  Arg Gln Arg Pro Leu  Ser Pro Ser
    1640                1645                1650

Ser Leu  Phe Ala Ala Ala Ala  Ser Met Ala Gly Val  Leu Pro Gly
    1655                1660                1665

Pro Leu  Pro Ser Ser Arg Ser  Ala Gly Ser Ser Ala  Leu Ser Pro
    1670                1675                1680

Gly Val  Glu Arg Ser Pro Arg  Glu Arg Val Ala Ala  Gln Ala Leu
    1685                1690                1695

Glu Ala  Thr Arg Arg Gly Asp  Val Asp Asp Arg Ser  Leu His Pro
    1700                1705                1710

Ser Ser  Ser Val Ser Ala Val  Arg Ser Leu Leu Pro  Ala Glu Pro
    1715                1720                1725

Ala Leu  Gly Gly Ala Ser Pro  Phe Ala Ser Ser Ala  Leu Ala Met
    1730                1735                1740

Gly Leu  Pro Glu Ala Gly Ala  Ser Gln Ala Gly Ala  Asp Ala Pro
    1745                1750                1755

Leu Ala  Ser Pro Ser Ile Ala  Leu Ala Thr Val Ala  His Leu Lys
    1760                1765                1770

Ala Ala  Glu Lys Ala Leu Leu  Asp Ser Val Pro Asp  Ser Ala Arg
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    1775                1780                1785

Val Val  Ser Leu Gln Phe Glu  Arg Thr Gln Gln Arg  Trp Val Cys
    1790                1795                1800

Lys Trp  Gln Arg His Lys Pro  Ala Gly Ala Pro Ala  Asn Arg Lys
    1805                1810                1815

Glu Pro  Trp His Arg Arg Cys  Phe Ser Val Ile Lys  Tyr Gly Tyr
    1820                1825                1830

Glu Gly  Ala His Ala Leu Ala  Ala Ala Val Ala Lys  Lys Leu Arg
    1835                1840                1845

Asp Gly  Arg Arg Ala Leu Leu  Gln Lys Gln Arg Arg  Leu Glu Glu
    1850                1855                1860

Glu Gly  Leu Ala Glu Ala Glu  Glu Ala Pro Arg Asp  Glu Glu Glu
    1865                1870                1875

Val Gly  Asp Ala Glu Asp Glu  Glu Pro Leu Gly Ala  Ala Glu Glu
    1880                1885                1890

Ala Glu  Glu Thr Val Ser Pro  Arg Val Asp Ala Gly  Gly Asp Arg
    1895                1900                1905

Ser Ala  Ser Gly Ser Ala Glu  Ala Gly Lys Gln
    1910                1915


<210> SEQ ID NO 18
<211> LENGTH: 485
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 18

Met Cys Val Lys Lys Glu Gly Gly Asp Gly Lys Arg Gly Asn Glu Lys
1               5                   10                  15

Asn Gln Val Asn Asp Lys Gly Val Lys Arg Thr Gly Arg Asp Val Glu
            20                  25                  30

Ser Arg His Ala Pro Ser Val Pro His Leu Glu Lys Leu Val Asp Met
        35                  40                  45

Ala Met Val Tyr Ser Ser Cys Leu Pro Pro Cys Asp Ser Ser Gln Gly
    50                  55                  60

Gly Asp Gly Glu Arg Val Lys Arg Asn Ala Gly Ala Lys Arg Lys Gln
65                  70                  75                  80

Gly Gln Gly Glu Ser Gln Asp Arg Leu Lys Ala Leu His Asp Ser His
                85                  90                  95

Pro Leu Gln Cys Val Trp Tyr Leu Glu Ser Ser Pro Ala Thr Asn Ile
            100                 105                 110

Thr Leu Pro Ser Glu Ser Gly Ser Leu Lys Ser Pro Ser Ser Pro Ser
        115                 120                 125

Lys Arg Ala Ser Pro Asp Arg Val Met Glu Val Ser Ala Ser Leu Cys
    130                 135                 140

Lys Glu Glu Gln Lys Arg Arg Glu Gly Pro Arg Glu Gly Gln Trp Cys
145                 150                 155                 160

Cys Ser Trp Ser Phe Pro Arg Gly Arg Pro Thr Gly Thr Lys Phe Ser
                165                 170                 175

Val Lys Leu Phe Gly Tyr Glu Glu Ala Lys Arg Leu Ala Leu Tyr Thr
            180                 185                 190

Ala Leu Tyr Ala Tyr Ser Pro Glu Glu Arg Cys Asp Val Leu Gln Glu
        195                 200                 205

Leu Ile Asp Glu Val Leu Ala Thr Ala Ser Ser Ala Ser Leu Ser Ala
    210                 215                 220
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Ser His Leu Pro Asn Pro Glu Arg Phe Pro Ala Ile Leu Glu Leu Gln
225                 230                 235                 240

Pro Gln Pro Leu Ser Leu Ser Ser Ser Leu Ser Pro Ser Leu Cys Val
                245                 250                 255

Arg Leu Asp Ala Cys Ala Phe Pro Ser Pro Val Leu Ser Gly Ser Pro
            260                 265                 270

Leu Cys Ser Ser Pro Gly Leu Ser Ser Arg Gly Arg Asn Gly Ser Lys
        275                 280                 285

Ala Ala Arg Glu Thr Leu Ser Ile Asp Arg Gly Ile Arg Leu Ser Ser
    290                 295                 300

Gln Ser Ser Ala Ser Ser Asn Ala Met Pro Ser Gln Phe Pro Gln Arg
305                 310                 315                 320

Trp Gln Ala Thr Glu Val Arg Met Ser Leu Leu Cys Arg Ser Ser Phe
                325                 330                 335

Arg Ala Ser Arg Arg Arg Glu Gly Gly Asn His Gly Glu Ala Glu Ala
            340                 345                 350

Glu Ala Lys Arg Ala Gly Gln Thr Arg Glu Lys Thr Gly Arg Arg Asp
        355                 360                 365

Lys Gly Asn His Pro His Asp Leu Ser Val Asn Asn Arg Lys Glu Pro
    370                 375                 380

Asn Lys Leu Glu Lys Ser His Ser Ser Cys Ser Pro Arg Arg Ser Leu
385                 390                 395                 400

Phe Ser Ser Ile Gln Val Gln Gln Asp Glu Arg Ser Gly Gly Arg Leu
                405                 410                 415

Leu His Gly Phe Arg Gly Asp Met Glu Glu Gly Lys Arg Ala Ser Arg
            420                 425                 430

Ala Asn Lys His Val Glu Ala Lys Lys Gly Glu Val Thr Gly Arg Arg
        435                 440                 445

Lys Gly Val Cys Gly Gly Ala Leu Phe Gly Cys Phe Pro Ala Arg Arg
    450                 455                 460

Gly Arg Glu Arg Gly Glu Asp Glu Gly Glu Arg Glu Lys Ala Gly Gln
465                 470                 475                 480

Val Asn Ala Gln His
                485


<210> SEQ ID NO 19
<211> LENGTH: 3837
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 19

Met Ala Phe Ala Pro Arg Thr Ser Pro Arg Leu Ser Ala Gly Ala Gly
1               5                   10                  15

Gly Pro Ser Glu Ala Ser Arg Gly Gly Thr Ala Ala Gly Ala Pro Leu
            20                  25                  30

Gly Pro Val Glu Thr Pro Thr Gly Arg Glu Pro Ser Ser Pro Phe Leu
        35                  40                  45

Arg Ser Ala Ser Ala Lys Arg Val Thr Arg Ala Arg Ala Gly Phe Leu
    50                  55                  60

Ala Ser Ser Pro Asp Asn Gln Ser Arg Ser Thr Ser Pro Leu Arg Pro
65                  70                  75                  80

Ala Asp Ala Leu Arg Ile Ser Glu Ala Ser Ser Ala Ser Ala Pro Gly
                85                  90                  95

Val Arg Arg Ser Leu Arg Ala Ser Asn Ala Arg Pro Pro Val Ser Ala
            100                 105                 110
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Ser Arg Gly Leu Met Gly Lys Ala Gly Glu Glu Gly Glu Gly Ala Ser
        115                 120                 125

Gly Gly Gly Arg Pro Gly Gly Ala Arg Arg Thr Ser Gly Gly Gly Glu
    130                 135                 140

Asp Val Cys Ala Ser Pro Arg Asp Ile Ser Tyr Arg Asp Lys Gly Ala
145                 150                 155                 160

Gly Gly Asp Ala Thr Ala Ser Ser Asn Ser Gln Ser Pro Ser Ser Val
                165                 170                 175

Asp Ala Ala Val Ser Ser Ser Tyr Ser Val Ala Ser Ser Val Ser Ser
            180                 185                 190

Pro Pro Ala Ser Ser Leu Ser Ser Ala Leu Ser Ser Ser Phe Ser Ser
        195                 200                 205

Ser Thr Leu Ser Arg Ser Gly Ser Ser Val Cys Pro Arg Ala Thr Ser
    210                 215                 220

Ala Val Ser Ala Thr Leu Gln Gln Gly Glu Arg Ser Gln Glu Ser Ser
225                 230                 235                 240

Leu Leu Ala Gly Glu Arg Glu Thr Asp Arg Asp Ala Asn Arg Pro Gln
                245                 250                 255

Arg Glu Thr Asp Glu Gly Gln Gly Ala Lys Ser Glu Thr Asp Arg Ala
            260                 265                 270

Pro Glu Asp Asp Arg Gly Arg Ser Arg Arg Gly Ser Ala Ser Pro Val
        275                 280                 285

Gln Gly Pro Phe Ser Pro Arg Gly Phe Phe Ser Asn Ala Val Thr Lys
    290                 295                 300

Glu Asn Ser Ala Tyr Pro Ala Thr Ser Gly Gln Ser Gly Gln Glu Val
305                 310                 315                 320

Gly Cys Arg Pro Asn Ser Thr Leu Ser Ser Val Ser Val Cys Ser Leu
                325                 330                 335

Ser Ser Arg Pro Pro Ser Thr Leu Ala Ser Asp Gln Leu Leu Ser Val
            340                 345                 350

Pro Asn Gly Asp Ala Ser Thr Val Ser Thr Ser Ser Pro Ser Leu Ser
        355                 360                 365

Cys Ser Cys Ser Ser Phe Ser Ser Ser Ser Ser Ser Leu Ser Ser Ser
    370                 375                 380

Ser Leu Leu Ser Ser Ser Pro Leu Ser Ser Thr Pro Ser Ser Phe Phe
385                 390                 395                 400

Ser Ser Ser Ser Ser Ser Ser Ser Ser Ala Ser Val Ala Pro Pro Gly
                405                 410                 415

Glu Gly Lys Gly Arg Pro Pro Val Arg Ser Gly Arg Gly Ala Cys Pro
            420                 425                 430

Arg Lys Pro Ala Gly Pro Pro Pro Arg Leu Cys Val Pro Tyr Gln Cys
        435                 440                 445

Gln Phe Asn Val Glu Lys Arg Glu Trp Arg Ala Arg Tyr Leu Phe Arg
    450                 455                 460

Gly Gln Lys Lys Met Arg Val Phe Ser Leu Ala Arg Tyr Ser Pro Glu
465                 470                 475                 480

Val Ala Val Ser Leu Ala Glu Leu Phe Leu Thr Phe Leu Ala Asp Asn
                485                 490                 495

Asp Gly Ile Pro Arg Ser Glu Val Ile Ala Tyr Trp Ala Glu Thr Leu
            500                 505                 510

Ala Arg Gly Pro Val Thr Ala Thr Thr Gly Thr Asn Pro Lys Gly Gly
        515                 520                 525
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Asn Leu Leu Gly Pro Gly Ala Ser Glu Glu Glu Thr Val Gly Gly Glu
    530                 535                 540

Gly Gly Glu Asp Ala Glu Ser Arg Ala Ala Glu Lys Glu Arg Glu Glu
545                 550                 555                 560

Glu Gly Lys Ala Ser Ser Ser Ser Gly Ser Ser Asp Gln Asn Ile Thr
                565                 570                 575

Arg Val Glu Ser Ser Glu Ala Lys Glu Asp Gly Glu Glu Asn Ser Ala
            580                 585                 590

Ser Ser Lys Pro Pro Gly Ala Ala Ser Ala Ala Thr Glu Pro Ala Gly
        595                 600                 605

Gly Asp Ala Asp Gly Arg Pro Gly Arg Ala Ala Ser Gly Pro Gly Asp
    610                 615                 620

Ala Cys Arg Ser Val Thr Ser Thr Glu Thr Glu Ala Ala Val Ala Val
625                 630                 635                 640

Ala Pro Glu Ala Lys Gly Gly Pro Ser Ser Asp Val Ser Cys Thr Leu
                645                 650                 655

Asp Lys Ser Arg Glu Ser Arg Gly Asn Gly Val Ala Gly Lys Arg Glu
            660                 665                 670

Asn Pro Ala Trp Ala Val Ser Pro Ser Ser Phe Ala Ala Phe Val Glu
        675                 680                 685

Thr Ala Lys Ala Arg Gln Trp Val Thr Glu Ala Ser Arg Leu Gln Ala
    690                 695                 700

Ala Ser Leu Pro Pro Leu Ala Pro Ala Glu Arg Pro Ala Arg Pro Pro
705                 710                 715                 720

Ile Leu Pro Thr Leu Ala Ser Ser Arg Ala Arg Arg Cys Thr Ser His
                725                 730                 735

Ser Leu Ile Ser Gly Leu Ser Ala Arg Glu Gly Ser Gln Arg Thr Val
            740                 745                 750

Ser Gln Gly Asp Ser Leu Ser Pro Ala Ser Gly Leu Ala Gly Glu Pro
        755                 760                 765

Gly Ala Val Arg Glu Ala Glu Gly Arg Glu Ala Ile Ala Val Asp Asp
    770                 775                 780

Glu Thr Gly Gly Glu His Arg Asp Phe Pro His Ser Gln Gly Pro Ala
785                 790                 795                 800

Gly Arg Gly Arg Leu Ala Gly Ala Arg Pro Ser Ser Ser Asp Met Arg
                805                 810                 815

Gly Glu Lys Arg Gly Arg Arg Ala Leu Arg Glu Gly Glu Ser Lys Arg
            820                 825                 830

Pro Cys Arg Arg Arg Glu Asp Leu Lys Ser Glu Glu Gly Gln Arg Glu
        835                 840                 845

Arg Arg Arg Arg Asp Thr Ala Trp Pro Ala Gly Arg Arg Glu Ala Ser
    850                 855                 860

His Gly Arg Gln Asp Ser Arg Val Lys Glu Glu Thr Pro Ala Pro Asp
865                 870                 875                 880

Ala Gly Ala Ala Leu Ala Leu Asp Gly Arg Ala Ala Ala Ala Arg Asp
                885                 890                 895

Arg Pro Gln Lys Ala Pro Ser Pro Phe Gly Thr Pro Glu Ala Leu Ser
            900                 905                 910

Ser Ser Leu Thr Gly Ser Gly Leu His Pro Asp Gly Arg Asn Pro His
        915                 920                 925

Gly His Pro Ala Leu Arg Val Lys Leu Ala Ala Gly Arg Gly Asn Gly
    930                 935                 940

Leu Leu Ala Ala Ser Pro Ala Ser Pro Ser Ser Ala Ser His Ala Ser
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945 950 955 960

Ser Leu Ala Ser Pro Ser Ala Ser Trp His Ala Ala Gln Gly Glu Ala
965 970 975

Glu Ile Pro Gly Ala Ser Thr Gly Phe Val Asp Ser Pro Cys Ser Ala
980 985 990

Asn Gly Ser Leu Asp Asp Ser Gly Leu Gly Gly Pro Ala Ala Ala Leu
995 1000 1005

Gln Lys Ser Trp Arg Asp Arg Lys Arg Asn Arg Lys Lys Leu Ser
1010 1015 1020

Lys Ser Met His Arg Lys Ser Leu Ala Ser Leu Gly Met Arg Ala
1025 1030 1035

Pro Pro Gln Asn Ala Cys Leu Ala Asp Pro Ser Asp Val Gly Leu
1040 1045 1050

Gly Val Gln Met Pro Ser Asp Ala Gly Thr Val Pro Gly Ile Ser
1055 1060 1065

Pro Pro Ser Phe Gly Ala Ser Glu Gln Lys Ala Ser Ser Ser Ala
1070 1075 1080

Leu Gly Leu Ala Phe Arg Ala Ser Ser Ser Phe Ser Pro Lys Asn
1085 1090 1095

Gly Asp Val Glu Pro Ala Gly Arg Asn Pro Pro Gln Phe Leu Pro
1100 1105 1110

Thr Ala Ser Val Gln Arg Ala Asp Pro Pro Gly Thr Gly Ala Pro
1115 1120 1125

Pro Ser Gln Gln Val Val Ser Ser Ala Ser Pro Cys Ser Pro Ser
1130 1135 1140

Ala Leu Ala Ala Thr Ala Ser Pro Gly Ala Cys Arg Gly Gly Ala
1145 1150 1155

Ser Arg Asn Gly Asp Pro Gln Gly Glu Arg Phe Ser Phe Pro Ala
1160 1165 1170

Ser Pro Thr Ser Gln Tyr Arg Trp Tyr Ala His Pro Asp Gly Gly
1175 1180 1185

Ala Thr Gly Pro Ser Cys Cys Arg Gln His Val Gly Gly Ser Gly
1190 1195 1200

Gly Gly Gly Trp Pro Val Val Trp Leu Lys Gln Leu Glu Met Ala
1205 1210 1215

Val Asn Gly Pro Pro Lys Phe Cys Ser Tyr Val Glu Ala Val Asp
1220 1225 1230

Lys His Leu Arg Leu Gly Gly Leu Arg Arg Pro Val Ala Phe Leu
1235 1240 1245

Pro Leu Ala Ser Arg Pro Ala Ser Pro Thr Gly Leu Gly Gly Gly
1250 1255 1260

Leu Gly Ala Pro Gly Pro Ala Leu Arg Gln Ala Ser Glu Lys Ala
1265 1270 1275

Leu Ala Ala Glu Gly Arg Gln Gly Gln Asn Glu Glu Lys Gln Val
1280 1285 1290

Gly Trp Lys Ser Ala Thr Gly Ser Lys Ala Gly Met Phe Gln Gly
1295 1300 1305

Asp Ser Gly Glu Thr Thr Ser Glu Arg Gly Ala Glu Glu Ala Glu
1310 1315 1320

Gly Thr Gly Gly Gly Arg Arg Gly Ile Leu Gly Lys Glu Glu Glu
1325 1330 1335

Asp Arg Asn Gly Gly Glu Gly Glu Lys Ala Ala Thr Pro Thr Met
1340 1345 1350

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Gly Gly  Ala Ser Pro Ala Ala  Ser Asp Asp Ala Leu  Ser Pro Met
    1355                 1360                 1365

Lys Ala  Asp Arg Pro Ala Glu  Ala Leu Gly Thr Gly  Gly Ser Ala
    1370                 1375                 1380

Pro Thr  His Ala Asp Ser Arg  Arg Ala Pro Gly Met  Pro Glu Gly
    1385                 1390                 1395

Glu Lys  Met Thr Gly Pro Ser  Lys Glu Gln Glu Met  Ala Glu Ala
    1400                 1405                 1410

Gly Glu  Arg Asp Arg Cys Glu  Arg Ser Leu Glu Arg  Asn Ala Glu
    1415                 1420                 1425

Leu Val  Leu Lys Glu Asn Val  Ser Met Thr Ser Ala  Ser Asp Val
    1430                 1435                 1440

Ser Glu  Ala Ala Glu Glu Lys  Gly Ala Pro Lys Lys  Leu Ala Ser
    1445                 1450                 1455

Ser Pro  His Ser Val Glu Ser  Pro Cys Gly Arg Thr  Ala Glu Lys
    1460                 1465                 1470

Thr Gly  Thr Leu Asn Thr Ser  Glu Lys Gly Glu Asn  Thr Arg Thr
    1475                 1480                 1485

Ala Glu  Gly Asp Asp Pro Gly  Thr Thr Ile Val Lys  Glu Glu Phe
    1490                 1495                 1500

Pro Leu  Leu Pro Ala Pro Glu  Thr Pro Val Thr Val  Thr Ala Gln
    1505                 1510                 1515

Asp Leu  Leu Ser Pro Thr Val  Tyr Thr Pro Arg Trp  Gln Ala Thr
    1520                 1525                 1530

Val Gly  Lys Ser Leu Glu Leu  Gly Ser Leu Asn Cys  Glu Lys Ser
    1535                 1540                 1545

Cys Glu  Arg Gly His Trp Ala  Asp Ala Ser Ala Cys  Asp Leu Glu
    1550                 1555                 1560

Thr Lys  Asp Leu Arg Leu Pro  Glu Asp Asn Lys Ser  Glu Glu Leu
    1565                 1570                 1575

Lys Lys  Glu Thr Gly Met Phe  Leu Gly Val Glu Gly  Glu Gln Val
    1580                 1585                 1590

Glu Glu  Ala Lys Ser Ser Lys  Glu Ala Phe Ser Pro  Glu Glu Arg
    1595                 1600                 1605

Glu Arg  Glu Glu Gln Lys Glu  Ser Ser Lys Ala Ala  Gly Gly Gly
    1610                 1615                 1620

Asp Ser  Cys Arg Thr Pro Arg  Gln Gln Glu Ala Thr  Pro Arg Ala
    1625                 1630                 1635

Ser Glu  Glu Cys Gln Pro Glu  Ser Arg Ile Asp Met  Lys Val Ser
    1640                 1645                 1650

Pro Asn  Thr Glu Met Met Val  Glu Lys Leu Glu Glu  Thr Arg Val
    1655                 1660                 1665

Gln Asn  Thr Glu Glu Pro Glu  Lys Val Glu Glu Lys  Glu Glu Gly
    1670                 1675                 1680

Gly Ser  Val Cys Arg Asp Val  Ser Val Ala Ser Pro  Leu Glu Ser
    1685                 1690                 1695

Pro Asn  Ser Arg Leu Ser Glu  Lys Gly Asp Gln Ser  Glu Thr Pro
    1700                 1705                 1710

Ala Gly  Val Ala Pro Pro Ser  Ser Ser Ala Leu Glu  Ala Arg Ala
    1715                 1720                 1725

Gly Arg  Asp Ser Ala Leu Leu  Ser Ala Ser Leu Pro  Leu Ser Pro
    1730                 1735                 1740
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Arg Ala  Ser Cys Pro Pro Thr  Gln Ser Ala Ser Pro  Ala Ser Arg
    1745                1750                1755

Asp Pro  Thr Pro Ala Ser Leu  Arg Val Ser Ser Val  Ala Ser Gly
    1760                1765                1770

Asp Arg  Asn Gly Pro Thr Gly  Ile Leu Phe Arg Pro  Leu Ser Ser
    1775                1780                1785

Pro His  Lys Arg Val Ser Phe  Cys Leu Arg Gly Gly  Ala Glu Pro
    1790                1795                1800

Pro Gln  Arg Pro Leu Ser Glu  Ala Val Pro Tyr Pro  Leu Asn Ala
    1805                1810                1815

Arg Leu  Gln Glu Ile Val Ser  Arg Phe Arg Leu Leu  Gln Gly Val
    1820                1825                1830

Ser Ala  Ala Arg Val Ser Ser  His Gly Lys Gly Glu  Thr Ser Ser
    1835                1840                1845

Gln Ala  Thr Pro Lys Ala Val  Gln Gly Glu Ala Thr  Val Lys Glu
    1850                1855                1860

Lys Ala  Thr Val Thr Pro Thr  Glu Ser Ala Lys Ser  Leu Ala Gly
    1865                1870                1875

Gly Gln  Ala Glu Thr Glu Lys  Gly Glu Ser Pro Ser  Gly Ala Glu
    1880                1885                1890

Ala Ala  Thr Gln Lys Ala Asp  Glu Lys Glu Lys Thr  Pro Asp Thr
    1895                1900                1905

Asp Ala  Thr Gln Ser Arg Ser  Thr Ser Ser Gly Phe  Glu Thr Gln
    1910                1915                1920

Glu Ala  Lys Thr Ala Pro Ala  Ser Ile Leu Pro Ala  Ser Ser Leu
    1925                1930                1935

Pro Ser  Ser Asp Arg Pro Ser  Ala Ser Cys Ser Asp  Thr His Ala
    1940                1945                1950

Ser Arg  Asp Ala Val Pro Leu  Ala Ser Ser Pro Ser  Ser Ser Ser
    1955                1960                1965

Ser Pro  Ala Leu Arg Arg Cys  Ser Val Arg Gly Lys  Asp Leu Val
    1970                1975                1980

Ser Ala  Pro Val Asp Ser Phe  Ser Glu Gly Asp Ser  Ser Asp Ala
    1985                1990                1995

Arg Pro  Phe Val Ser Val Arg  Asp Leu Ala Val Lys  Leu Tyr Arg
    2000                2005                2010

Trp Leu  Glu Gln Gly Glu Gly  Leu Pro Ala Ala Ala  Gly Glu Pro
    2015                2020                2025

Gln Gly  Ala Cys Gly Val Gly  Ala Lys Ala Gln Ala  Arg Glu Ala
    2030                2035                2040

Leu Arg  Ile Asp Thr Val Pro  Phe Ile Ser Arg Trp  Arg Gln Met
    2045                2050                2055

Leu Glu  Arg Ser Leu Ser Ile  Ala Ser Asp Leu Arg  Lys Leu Asp
    2060                2065                2070

Leu Gln  Val Val His Leu Val  Glu Leu Thr Glu Ala  Leu His Ile
    2075                2080                2085

Ala Val  Tyr Ile Cys Gly Gln  Leu Arg Arg Arg Leu  Arg Glu Gly
    2090                2095                2100

Ala Ala  Pro Asp Ala Gly Ala  Ala Glu Asp Leu Ala  Pro Val Asp
    2105                2110                2115

Val Asp  Asp Pro Arg Gly Cys  Ser Gln Gln Ser Gly  Asp Thr Arg
    2120                2125                2130

Asp Ser  Ser Ser Pro Ala Thr  Pro Gly Gly Arg Leu  Ala Gly Gly
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    2135                2140                2145

Ala Gly  Gly Ala Ala Thr Ser  Pro Lys Gly Gln Ala  Phe Ala Pro
    2150                2155                2160

Arg Gly  Gly Glu Gly Glu Ile  Lys Pro Gln Glu Thr  Gly Asn Ser
    2165                2170                2175

Gly Asp  Ser Lys Ala Glu Gly  Lys Glu Ala Ser Gly  Asp Ala Asn
    2180                2185                2190

Thr Ser  Glu Gly Lys Arg Leu  Ser Gly Glu Val Asp  Lys Thr Ala
    2195                2200                2205

Glu Val  Glu Thr Ala Gly Ser  Glu Asp Ile Asn Val  Glu Arg Gly
    2210                2215                2220

Val Pro  Gly Ala Gln Ala Glu  Thr Ala Arg Thr Glu  Met Asn Gly
    2225                2230                2235

Gly Val  Val Lys Gly Gln Glu  Thr Ser Gly Asp Ile  Leu Ser Val
    2240                2245                2250

Gly Ser  Ser Gln Val Leu Ser  Leu Ser Ser Pro Ser  Leu Ser His
    2255                2260                2265

Leu Ala  Ser Ser Ser Gly Lys  Gly Pro Leu Lys Pro  Thr Ser Ser
    2270                2275                2280

Pro Ser  Ser Ser Leu Tyr Ala  Leu Ser Pro Ser Ser  Ser Ala Ala
    2285                2290                2295

Ser Pro  Phe Ser Ala Gln Leu  Ala Ser Pro Ser Ser  His Ala Pro
    2300                2305                2310

Leu Ser  Leu Ser Phe Arg Ser  Ser Ser Ser Pro Thr  Ser Leu Ser
    2315                2320                2325

Ser Pro  Leu Ala Ser Tyr Pro  Phe Pro Gln Thr Leu  Gln Gln Thr
    2330                2335                2340

Ser Ala  Ser Pro Ser Ser Ser  Ala Ser Ala Arg Pro  Ser Cys Ala
    2345                2350                2355

Ser Val  Lys Pro Leu Arg Glu  Ala Gly Asp Leu Val  Arg Ala Ala
    2360                2365                2370

Ala Arg  Ala Ala Leu Glu Gln  Ala Gln Val Phe Gly  Val Gly Gly
    2375                2380                2385

Lys Leu  Ser Asp Ala Thr His  Gln Leu Ala Ala Arg  Val Thr Val
    2390                2395                2400

Ala Val  Arg Ala Ala Met Leu  Ala Lys Gly Glu Gly  Gly Leu Thr
    2405                2410                2415

Arg Gly  Asp Val Asp Leu Leu  Val Glu Glu Thr Glu  Arg Phe Val
    2420                2425                2430

Arg Glu  Ala Arg Phe Lys Ala  Gln Glu Thr Ala Ala  Glu Thr Thr
    2435                2440                2445

Ala Leu  Pro Asp Gly Val Ala  Glu Val Val Ser Ser  Glu Ala Gly
    2450                2455                2460

Leu Gly  Leu Gln Thr Thr Asn  His Ala Pro Val Ser  Pro Ala Ala
    2465                2470                2475

Ala Pro  Ser Ala Gly Gly Ala  Phe Ala Gly Leu Thr  Glu Ala Val
    2480                2485                2490

Glu Val  Glu Ala Arg Gln Leu  Pro Glu Ala Ser Glu  Arg Val Gly
    2495                2500                2505

Arg Val  Ser Ser Pro Arg Gly  Ser Leu Gly Phe Glu  Ala Met Asp
    2510                2515                2520

Leu Ala  Gly Glu Leu His Leu  Val Lys Val Leu Asn  Ala Phe His
    2525                2530                2535
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Arg His  Thr Glu Cys Leu Met  Asn Glu Arg Glu Arg  Leu Ile Gln
    2540                 2545                 2550

Ala Thr  Asn Glu Asp Leu Ser  Phe Leu Leu His Ala  Met Glu Leu
    2555                 2560                 2565

Ala Leu  Pro Ser Gly Leu Asp  Thr Pro Leu Leu Ser  Ile Leu Glu
    2570                 2575                 2580

Gly Asp  Val Asp Ile Leu Pro  Pro Leu Pro Pro Pro  Asn Val Glu
    2585                 2590                 2595

Ala Leu  Ile Tyr Leu His Ala  Val Ser Leu Ala Gln  Ala Asp Ala
    2600                 2605                 2610

Ser Ala  Ser Pro Ser Ser Pro  Ser Ala Val Ala Pro  Cys Leu Leu
    2615                 2620                 2625

Ser Pro  Ser Ala Arg Leu Leu  Leu Ala His Phe Ala  Gly Ala Ser
    2630                 2635                 2640

Pro Thr  Ala Gly Gly Leu Gly  Gly Asp Ser Ala Lys  Gly Arg Thr
    2645                 2650                 2655

Met Ser  Ser Phe Pro Gly Arg  Pro Gly Glu Glu Arg  His Arg Ala
    2660                 2665                 2670

Asp Glu  Arg Lys Gly Ser Val  Leu Pro Val Arg Arg  Gly Arg Pro
    2675                 2680                 2685

Pro Ser  Ser Ala Arg Leu Asn  Ala Leu Arg Arg Leu  His Ala Val
    2690                 2695                 2700

Gly Glu  Pro Ala Ala Asp Ala  Gly Leu Asp Thr Val  Asn Gly Arg
    2705                 2710                 2715

Phe Arg  Ser Lys Arg Leu Arg  Ala Met Ser Gln Glu  Glu Glu Ala
    2720                 2725                 2730

Arg Arg  Ala Ala Thr His Ala  Ser Pro Thr Ile Pro  Tyr Pro Leu
    2735                 2740                 2745

Ser Arg  Tyr Leu His Arg Pro  Pro Arg Leu Leu Ser  Pro Thr Asp
    2750                 2755                 2760

Ala Gly  His Phe Ala Ser Ser  Tyr Ser Ser Pro Leu  Ser His Pro
    2765                 2770                 2775

Leu Ser  Lys Gly Ser Ser Leu  Thr Ser Pro Lys Arg  Gln Arg Arg
    2780                 2785                 2790

Ser Val  Cys Ser Glu Ala Pro  Glu His Glu Arg Lys  Asn Leu Arg
    2795                 2800                 2805

Ser Leu  Phe Lys Ser Pro Ser  Ala Gln Arg Glu Glu  Ala Pro Arg
    2810                 2815                 2820

Ser Leu  Thr Arg Pro Phe Gly  Pro Leu Lys Gly Glu  Gly Phe Ser
    2825                 2830                 2835

Pro Ala  Ser Leu Gly Thr Leu  Gly Ser Arg Arg Gln  Ser Glu Leu
    2840                 2845                 2850

Gly Ile  Arg Arg Arg Asp Ala  Leu Val Ala Phe Pro  Pro Ala Gly
    2855                 2860                 2865

Met Pro  Cys His Pro Ala Ser  Pro Gly Arg Arg Leu  Glu Arg Pro
    2870                 2875                 2880

Arg Val  Asp Gly Ala Asp Met  Asp Gly Glu Arg Arg  Arg Arg Thr
    2885                 2890                 2895

Arg Cys  Ala Gly Asp Arg Leu  Glu Glu Arg Arg Arg  Pro Leu Gly
    2900                 2905                 2910

Pro Val  Tyr Ile Pro Thr Lys  Val Arg Asp Pro Ala  Thr Gly Arg
    2915                 2920                 2925
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Val Ala  Val Cys Ala Cys Asp  Thr Glu Arg Gly Glu  Arg Val Arg
    2930                 2935                 2940

Lys Val  Gln Leu Phe Glu Lys  Pro His Val Gly Ala  Phe Trp Cys
    2945                 2950                 2955

Ala Arg  Tyr Gly Pro Asn Asp  Glu Phe Val Arg Cys  Phe Ser Ile
    2960                 2965                 2970

Glu Lys  Val Gly Ser Leu Lys  Ala Leu Val Ser Ala  Val Arg Phe
    2975                 2980                 2985

Arg Gln  Tyr Val Thr Gly His  Ser Leu Gly Tyr Gly  Val Gly Asn
    2990                 2995                 3000

Cys Val  Pro Val Glu Thr Ile  Arg Ser Ala Gly Arg  Arg Asp Arg
    3005                 3010                 3015

Asn Gly  Asp Val Ala Pro Asp  Arg Pro Leu Lys Gln  Ala Ala Ala
    3020                 3025                 3030

Ser Pro  Pro Pro Ala Gly Val  Ala Gly Ala Leu Gly  Arg Gly Glu
    3035                 3040                 3045

Val Gly  Gln Ala Gln Asp Glu  Ser Gly Glu Thr Arg  Asp Ala Val
    3050                 3055                 3060

Glu Glu  Glu Gly Arg Gly Gln  Glu Pro Leu Gly Ser  Gly Glu Gly
    3065                 3070                 3075

Ala Ser  Gly Val Ala Ala Lys  Glu Gly His Gly Ser  Ser Arg Gly
    3080                 3085                 3090

Glu Gly  Glu Gly Ala Glu Gly  Arg Thr Asp Ser Ala  Ala Gly Ser
    3095                 3100                 3105

Thr Ala  Gly Asp Arg Ser Thr  Glu Asp Ser Ser Arg  Leu Leu Ser
    3110                 3115                 3120

Glu Gly  Arg Asp Ala Lys His  Gly Ser Ser Pro Ala  Gly Gly Ser
    3125                 3130                 3135

Glu Ala  Leu Ala Pro Gly Gly  Glu His Ala Leu Ala  Glu Gly Ser
    3140                 3145                 3150

Glu Lys  Val Gly Arg Ala Gln  Glu Thr Glu Ala Arg  Lys Glu Asp
    3155                 3160                 3165

Leu Arg  Thr Ser Gln Asn Glu  Thr His Ser Gly Glu  Asp Val Ser
    3170                 3175                 3180

Ser Leu  Asn Glu Lys Ala Leu  Asp Ser Pro Arg Ser  Ser Ala Pro
    3185                 3190                 3195

Gln Gly  Lys Ser Asp Gln Gly  Arg Glu Pro Ile Ala  Leu Arg Ile
    3200                 3205                 3210

Arg Ser  Thr Leu Pro Pro Ser  Glu Val Asp Lys Gln  Glu Ala Ala
    3215                 3220                 3225

Gly Gln  Gly Gly Ser Ala Ser  Glu Leu Ala Phe Pro  Thr Gly Val
    3230                 3235                 3240

Ser Leu  Ala Ser Pro Val Ser  Pro Phe Ser Ala Leu  Ala Arg Ser
    3245                 3250                 3255

Pro Ile  Ser Ala Arg Ala Ser  Ser Val Ser Pro Gly  Ala Cys Asp
    3260                 3265                 3270

Arg Pro  Asp Val Ser Arg Arg  His Ser Gly Ser Ser  Asp Glu Ala
    3275                 3280                 3285

Ser Glu  Ala Leu Trp Asp Leu  Gly Glu Asp Leu Gly  Phe Ala Gly
    3290                 3295                 3300

Asp Asp  Ala Asn Phe Pro Phe  Leu Asp Ser Glu Asn  Ser Ala Leu
    3305                 3310                 3315

Leu Phe  Ala Pro Pro Arg His  Leu Met Ser Pro Gly  Ser Ala Ser
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    3320                3325                3330

Pro Thr  Gly Gly Gly Leu Gly  Ile His Tyr Asp Lys  Thr Lys His
    3335                3340                3345

Arg Trp  Lys Ala Thr Trp Thr  Thr Leu Asp Gly Gln  Arg Ala Ser
    3350                3355                3360

Thr Ser  Phe Ser Val Lys Val  Leu Gly Met Glu Arg  Ala Arg Glu
    3365                3370                3375

Leu Ala  Leu Glu Ala Arg Gln  Arg Ala Leu Ala Gly  Leu Asp Pro
    3380                3385                3390

Arg Glu  Val Arg Asp Glu Met  Val Ala Gly Gly Ala  Ala Ala Arg
    3395                3400                3405

Asp Arg  Glu Arg Glu Arg Gly  Arg Gln Asp Gly Arg  Arg Glu Gly
    3410                3415                3420

Ser Glu  Arg Arg Val Gly Phe  Glu Ala Glu Ala Glu  Gly Thr Glu
    3425                3430                3435

Ala Ala  Ser Glu Arg Leu Arg  Arg Arg Gly Glu Arg  Glu Asp Gly
    3440                3445                3450

Asp Glu  Glu Arg Arg Arg Lys  Lys Thr Arg Gly Asp  Glu Leu Arg
    3455                3460                3465

Gly Ala  Glu Gly Asp Arg Glu  Glu Arg Glu Leu Arg  Arg Arg Lys
    3470                3475                3480

Thr Ser  Glu Glu Arg Arg Lys  Gly Lys Asn Glu Ala  Ala Lys Asn
    3485                3490                3495

Glu Ala  Ala Lys Asn Glu Ala  Ala Lys Asn Glu Gly  Gly Lys Gly
    3500                3505                3510

Glu Thr  Trp Lys Val Arg Glu  Gly Gly Lys Thr Pro  Leu Gly Val
    3515                3520                3525

Lys Ser  His Arg Ala Lys Val  Val Gly Gln Thr Val  Glu Arg Arg
    3530                3535                3540

Gly Glu  Glu Arg Arg Arg Asp  Leu Arg Gly Ser Arg  Arg Glu Glu
    3545                3550                3555

Gly Lys  Thr Val Trp Gly Gln  Glu Gln Asp Ala Glu  His Gln Val
    3560                3565                3570

Phe Glu  Gly Val Lys Glu Asp  Asp Asn Glu Arg Gly  Arg Arg Arg
    3575                3580                3585

Glu Arg  Arg Arg Phe Glu Glu  Arg Asp Ser Leu Arg  Gly Ser His
    3590                3595                3600

Gly Ala  Thr Pro Ser Asp Glu  Gln Arg Gln Met Arg  Arg Gln Thr
    3605                3610                3615

Ile Leu  Gly Ser Arg Glu Val  Asp Gly Lys Pro Leu  Ser Phe Asp
    3620                3625                3630

Asp Thr  His Arg Val Asp Ala  Gln Leu Gly Ile Gln  Asn Glu Val
    3635                3640                3645

Ala Phe  Pro Gly Pro Gln Gly  Val Gly Gly Ala Gly  Asn Ser Leu
    3650                3655                3660

Gln Phe  Gly Arg Glu Gly Glu  Arg Phe Ala Ser Ser  Ser Pro Val
    3665                3670                3675

Ala Phe  Leu Arg Thr Lys Glu  Glu Asp Glu Glu Ile  Val Glu Val
    3680                3685                3690

Phe Leu  Thr Pro Glu Gly Ser  Gly Ser Glu Arg Asp  Lys Ala Ser
    3695                3700                3705

Ser Val  Ser Ala Ser Ser Ala  Pro Arg Asp Ser Arg  Pro Ala Ser
    3710                3715                3720
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Pro Arg  Leu Arg Ala Ser Arg  Leu Arg Glu Ser Ala  Arg Leu Gln
    3725                3730                3735

Arg Arg  Leu Glu Glu Ala Glu  Val His Asp Arg Gly  Ser Arg Pro
    3740                3745                3750

Leu Arg  Pro Glu Glu Arg Arg  Val Ala Lys Arg His  Val Ala Glu
    3755                3760                3765

Glu Asn  Val Asp Ala Thr Phe  Ser Ala Gly Ala Gly  Gly Thr Lys
    3770                3775                3780

Lys Ile  Arg Pro His Ser Ser  His Asp Phe Ser Ala  Glu Gly Leu
    3785                3790                3795

Ser Lys  Phe Gln Glu Leu Leu  Thr Trp Asp Cys Glu  Val Glu Ile
    3800                3805                3810

Asp Gly  Thr Asp Ala His Val  Trp Arg Ala Val Ala  Ala Leu Pro
    3815                3820                3825

Gly Pro  Arg Pro Arg Pro Arg  Tyr Val
    3830                3835


<210> SEQ ID NO 20
<211> LENGTH: 1292
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 20

Met Cys Gln Glu Arg Lys Pro Arg Glu Leu Ser Leu Arg Asn Asn Ser
1               5                  10                  15

Arg Ala Arg Glu Arg Arg Gly Ser Lys Leu Glu Pro Gly Val Ser Cys
            20                  25                  30

Leu Ser Leu Ser Ala Cys Pro Ser Val Ala Pro Asn Asp Arg Gly Gly
        35                  40                  45

Val Thr Thr Pro Arg Ser Leu His Ala Trp Thr Arg Glu Val Ser Ala
    50                  55                  60

Cys Arg Leu Pro Arg Gln Gln Val Ser Arg Pro Leu Pro Arg Arg Ser
65                  70                  75                  80

Leu Ser Arg Pro Arg Ser Glu Pro Asp Ala Ser Pro Val Lys Gly Pro
                85                  90                  95

Gly Gln Arg Val Glu Ala Ser Ala Val Glu Gly Gly Pro Ser Ala Ser
            100                 105                 110

Ser Ala Glu Arg Leu Gln Val Asp Asp Gly Leu Ala Ala Met Arg Lys
        115                 120                 125

Thr Lys Lys Gly Lys Gly Glu Glu Gly Gly Glu Glu Thr Glu Arg Trp
    130                 135                 140

Ala Thr Gln Ala Val Glu Gln Gln Gly Thr Leu Lys Pro Ala Gly Glu
145                 150                 155                 160

Glu Thr Ala Val Pro Gly Ala Ser Glu Arg Ser Ala Ser Pro Gln Gln
                165                 170                 175

Ala Met Glu Gly Ser Cys Gly Val Glu Thr Pro Glu Thr Phe Phe Gly
            180                 185                 190

Val Ser Thr Gly Asn Ser Gln Gly Ser Pro Ser Pro Glu Ser Val Ala
        195                 200                 205

Gly Glu Glu Ala Arg Pro Glu Arg Glu Asn Ala Glu Lys Ser Ala Thr
    210                 215                 220

Gly Gly Ser Ala Ser Lys Ala Lys Lys Pro Ser Arg Glu Ser Ala Arg
225                 230                 235                 240

Arg Pro Asp Thr Ala Leu Ile Asp Arg His Leu Ile Ala Ala Ser Pro
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               245                 250                 255

Ser Pro Ser Ser Ala Arg Arg Ser Ser Thr Cys Ser Pro Ser Pro His
            260                 265                 270

Ser Arg Glu Gly Glu Asp Lys Pro Gly Ser Gly Ala Pro Pro Ala Ser
        275                 280                 285

Ser Pro Ser Ala Asn Ala Gly Ala Leu Glu Pro Ala Glu Lys Gly Thr
    290                 295                 300

Leu Gly Ser Pro Pro Gln Asp Val Leu Pro Ala Leu Pro Ala Ser Ser
305                 310                 315                 320

Ser Ser Pro Ser Thr Gly Gly Gly Ser Pro Leu Ser Pro Pro Pro Gly
                325                 330                 335

Gln Ala Pro Arg Ala Glu Ser Gly Ala Pro Gly Ser Gly Ala Leu Ser
            340                 345                 350

Leu Arg Arg Ser Leu Arg His Arg Gln Pro Val Arg Pro Ala Ala Ile
        355                 360                 365

Ala Val Ser Pro Leu Gly Gly Pro Gly Ser Ser Leu Ser Ser Arg Ser
    370                 375                 380

Ala Ser Pro Thr Arg Arg Gly Gly Val Ser Pro Cys Gly Pro Ala Thr
385                 390                 395                 400

Ala Val Gly Lys Gly Ala Gly Ala Ala Ser Gly Ala Ala Ala Leu Pro
                405                 410                 415

Gly Val Gly Ala Lys Ala Pro Pro Ser Ala Thr Pro Leu Ala Gly Leu
            420                 425                 430

Ser Gly Arg Ser Leu Leu Ala Ser Val Ser Pro Ser Ala Ala Ala Leu
        435                 440                 445

Gly Pro Gly Ala Pro Gly Lys Lys Lys Ala Gly Gln Val Gln Gly Ala
    450                 455                 460

Ala Lys Ala Arg Gly Ala Pro Pro Phe Val Leu Ala Glu Tyr Trp Pro
465                 470                 475                 480

Gly Val Thr Leu Asp Glu Met Glu Lys Gly Glu Leu Ser Trp Ala Arg
                485                 490                 495

Ala Ala Ala Gly Leu Pro Leu Pro Ala Ser Pro His Lys Val Pro Gly
            500                 505                 510

Gly Pro Ala Pro Pro Val Gly Gly Pro Pro Ala Arg Asp Glu Asp Ser
        515                 520                 525

Val Ala Ala Cys Ala Gly Glu Lys Gly Lys Glu Lys Ala Phe Leu Gly
    530                 535                 540

Ser Gly Arg Ser Gln Ala Ala Gln Gly Leu Pro Gly Ile Asp Ala Val
545                 550                 555                 560

Ala Ala Ala Cys Trp Gly Gly Ala Gly Val Asp Ser Arg Val Leu Ala
                565                 570                 575

Pro Ala Glu Gly Glu Ala Ser Gly Ala Phe Gly Pro Gly Gly Glu Lys
            580                 585                 590

Lys Lys Val His Ala Ser Ser Asp Ser Ser Gly Gly Ser Arg Ala Ala
        595                 600                 605

Leu Gly Gly Arg Ala Ser Val Gln Gly Lys Ala Arg Lys Pro Ala Gly
    610                 615                 620

Trp Glu Glu Glu Arg Gly Arg Arg Asp Asp Arg Ser Arg Gly Arg Arg
625                 630                 635                 640

Asp Glu Thr Asp Gly Pro Arg Phe Asp Val Thr Trp Phe Val Asp Asp
                645                 650                 655

Ser Pro Leu Ala His Thr Arg Lys Arg Thr Arg Trp Asp Ser Leu Trp
            660                 665                 670
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Val Arg Pro Ala Ser Pro Val Arg Val Val Gly Asp Ser Ala Pro Glu
        675                 680                 685

Glu Ser Pro Glu Arg Arg Glu Gly Gly Gly Arg Ala Pro Asp Leu Gln
    690                 695                 700

Ala Ser Met Ala Lys Arg Arg Thr Ala Asp Ser Gly Leu Glu Glu Glu
705                 710                 715                 720

Ala Gln Val Glu Arg Gly Phe Ser Ser Ser Asp Ser Asp Asp Cys Asp
                725                 730                 735

Trp His Leu Pro Thr Arg Thr Val Ser Ser Ser Leu Ala Pro Phe Ala
            740                 745                 750

Ala Ser Lys Ala His Leu Val Pro Arg Cys Cys Tyr Cys Leu Leu Pro
        755                 760                 765

Arg Arg Leu Pro Gly Arg His Thr Glu Ala Gly Gly Pro Pro Arg Asp
    770                 775                 780

Leu Leu Gly Trp Ser Thr Ser Val Glu Ser Glu Glu Thr Arg Gly Arg
785                 790                 795                 800

Tyr Leu Gln Leu Tyr Cys Ala Cys Thr Lys Arg Pro Phe Gly Glu Ser
                805                 810                 815

Val Leu Gln Gly Ala Ala Gly Arg Arg Gly Leu Leu Leu Pro Val Ala
            820                 825                 830

Thr Asn Ala Leu Leu Tyr Ser Val Arg Arg Val Ala Leu Asp Gly Ala
        835                 840                 845

Ala Ser Glu Gln Lys Ser Glu Ala Leu Pro Thr Ser Ala Val Ser Arg
    850                 855                 860

Pro Ser Ser Ala Val Arg Ala Arg Ser Ser Cys Ala Ser Ser Gly Cys
865                 870                 875                 880

Asp Asp Gly Arg Ala Glu Val Ala Pro Gly Ala Pro Ala Glu Thr Ile
                885                 890                 895

Tyr Arg Trp Arg Asp Pro Cys Thr Leu Gln Thr Phe Ser Ser Ser Leu
            900                 905                 910

Asp Arg Ile Gln Gly Ser Leu Ala Ala Thr Ala Ala Val Ala Ala Ala
        915                 920                 925

Ala Glu Ser Ala Gly Lys Pro Val Ala Phe Leu Pro Arg Leu Tyr Trp
    930                 935                 940

Asp Ser Gln Ala Asp Cys Tyr Ile Ala Ser Cys Leu Arg Trp Glu Glu
945                 950                 955                 960

Glu Ala Gln Pro Thr Pro Ala Ala Glu Arg Gly Glu Lys Arg Asn Gly
                965                 970                 975

Val Glu Arg Pro Ala Glu Ala Arg Glu Arg Gly Arg Asp Glu Lys Lys
            980                 985                 990

Pro Glu Asp Pro Ser Val Pro Gly  Leu Arg Arg Arg Ser  Leu Lys Leu
        995                 1000                1005

Leu Gln  Lys Lys Phe Ser Val  Ala Phe Leu Gly Asp  Ala Lys Ala
    1010                1015                1020

His Phe  Tyr Ala Ser Glu Trp  Leu Lys Trp Gln His  Lys Gly Gln
    1025                1030                1035

Arg Met  Met Asp Glu Glu Asp  Arg Arg Gln Glu Val  Ala Arg Gln
    1040                1045                1050

Met Leu  Leu Val Ser Pro Leu  Leu Ala Gly Arg Lys  Ala Pro Ala
    1055                1060                1065

Lys Ala  Pro Gly Gly Cys Ser  Lys Lys Ala Ser Ser  Leu Ser Ala
    1070                1075                1080
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Ala Gln  Leu Ala Leu Ala Ser  Gly Arg Pro Leu Thr  Pro Glu Glu
    1085                1090                1095

Glu Ala  Glu Leu Lys Arg Gln  Leu Glu Asn Lys Glu  Arg Gln Lys
    1100                1105                1110

Lys Gln  Lys Leu Leu Arg Gln  Gln Trp Arg Arg Gln  Gln Ala Arg
    1115                1120                1125

Glu Ala  Lys Leu Arg Leu Arg  Glu Ala Glu Ala Ala  Ala Ala Ala
    1130                1135                1140

Ala Ala  Ala Ala Gly Ala Pro  Ser Ala Pro Gly Thr  Thr Gly Ala
    1145                1150                1155

Ser Gln  Thr Arg Ser Pro Gln  Ser Gln Gln Lys Ser  Glu Ser Leu
    1160                1165                1170

Pro Val  Leu Arg Ser Lys Thr  Glu Val Leu Gln Pro  Ser Pro Gly
    1175                1180                1185

Ala Ser  Phe Ala Pro Ala Ser  Ser Arg Ser Thr Leu  Pro Ala Gly
    1190                1195                1200

Glu Ser  Gly Ala Ala Pro Cys  Glu Gly Val Gly Thr  Arg Arg Ser
    1205                1210                1215

Ala Ala  Ser Ala Thr Ser Val  Ala Pro Glu Lys Val  Thr Gly Arg
    1220                1225                1230

Lys Ser  Glu Thr Ala Arg Asp  Ala Ala Ser Ala Ser  Leu Glu Ala
    1235                1240                1245

Ala Lys  Ser Thr Met Val Thr  Arg Gly Gly Gly Arg  Gly Ser Ser
    1250                1255                1260

Val Val  Ala Val Thr Arg Ser  Thr Ser Ser Pro Ser  Gly Arg Ala
    1265                1270                1275

Ala Ser  Val Ala Ser Ser Thr  Leu Gly Gly Phe Gly  Ala Arg
    1280                1285                1290


<210> SEQ ID NO 21
<211> LENGTH: 2406
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 21

Met Ala Ala Pro Ala Pro Ser Ala Glu Ala Arg Pro Ala Lys Arg Arg
1               5                  10                  15

Cys Phe Pro Leu Pro Arg Glu Thr Pro Val Ser Ser Glu Asp Glu Thr
            20                  25                  30

Arg Lys Thr Leu Gln His Asp Thr Leu Gly Cys Leu Pro Arg Ser Ser
        35                  40                  45

Ser Gly Gln Pro Glu Leu Ala Ala Ala Ser Ala Ala Ser Gln Val Gly
    50                  55                  60

His Leu Ser Ser Ala Ala Leu Leu Gln Leu Val Gln Thr Gln Ser Ala
65                  70                  75                  80

Gly Gly Val Pro Gln Ala Val Leu Arg Asn Leu Phe Ser Ser Ile His
                85                  90                  95

Arg Asn Pro Lys Pro Leu Pro Ala Asn Ala Leu Ala Ala Thr Pro Asn
            100                 105                 110

Ser Ser Leu Tyr Ala Ser Leu Thr Ser Leu Ser Ser Ala Ala Ala Leu
        115                 120                 125

Pro Gly Ala Gly Pro Ala Tyr Ser Gln Ala Pro Ser Pro Ala Ser Ala
    130                 135                 140

Asp Leu Leu Gln Ser Glu Gln Phe Arg Ser Ala Ala Lys Asn Pro Ser
145                 150                 155                 160
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Pro Asn Glu Ala Ser Pro Ile Leu Ala Leu Leu Gly Glu Ala Ala Arg
                165                 170                 175

Ala Ala Thr Thr Pro Arg Thr Val Pro Ala Leu Ser Ala Val Cys Pro
            180                 185                 190

Ala Ala Ser Ser Gly Val Ser Leu Pro Pro Ala Ser Asp Thr Leu Ala
        195                 200                 205

Leu Ala Gln Ser Ser Leu Ser Ser Ser Thr Gly Cys Ala Ser Asp Val
    210                 215                 220

Lys Ala Ser Arg Pro Glu Glu His Pro Ala Phe Ala Ser Gly Thr Ala
225                 230                 235                 240

Asn Arg Gln Ser Leu Leu Gln Ala Leu Leu Leu Ser Thr Ala Pro Leu
                245                 250                 255

Ala Phe Ser Gly Pro Ser Leu Ser Ser Ala Ser Thr Thr Leu Pro Ala
            260                 265                 270

Ser Ser Gly Ala Val Ser Ser Arg Asn Ala Gly Ala Tyr Gln Phe Glu
        275                 280                 285

Arg Leu Leu Gln Ala Glu Ala Ala Lys Val Lys Ala Leu Leu Pro Asn
    290                 295                 300

Thr Thr Ser Lys Ser Met Ser Gln Ser Ser Val Pro Gln Arg Asp Leu
305                 310                 315                 320

Thr Arg Lys Thr Ser Leu Phe Pro Asp Pro Arg Gly Leu Ser Ala Asp
                325                 330                 335

Asp Ala Ser Arg Arg Tyr Asn Thr Arg Gly Ala Asn Ser Gly Gly Ala
            340                 345                 350

Gly Leu Arg Arg Gly Thr Gly Val His Ala Thr Thr Glu Gln Ser Gly
        355                 360                 365

Ala Leu Asp Ala Gly Glu Arg Thr Arg Pro Phe Gly Ala Gly Glu Asp
    370                 375                 380

Glu Ser Ala Gln Gly Lys Pro Asp Ser Arg Gly Arg Gln Arg Pro Gly
385                 390                 395                 400

Ala Leu Asp Ala Ser Asn Ile Leu Gly Leu Leu Ala Ala Phe Gln Pro
                405                 410                 415

Ser Gln Ala Pro Ala Ile Arg Asp Leu Ser Ala Pro Ser His Leu Ser
            420                 425                 430

Ala Ala Ala Thr Gly Ala Leu Pro Leu Thr Ala Ser Phe Thr Ala Ser
        435                 440                 445

Ala Leu Ala Ser Ser Gln Cys Leu Pro Ala Gly Thr Pro Ala Ser Ser
    450                 455                 460

Ser Ala Ser Pro Pro Phe Ser Glu Val Leu Ser Thr Thr Glu Glu Ser
465                 470                 475                 480

Ser Thr Thr Lys Glu Thr Asp Ala Ser Ala Ser Thr Leu Leu Ala Phe
                485                 490                 495

Leu Gln Lys Tyr Ser Ala Val Ser Gly Leu Gly Gly Ala Ser Asp Phe
            500                 505                 510

Leu Gly Gln Leu Gln Gly Lys Thr Ser Leu Pro Pro Leu Ser Leu Ala
        515                 520                 525

Glu Pro Ser Ser Ala Leu Pro Ser Ser Phe Leu Gly Gly Ser Asp Gly
    530                 535                 540

Gly Thr Ile Asp Thr Arg Asn Gly Asn Gly Glu Lys Thr Thr Pro Pro
545                 550                 555                 560

Ile His Leu Phe Gln Ser Ala Phe Arg Met Pro Ser Pro Ser Gln Gln
                565                 570                 575
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```
Asn Leu Leu Asp Ala Leu Leu Ala Ser Ser Cys Thr Thr Ala Thr Ser
            580                 585                 590

Arg Ser Asp Gly Ser Gly Asn Leu Gly Cys Pro Val Val Asp Glu Arg
        595                 600                 605

Asn Ala Lys Leu Ala Gly Pro Ala His Pro Leu Pro Cys Ser Phe Pro
    610                 615                 620

Gln Ile Ser Ser Ser Ser Gly Glu Pro Gly Arg Lys Thr Gly Gly Arg
625                 630                 635                 640

Val His Arg Gln Gly Thr Ser Gln Ser Gly Gly Arg Val Arg Ser Gly
                645                 650                 655

Lys Asn Gly Gly Ser Ala Ala Pro Pro Arg Gln Ser Ser Ser Asp Asn
            660                 665                 670

Val Pro Ser Thr Pro Thr Val Ser Ser His Glu Ala Pro His Arg Ala
        675                 680                 685

Gly Phe Pro Ser Gln Thr Pro Tyr Glu Leu Ser Ala Ser Pro Ser His
    690                 695                 700

Gln Leu Asp Leu Leu Arg Leu Gly Ala Phe Leu Gly Gly Ala Gly Lys
705                 710                 715                 720

Gln Asp Ala Ser Val His Ser Asp Glu Thr Gly Thr Leu Ser Gly Glu
                725                 730                 735

Pro Ser His Arg Ser Cys Ser Leu Ser Arg Gly Leu Thr Gln Glu Ser
            740                 745                 750

Val Leu Gln Leu Ser Asp Thr Thr Ser Thr Ser Arg Glu Gly Glu Pro
        755                 760                 765

Asn Glu Pro Ser Gln Gly Cys Val Asn Val Ala Ala Ser Leu Pro Ala
    770                 775                 780

Phe Gly Pro Gln Pro Ser Ser Gly Ala Ala Lys Ala Arg Glu Gly Arg
785                 790                 795                 800

Arg Gly Ala Gly Gly Ala Gly Ala Ala Pro Pro Val Pro Leu Arg Ala
                805                 810                 815

Asp Val Thr Leu Gly Gly Asn Arg Pro His Tyr His Val Ala Lys Gln
            820                 825                 830

Glu Trp Arg Val Arg Tyr Tyr Met Asn Gly Lys Arg Lys Met Arg Thr
        835                 840                 845

Tyr Ser Ala Lys Phe Tyr Gly Tyr Glu Thr Ala His Thr Met Ala Glu
    850                 855                 860

Asp Phe Ala His Tyr Val Asp Lys His Glu Ala Leu Pro Asp Ser Met
865                 870                 875                 880

Met Met Thr Ala Met Met Leu Gln Ala Gln Ala Asn Ser Ala Ala Ser
                885                 890                 895

Ser Gly Gln Thr Val Pro Leu Ala Arg Gly Ile Arg Ala Ser Ser Ala
            900                 905                 910

Ser Thr Gly Ala Gly Gly His Val Ser Lys Ser Ala Thr Lys Gly Ser
        915                 920                 925

Val Ala Ala Ser Ser Glu Gly Ser Thr Ser Met Gly Ser Asp Ala Thr
    930                 935                 940

Arg Ser Gln Glu Gly Glu Ala Ala Glu Leu Cys Pro Leu Ala Ala Gly
945                 950                 955                 960

Leu Ser Arg Pro Leu Ala Ser Met His Ser Ala Ala Gly Asn Ala Val
                965                 970                 975

Ala Gln Gly Arg Gln Glu Ser Lys Glu Glu Ala Pro Gly Gly Gln Ala
            980                 985                 990

Trp Phe Gly Glu Pro Gly Lys Phe  Arg Ala Ser Ser Glu  Ala Ala Leu
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        995                 1000                1005

Cys Gly  Ser Gly Ser Ser Ala  Glu Gly Arg Asp Gly  His Glu Ser
    1010                1015                1020

Glu Val  Leu Trp Ala Thr Leu  Gly Lys Val His Asp  Ala Ser Gln
    1025                1030                1035

Gly Lys  Lys Ile Lys Pro Glu  Lys Pro Leu Thr Val  Ala Arg Gly
    1040                1045                1050

Arg Leu  Ala Leu Gly Ala Glu  Asp Lys Ser Gln Asn  Leu Gly Val
    1055                1060                1065

Asp Leu  Val Asp Ser Gly Glu  Ala Gln Gly Leu Pro  Gly Val Arg
    1070                1075                1080

Gln Pro  Arg Gln Met Lys Asn  Ser Glu Glu Cys Ser  Leu Arg Asp
    1085                1090                1095

Ser Asp  Lys Gly Met Ala Leu  Ser Lys Arg Phe Gly  Phe Leu Pro
    1100                1105                1110

Ser Gln  Thr Pro Ser Cys Asp  Ser Met Thr Leu Pro  Phe Pro Gly
    1115                1120                1125

Gly Phe  Asp Ala Leu Ser Leu  Ser Ser Ala Leu Ser  Ser Cys Ala
    1130                1135                1140

Ser Leu  Pro Val Ala His Glu  Gly Asn Asn Phe Gln  Lys Gly His
    1145                1150                1155

Thr Gly  Asp Ile Val Ala Leu  Ala Ser Gln Ser Gly  Thr Gln Arg
    1160                1165                1170

Pro Ala  Ser Val Val Leu Ser  Arg Asp Ala Asn Val  Ser Gly Ser
    1175                1180                1185

Ser Pro  Ser His Pro Thr Trp  Gln Arg Glu Gly Ala  Ala Val Ser
    1190                1195                1200

Gly Arg  Ala Asp Glu Phe Ser  Ser Leu Ser Val Thr  Pro Ser Thr
    1205                1210                1215

Val Pro  Leu Ser Ser Phe Thr  Met Glu Asp Ile Lys  Gly Glu Lys
    1220                1225                1230

Gly Asp  Pro Ser Arg Arg Phe  Ala Leu Val Gly Glu  Ser Met Lys
    1235                1240                1245

Asn Val  Ser Ala Pro Glu Val  Gln Ala Leu Phe Pro  Thr Ser Ser
    1250                1255                1260

Ile Ala  Asn Ala Glu Leu Leu  Pro Val Asp Phe Leu  His Ser Asn
    1265                1270                1275

Ser Cys  Ser Ala Asp Lys Leu  Glu Ser Ser Ile Pro  Arg Gly Leu
    1280                1285                1290

Ala Gly  Asn Asn Pro Ser Met  Thr Ala Thr Ala Val  Ala Ala Thr
    1295                1300                1305

Ala Val  Ser His Gln Ile Phe  Asp Thr Ile Thr Leu  Phe Gly Glu
    1310                1315                1320

Phe Leu  Arg Glu Phe Ala Lys  Glu Lys Val Asn Glu  Phe His Glu
    1325                1330                1335

Tyr Gly  Leu Glu Ala Ser Pro  Leu Thr Val Glu Ala  Ser Ser Glu
    1340                1345                1350

Val Ser  Leu Phe Gly Lys Ala  Thr Phe Gly Arg Cys  Pro Val Ala
    1355                1360                1365

Gly Gly  Ser Thr Pro Ala Gly  Ile Ser Lys Met Ser  Gly Glu Thr
    1370                1375                1380

Leu Ser  Gly Leu Ser Ala Ser  Glu Leu Ser Leu Val  Ser Ala Arg
    1385                1390                1395
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Thr Asn  Thr Thr Thr Gly Glu  Glu Gln Phe Ala Leu  Ala Arg Gly
    1400                1405                1410

Leu Phe  Pro Gly Asp Ser Glu  Gly Asp Arg Asp Glu  Lys Lys Pro
    1415                1420                1425

Gln Leu  Ser Gln Gln Glu Leu  Leu Val Leu Ser His  Ala Leu Val
    1430                1435                1440

Asn Leu  Thr Ser Ser Thr Tyr  Val Leu Met His Thr  Leu Lys Ala
    1445                1450                1455

Ser Leu  Ser Lys Ser Thr Glu  Ala Val Gln Leu His  Gln Pro Leu
    1460                1465                1470

Leu Glu  Ala Ala Ser Glu Ala  Lys Ala Thr Asp Glu  Ala Lys Thr
    1475                1480                1485

Arg Glu  Glu Gln Glu Ser Ser  Glu Cys Asp His Glu  Tyr Pro Pro
    1490                1495                1500

Arg Ser  Ser Leu Glu Ala Thr  Thr Gly Ala Leu Pro  Phe Arg Leu
    1505                1510                1515

Ser Pro  Ala Leu Ser Ala Ser  Ser Lys Asp Leu Pro  Ser Leu Ser
    1520                1525                1530

Ala Ser  Ala Ser Leu Glu Ser  Val Thr Pro Phe Ala  Gly Leu Pro
    1535                1540                1545

Leu Glu  Glu Gly Thr Leu Ser  Ala Ser Val Gly Leu  Ala Ser Ser
    1550                1555                1560

Asp Asp  Glu His Asp Thr Ser  Leu Leu Phe Lys Thr  Glu Ala Ala
    1565                1570                1575

Lys Lys  Arg Ser Leu Phe Ser  Thr Ala Ala Asp Gly  Asp Glu Ser
    1580                1585                1590

Arg Thr  Tyr Asn Asp Gly Leu  Gly Gln Pro Met Glu  Glu Glu Ile
    1595                1600                1605

Arg Ser  Cys Val Ser Thr Ser  Cys Gly Glu Ala Val  Ala Thr Thr
    1610                1615                1620

Thr Leu  Ser Ala Ile Gly Pro  Gly Thr Gly Ala Ser  Gly Ala Leu
    1625                1630                1635

Leu Asp  Ser Glu Ser Arg Glu  Ser Leu Gly Glu Lys  Pro Gly Ala
    1640                1645                1650

Ala Leu  Arg Ala Gly Ala His  Thr Pro Ala Pro Ser  Arg Ala Pro
    1655                1660                1665

Thr Pro  Ser Arg Thr Phe Ser  Phe Thr Ser Ser Ser  Thr Ala Thr
    1670                1675                1680

Ser Ala  Ala Leu Leu Cys Asp  Ser Asn Val Val His  Glu Lys Leu
    1685                1690                1695

Arg Ala  Gln Gly Lys Asp Ser  Glu Ala Gly Glu Arg  Lys Gly Asp
    1700                1705                1710

Ser Glu  Lys Glu Glu Glu Val  Glu Met Trp Lys Glu  Glu Asp Glu
    1715                1720                1725

Glu Val  Gln Arg Cys Thr Gly  Ser Ala Glu Thr Asp  Ser Thr Glu
    1730                1735                1740

Ala Thr  Arg Gly Glu Glu Ala  Trp Arg Arg Gly Lys  Gln Ser Glu
    1745                1750                1755

Lys Lys  Pro Ser Val Ile Thr  Thr Ala Leu Asn Leu  Leu Glu Thr
    1760                1765                1770

His Arg  His Leu Ala Leu Thr  Ile Ser Gln Leu Lys  Arg Pro Val
    1775                1780                1785
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Ala Gln  Gln Leu Arg Phe Ile  Leu Pro Ile Ala Ala  Pro Gln Leu
    1790                 1795                 1800

Leu Pro  Cys Ile Leu Pro Pro  Ala Ser Phe Gln Gly  Pro Gly Glu
    1805                 1810                 1815

Ser Gly  Asp Gly Lys Ala Glu  Ala Glu Ala Lys Gly  Ser Ser Ser
    1820                 1825                 1830

Leu Gly  Gln Val Leu Glu Thr  Ala Leu Gly His Gly  Thr Arg Leu
    1835                 1840                 1845

Ala Pro  Ser Ala Ser Ala Met  Val Pro Pro Arg Lys  Asp Glu Ala
    1850                 1855                 1860

Ala Ser  Ala Val Pro Glu Ala  Lys Thr Phe Thr Gly  Leu Ala Asn
    1865                 1870                 1875

Ala Gly  Val Met Arg Glu Ala  Ala Ser Arg Thr Leu  Glu Ala Glu
    1880                 1885                 1890

Gln Val  Ser Arg Lys Arg Ser  Arg Glu Glu Val Val  Asp Ser Glu
    1895                 1900                 1905

Thr Ala  Gly Asp Glu Gly Asp  Met Glu Asn Val Pro  Glu Thr Leu
    1910                 1915                 1920

Asp Ala  Thr Thr Ser Pro Gly  Ser Arg Gln Tyr Asp  Lys Ser Pro
    1925                 1930                 1935

Ser Asn  Gly Gly Thr Lys Pro  Pro Ala Thr Ala Lys  Ser Arg Val
    1940                 1945                 1950

Ile Arg  Asp Gln Ala Ala Leu  Glu Arg Leu Leu Leu  Ala Pro Phe
    1955                 1960                 1965

Gln Asp  Thr Pro Thr Cys Ser  Cys Thr Asp Arg Pro  Cys Pro Cys
    1970                 1975                 1980

Asp Arg  Gln Gln Val Ala Asp  Met Ile Tyr Leu Phe  Tyr Ala Val
    1985                 1990                 1995

Pro Ala  Arg Gln Gln Ala Glu  Ser Ser Lys Glu Gly  Ser Thr Gln
    2000                 2005                 2010

Arg Leu  Gln Phe Ala Ala Arg  Asp Thr Asn Glu Arg  Lys Asp Ala
    2015                 2020                 2025

Arg Thr  Gly Glu Glu Thr Gln  Gly Gly Glu Thr Glu  Ala Lys Glu
    2030                 2035                 2040

Val Ile  Arg Asp Pro Glu Glu  Arg Gly Val Cys Glu  Gly Ser Ser
    2045                 2050                 2055

Ser Gln  Asn Ala His Thr Gln  Phe Asp Ala Glu Thr  Ala Ser Ser
    2060                 2065                 2070

Ser Met  Ser Ser Asp Pro Arg  Ala Asp Lys Glu Ser  Asn Ala Gln
    2075                 2080                 2085

Asp Ala  His Met Ala Asp Lys  Thr Ser Phe Val Ser  Asp Leu Pro
    2090                 2095                 2100

Gln Pro  Ser Gly Glu Phe Ala  Pro Ser Leu Leu Ser  Glu Thr Ser
    2105                 2110                 2115

Leu Asp  Val Ala Met Ala Asp  Ser Arg Gly Thr Thr  Ser Glu Ile
    2120                 2125                 2130

His Gly  Phe Phe Thr Arg Ser  Asp Glu Gln Lys Arg  Ala Ser Phe
    2135                 2140                 2145

Ser Ser  Ser Ser Leu Leu Ala  Ala Gly His Ala Val  Ala Ser Phe
    2150                 2155                 2160

Ser Ser  Ser Leu Ala Gly Val  Val Ser Gly Ala Gly  Glu Arg Arg
    2165                 2170                 2175

Glu Cys  Ala Gly Pro Ser Leu  Gly Asp Leu Ser Thr  Ile Gly Leu
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    2180                2185                2190

Leu Ser  Leu Ser Tyr Pro Ala  Met Leu Ala Phe Ile  Leu Pro Leu
    2195                2200                2205

Gln Ser  Leu Leu His Met Val  Ser Gly Met Ile Leu  Thr Leu His
    2210                2215                2220

Lys Lys  Leu Ile His Arg Phe  Ile Cys Ala His Leu  Arg Leu Val
    2225                2230                2235

Leu Asp  Asp Asp Met Arg Arg  Pro Ala Gly Gly Ala  Leu Lys Ser
    2240                2245                2250

Arg Gly  Ala His Gly Asp Thr  Glu Ala Ala Glu Ala  Gln Val Glu
    2255                2260                2265

Arg Arg  Arg Arg Glu His Glu  Arg Glu Glu Thr Thr  Asn Leu Ala
    2270                2275                2280

Ile Gly  Tyr Arg Glu Gly Asn  Ala Glu Ala Ser Asn  Thr Phe Pro
    2285                2290                2295

Leu Val  Asp Thr Val Ser Ser  Leu Leu Ser Pro Gly  Ser Leu Arg
    2300                2305                2310

Gln Glu  Asn Ser Glu Val Glu  Arg Arg Asp Asn Asp  Glu Glu Arg
    2315                2320                2325

Leu Glu  Leu Ile Thr Gly Ile  Ala Arg Glu Ser Pro  Lys Pro Ser
    2330                2335                2340

Glu Lys  Asp Ser Val Ser Pro  Phe Leu Ser Thr Ala  Pro Cys Pro
    2345                2350                2355

Gly Thr  Glu Ala Glu Ser Ser  Asp Cys Ser Ala Ser  Ser Ala Cys
    2360                2365                2370

Ser Gly  Thr Pro Thr Glu Gly  Thr Glu Gly Gly Glu  Thr Gly Asp
    2375                2380                2385

Ile Ala  Ser Phe Leu Ser Pro  Ser Gly Asp Val Lys  Gln Thr Ile
    2390                2395                2400

Met Leu  Ala
    2405


<210> SEQ ID NO 22
<211> LENGTH: 711
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 22

Met Pro Leu Lys Thr Ser Trp His Cys Ser Cys Asn Ala Thr Phe Pro
1               5                   10                  15

Gly Asp Leu Leu Met Val Val Ala Asn His Asp Arg Val Gly Asn Trp
            20                  25                  30

Asn Pro Gln Asn Ser Val Val Leu Ser Thr Asp Ala Ser Ser Phe Pro
        35                  40                  45

Thr Trp Arg Ser Gly Glu Val Cys Phe Asp Glu Gln Gln Pro Val Arg
    50                  55                  60

Leu Glu Tyr Lys Leu Ile Ile Arg Arg Ala Ser Gly Glu Ile Tyr Trp
65                  70                  75                  80

Glu Pro Ile Pro Thr Asn Arg Val Val Thr Leu Thr Ala Asn Thr Ser
                85                  90                  95

Ser Val Ile Glu Asn Val Trp Gly Ser Leu Ala Thr Cys Ser Ile Thr
            100                 105                 110

Phe Phe Pro Leu Gln Pro Ile Pro Ser Pro Ser Phe Tyr Lys His Ala
        115                 120                 125
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Glu Arg Thr Lys Lys Glu Ala Ser Ser Val His Leu His Ser Ala Ser
    130                 135                 140

Ile Ser Asp Asp Ser Gly Ser Asp Thr Gly Thr Cys Ser Gln Val Asp
145                 150                 155                 160

Glu Ser Arg Thr Gln Arg Asn Val Arg Gly Gln Pro Ala Ser Val Gly
                165                 170                 175

Thr Gly Lys Ala Thr Ala Ala Glu Arg Gly Gly Lys Gly Tyr Val Met
            180                 185                 190

Pro His His Gln Cys Ser Thr Ser Gln Arg Arg His Ser Ile Ser Thr
        195                 200                 205

Gln Ala Ala Asp Glu Ala Ala Gly Gly Gly Asn Arg Val Ser Phe Lys
    210                 215                 220

Arg Ser Ala Phe Ile Leu Ala Asn Thr Gly Pro Ile Thr Asn Tyr Tyr
225                 230                 235                 240

Thr Val Ser Lys Thr Ile Gly Arg Gly Thr Trp Gly Glu Val Lys Leu
                245                 250                 255

Val Ile Asp Asn Gly Thr Gly Ala Arg Arg Ala Ala Lys Lys Ile Pro
            260                 265                 270

Lys Cys Tyr Val Glu Asp Ala Asp Arg Phe Arg Gln Glu Ile Glu Ile
        275                 280                 285

Met Lys Ser Leu Asp His Pro Asn Ile Val Arg Leu Tyr Glu Thr Phe
    290                 295                 300

Glu Asp Met Thr Asp Phe Tyr Leu Val Met Glu Tyr Cys Thr Gly Gly
305                 310                 315                 320

Glu Leu Phe Asp Arg Leu Val His Gln Gly Val Phe Thr Glu Ala Leu
                325                 330                 335

Ala Cys Arg Ile Met Arg Gln Ile Leu Ala Ala Val Ala Tyr Cys His
            340                 345                 350

Ala His Arg Val Ala His Arg Asp Leu Lys Pro Glu Asn Phe Leu Phe
        355                 360                 365

Leu His Asp Asn Pro Glu Ser Pro Ile Lys Leu Ile Asp Phe Gly Leu
    370                 375                 380

Ala Ala Arg Phe Lys Ser Gly Gln Pro Met Arg Thr Arg Ala Gly Thr
385                 390                 395                 400

Pro Tyr Tyr Val Ser Pro Gln Val Leu Glu Gly Arg Tyr Gly Pro Glu
                405                 410                 415

Cys Asp Val Trp Ser Ala Gly Val Met Met Tyr Ile Leu Leu Cys Gly
            420                 425                 430

Tyr Pro Pro Phe Asn Ala Pro Ser Asp Arg Ala Ile Met Asn Lys Val
        435                 440                 445

Arg Ala Gly His Tyr Thr Phe Pro Asp Ser Glu Trp Ser Arg Val Ser
    450                 455                 460

Leu Gln Ala Lys Asp Leu Ile Ser Arg Leu Leu Asp Arg His Pro Arg
465                 470                 475                 480

Thr Arg Ile Ser Ala Glu Gln Ala Leu Arg His Ala Trp Phe Ala Met
                485                 490                 495

His Ala Pro Gly Asp His Phe Glu Pro Leu Gly Leu Asp Ile Leu Ser
            500                 505                 510

Lys Phe Arg Arg Phe Gln Gly Leu Ser Arg Leu Lys Lys Leu Ala Leu
        515                 520                 525

Thr Val Ile Ala Gln His Leu Glu Asp Ser Glu Ile Glu Gly Leu Lys
    530                 535                 540

Asn Leu Phe Thr Gln Leu Asp Thr Glu Gly Asp Gly Val Leu Thr Val
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545                 550                 555                 560

Glu Glu Ile Arg Lys Gly Ile Glu Arg Ser Gly Val His Leu Pro Pro
                565                 570                 575

Asp Met Val Leu Glu Asp Val Leu Arg Glu Val Asp Thr Ala Gly Thr
            580                 585                 590

Gly Ser Ile Asp Tyr Thr Glu Phe Ile Ala Ala Cys Leu His Gln Ser
        595                 600                 605

His Tyr Ile Arg Glu Glu Ala Cys Arg Ala Ala Phe Arg Val Leu Asp
    610                 615                 620

Ile Asn Gly Asp Gly Leu Val Ser Ala Gln Glu Leu Arg Gln Val Phe
625                 630                 635                 640

His Met Ala Gly Asp Leu Glu Thr Asp Ala Ala Ala Glu Leu Leu Glu
                645                 650                 655

Ala Asp Ala Asp Gly Asp Gly His Ile Thr Phe Asp Glu Phe Cys Gly
            660                 665                 670

Leu Met Arg Lys Val Pro Ser Leu Ala Leu Val Thr Glu His Thr Val
        675                 680                 685

Ser Met Met Arg Arg Thr Cys Ser Arg Thr Asn Ile Ser Glu Ala Ser
    690                 695                 700

Leu Thr Pro Arg Ala Thr Gly
705                 710


<210> SEQ ID NO 23
<211> LENGTH: 368
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 23

Met Val Ser Arg Thr Leu Ser Leu Ser Met Ser Leu Phe Arg Ala His
1               5                   10                  15

Leu Val Phe Tyr Arg Cys Ala Leu Asn Leu Asn Ser Ser Tyr Asn Phe
            20                  25                  30

Gly Phe Leu Val Ala Met Thr Phe Val Leu Gln Ile Ile Thr Gly Ile
        35                  40                  45

Thr Leu Ala Phe Arg Tyr Thr Ser Glu Ala Ser Cys Ala Phe Ala Ser
    50                  55                  60

Val Gln His Leu Val Arg Glu Val Ala Ala Gly Trp Glu Phe Arg Met
65                  70                  75                  80

Leu His Ala Thr Thr Ala Ser Phe Val Phe Leu Cys Ile Leu Ile His
                85                  90                  95

Met Thr Arg Gly Leu Tyr Asn Trp Ser Tyr Ser Tyr Leu Thr Thr Ala
            100                 105                 110

Trp Met Ser Gly Leu Val Leu Tyr Leu Leu Thr Ile Ala Thr Ala Phe
        115                 120                 125

Leu Gly Tyr Val Leu Pro Trp Gly Gln Met Ser Phe Trp Gly Ala Thr
    130                 135                 140

Val Ile Thr Asn Leu Leu Ser Pro Ile Pro Tyr Leu Val Pro Trp Leu
145                 150                 155                 160

Leu Gly Gly Tyr Tyr Val Ser Asp Val Thr Leu Lys Arg Phe Phe Val
                165                 170                 175

Leu His Phe Ile Leu Pro Phe Ile Gly Cys Ile Ile Ile Val Leu His
            180                 185                 190

Ile Phe Tyr Leu His Leu Asn Gly Ser Ser Asn Pro Ala Gly Ile Asp
        195                 200                 205
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Thr Ala Leu Lys Val Ala Phe Tyr Pro His Met Leu Met Thr Asp Ala
    210                 215                 220

Lys Cys Leu Ser Tyr Leu Ile Gly Leu Ile Phe Leu Gln Ala Ala Phe
225                 230                 235                 240

Gly Leu Met Glu Leu Ser His Pro Asp Asn Ser Ile Pro Val Asn Arg
                245                 250                 255

Phe Val Thr Pro Leu His Ile Val Pro Glu Trp Tyr Phe Leu Ala Tyr
            260                 265                 270

Tyr Ala Val Leu Lys Val Ile Pro Ser Lys Thr Gly Gly Leu Leu Val
        275                 280                 285

Phe Met Ser Ser Leu Ile Asn Leu Gly Leu Leu Ser Glu Ile Arg Ala
    290                 295                 300

Leu Asn Thr Arg Met Leu Ile Arg Gln Gln Phe Met Thr Arg Asn Val
305                 310                 315                 320

Val Ser Gly Trp Val Ile Ile Trp Val Tyr Ser Met Ile Phe Leu Ile
                325                 330                 335

Ile Ile Gly Ser Ala Ile Pro Gln Ala Thr Tyr Ile Leu Tyr Gly Arg
            340                 345                 350

Leu Ala Thr Ile Leu Tyr Leu Thr Thr Gly Leu Val Leu Cys Leu Tyr
        355                 360                 365


<210> SEQ ID NO 24
<211> LENGTH: 84
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 24

Lys Leu Cys Lys Tyr His His Phe Leu Cys Ser Leu Thr Ser Gln Leu
1               5                   10                  15

Ser Tyr Leu Ile Gly Leu Ile Phe Leu Gln Ala Ala Phe Gly Leu Met
            20                  25                  30

Glu Leu Ser His Pro Asp Asn Ser Ile Pro Val Asn Arg Phe Val Thr
        35                  40                  45

Pro Leu His Ile Val Pro Glu Trp Tyr Phe Leu Ala Tyr Tyr Ala Val
    50                  55                  60

Leu Lys Val Ile Pro Ser Lys Thr Gly Gly Leu Leu Val Phe Met Ser
65                  70                  75                  80

Ser Thr Cys Gln


<210> SEQ ID NO 25
<211> LENGTH: 244
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 25

Met Ile Ala Val His His His Pro Thr Gly Leu Leu Lys Thr Ala Lys
1               5                   10                  15

Ser Val Gly Phe Gln Tyr Pro Thr Thr Leu Arg Leu Phe His Ile Gly
            20                  25                  30

Tyr Val Leu Gly Val Ile Tyr Gly Leu Leu Leu Ser Leu Val Leu Thr
        35                  40                  45

Ala Arg Glu Asn Tyr Tyr Ser Asp Ala Ser Met Ile Ser Thr Ile Val
    50                  55                  60

Leu Gly Val Ile Ile Ser Glu Thr Gly Leu Phe Ile Ser Phe Phe Trp
65                  70                  75                  80

Gly Val Tyr Thr Thr Ser Trp Thr Thr Gly Leu Asp Leu Glu Gly Leu
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                85                  90                  95

Cys Leu Pro Asp Pro Ser Ser Ile Val Leu Phe Met Thr Ile Met Leu
            100                 105                 110

Ser Ala Leu Ser Ile Val Val Ser Ser Val Tyr Leu Lys Asn Gln His
        115                 120                 125

Leu Tyr Thr Ser Cys Thr Asn Ile Met Ile Phe Thr Leu Val Val Ser
    130                 135                 140

Phe Leu Met Leu Val Cys Thr Glu Tyr Leu Gly Leu Ser Ile Tyr Ile
145                 150                 155                 160

Asn Asp Asn Gly Phe Gly Asn Gly Leu Phe Ile Leu Thr Gly Ile His
                165                 170                 175

Phe Ser His Val Ile Val Gly Ala Ile Leu Gly Phe Phe Asn Gln Gly
            180                 185                 190

Met Tyr Ser Ser Leu Val Thr Tyr Leu Pro Val Asn Cys Ile Thr Leu
        195                 200                 205

Ser Lys Cys Lys Gly Thr Leu Cys Lys Ile Phe Ser Glu Pro Phe Thr
    210                 215                 220

Ile Leu Tyr Leu His Phe Val Glu Ala Val Trp Ile Met Ile His Val
225                 230                 235                 240

Thr Phe Tyr Leu


<210> SEQ ID NO 26
<211> LENGTH: 156
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 26

Met Ser Leu Phe Arg Ala His Leu Val Phe Tyr Arg Cys Ala Leu Asn
1               5                   10                  15

Leu Asn Ser Ser Tyr Asn Phe Gly Phe Leu Val Ala Met Thr Phe Val
            20                  25                  30

Leu Gln Ile Ile Thr Gly Ile Thr Leu Ala Phe Arg Tyr Thr Ser Glu
        35                  40                  45

Ala Ser Cys Ala Phe Ala Ser Val Gln His Leu Val Arg Glu Val Ala
    50                  55                  60

Ala Gly Trp Glu Phe Arg Met Leu His Ala Thr Thr Ala Ser Phe Val
65                  70                  75                  80

Phe Leu Cys Ile Leu Ile His Met Thr Arg Gly Leu Tyr Asn Trp Ser
                85                  90                  95

Tyr Ser Tyr Leu Thr Thr Ala Trp Met Ser Gly Leu Val Leu Tyr Leu
            100                 105                 110

Leu Thr Ile Ala Thr Ala Phe Leu Gly Tyr Ala Thr Ser Asn Tyr Thr
        115                 120                 125

Thr Leu Cys Gln Glu Gly Ser Gln Ile Thr Leu Ile Ile Phe Val Ile
    130                 135                 140

Leu Ile His Gly Val Gln Leu Val Leu Phe Leu Gln
145                 150                 155


<210> SEQ ID NO 27
<211> LENGTH: 403
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 27

Trp Ala His His Met Met Thr Val Gly Leu Glu Val Asp Thr Arg Ala
1               5                   10                  15
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Tyr Phe Ser Ala Met Thr Ile Met Ile Ala Ile Pro Thr Gly Thr Lys
            20                  25                  30

Ile Phe Asn Trp Leu Gly Thr Tyr Met Ala Ser His Asn Thr Thr Arg
        35                  40                  45

Thr Ile Asp Leu Trp Ala Ala Leu Cys Phe Ile Leu Leu Phe Thr Leu
    50                  55                  60

Gly Gly Thr Thr Gly Val Val Met Gly Asn Ala Gly Met Asp Ile Ala
65                  70                  75                  80

Leu His Asp Thr Tyr Tyr Ile Val Ala His Phe His Phe Val Leu Ser
                85                  90                  95

Leu Gly Ala Ile Leu Ala Thr Ile Cys Gly Phe Val Phe Tyr Ser Lys
            100                 105                 110

Asp Met Phe Gly Asp Thr Leu Asn Leu Phe His Val Asn Thr Gly Ser
        115                 120                 125

Ser Pro Tyr Leu Asn Ile Trp Phe Val Val Phe Leu Ala Ser Ile Met
    130                 135                 140

Leu Ile Phe Leu Pro Met His Ile Leu Gly Phe Asn Val Met Pro Arg
145                 150                 155                 160

Arg Ile Pro Asp Tyr Pro Asp Tyr Leu Cys Tyr Ile Asn Thr Trp Cys
                165                 170                 175

Ser Ile Val Gln Leu Val Val Tyr Thr Pro Gln Lys Lys Leu Ile Asn
            180                 185                 190

Asn Pro Val Ser Glu Met Ile Thr Pro Ser Thr Met Val Leu Ile Ile
        195                 200                 205

Leu Ala Ser Glu Tyr Tyr Ser Tyr Leu Thr Thr Ala Trp Met Ser Gly
    210                 215                 220

Leu Val Leu Tyr Leu Leu Thr Ile Ala Thr Ala Phe Leu Gly Tyr Val
225                 230                 235                 240

Leu Pro Trp Gly Gln Met Ser Phe Trp Gly Ala Thr Val Ile Thr Asn
                245                 250                 255

Leu Leu Ser Pro Ile Pro Tyr Leu Val Pro Trp Leu Leu Gly Gly Tyr
            260                 265                 270

Tyr Val Ser Asp Val Thr Leu Lys Arg Phe Phe Val Leu His Phe Ile
        275                 280                 285

Leu Pro Phe Ile Gly Cys Ile Ile Ile Val Leu His Ile Phe Tyr Leu
    290                 295                 300

His Leu Asn Gly Ser Ser Asn Pro Ala Gly Ile Asp Thr Ala Leu Lys
305                 310                 315                 320

Val Ala Phe Tyr Pro His Met Leu Met Thr Asp Ala Lys Cys Leu Ser
                325                 330                 335

Tyr Leu Ile Gly Leu Ile Phe Leu Gln Ala Ala Phe Gly Leu Met Glu
            340                 345                 350

Leu Ser His Pro Asp Asn Ser Ile Pro Val Asn Arg Phe Val Thr Pro
        355                 360                 365

Leu His Ile Val Pro Glu Trp Tyr Phe Leu Ala Tyr Tyr Ala Val Leu
    370                 375                 380

Lys Val Ile Pro Ser Lys Thr Gly Gly Leu Leu Val Phe Met Ser Ser
385                 390                 395                 400

Thr Cys Gln
```
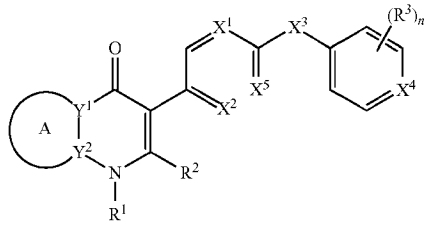

We claim:

1. A compound of the structure of

(a) Formula (I):



(I)

or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof,

wherein

ring A combines with Y^1 and Y_2 to form a C_{3-7}cycloalkenyl or heteroaryl ring,

wherein the C_{3-7}cycloalkenyl or heteroaryl is optionally substituted by halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR, cyano or phenyl;

Y^1 is N;

Y^2 is C or N;

X^1 is $C(R^{x1})$ or N,

wherein R^{x1} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^2 is $C(R^{x2})$ or N,

wherein R^{x2} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^3 is O, N(R), S or C_{1-3}alkyl;

X^4 is C or N;

X^5 is C or N;

R^1 is hydrogen or C_{1-3}alkyl;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR or —C(O)OR;

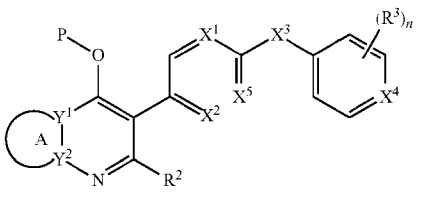
n is 0, 1, 2, 3 or 4;

each R^3 is independently halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5;

or two R^3 groups, together with the carbons to which they are attached, form a 1.3-dioxolane; and

each R is independently hydrogen or C_{1-3}alkyl; or

(b) Formula (I-p):



(I-p)

or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof,

wherein

ring A combines with Y^1 and Y_2 to form a C_{3-7}cycloalkenyl or heteroaryl ring, wherein the C_{3-7}cycloalkenyl or heteroaryl is optionally substituted by halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR, cyano or phenyl;

Y^1 is N;

Y^2 is C or N;

X^1 is $C(R^{x1})$ or N,

wherein R^{x1} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^2 is $C(R^{x2})$ or N,

wherein R^{x2} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^3 is O, N(R), S or C_{1-3}alkyl;

X^5 is C or N;

P is —C(O)OR', —C(O)R', —C(O)NR'_2, wherein R' is hydrogen, C_{1-3}alkyl or —CH_2OR;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, CH_2OH, —CH_2OR, —C(O)OR or —CH_2OP;

P is —C(O)OR', —C(O)R', —C(O)NR'_2 or —OP(O)(OR')OR', wherein each R' is independently hydrogen or C_{1-3}alkyl;

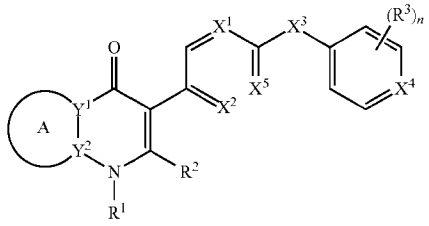
n is 0, 1, 2, 3 or 4;

each R^3 is independently halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5;

or two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane; and

each R is independently hydrogen or C_{1-3}alkyl.

2. The compound of claim 1, having the structure of (I):



(I)

or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof,

wherein

ring A combines with Y^1 and Y^2 to form a C_{3-7}cycloalkenyl or heteroaryl ring,

wherein the C_{3-7}cycloalkenyl or heteroaryl is optionally substituted by halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR, cyano or phenyl;

Y^1 is N;

Y^2 is C or N;

X^1 is $C(R^{x1})$ or N,

wherein R^{x1} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^2 is $C(R^{x2})$ or N,

wherein R^{x2} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^3 is O, N(R), S or C_{1-3}alkyl;

X^4 is C or N;

X^5 is C or N;

R^1 is hydrogen or C_{1-3}alkyl;

R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR or —C(O)OR;

n is 0, 1, 2, 3 or 4;

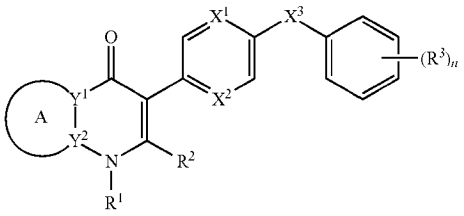
each R^3 is independently halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5;

or two R^3 groups, together with the carbons to which they are attached, form a 1,3-dioxolane; and

each R is independently hydrogen or C_{1-3}alkyl.

3. The compound of claim **1**, having the structure of (Ia):

(Ia)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof,

wherein

ring A combines with Y^1 and Y^2 to form a C_{3-7}cycloalkenyl or heteroaryl ring,

wherein the C_{3-7}cycloalkenyl or heteroaryl is optionally substituted by halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR, cyano or phenyl;

Y^1 is N;

Y^2 is C or N;

X^1 is $C(R^{x1})$ or N,

wherein R^{x1} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^2 is $C(R^{x2})$ or N,

wherein R^{x2} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^3 is O, N(R), S or C_{1-3}alkyl;

R^1 is hydrogen or C_{1-3}alkyl;

R^2 is hydrogen, C_{1-3}alkyl or —C(O)OR;

n is 0, 1, 2, 3 or 4;

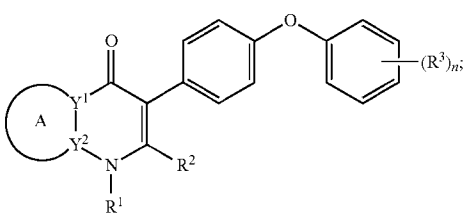
each R^3 is independently halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5; and

each R is independently hydrogen or C_{1-3}alkyl.

4. The compound of claim **3**, having

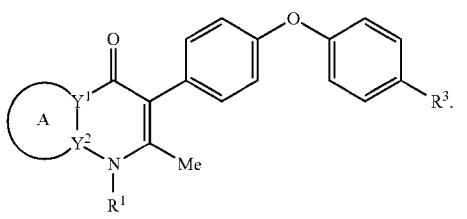
(a) the structure of Formula (Ib):

(Ib)



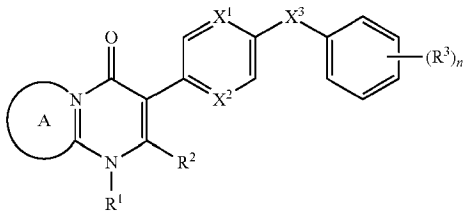
(b) the structure of Formula (Ic):

(Ic)



5. The compound of claim **3**, having the structure of Formula (III):

(III)

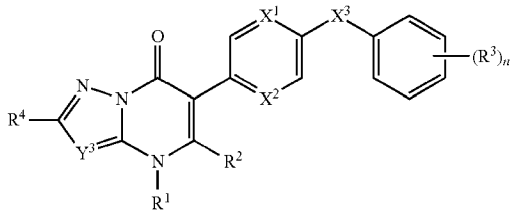


wherein

ring A combines with the nitrogen atom and carbon atom with which it is attached to form a heteroaryl ring.

6. The compound of claim **5**, having the structure of Formula (IIIa):

(IIIa)



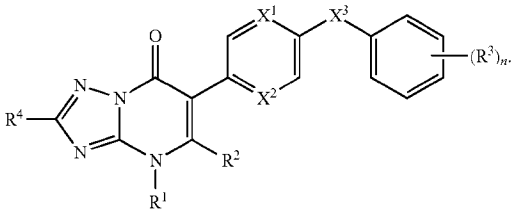
wherein

Y^3 is $C(R^5)$ or N; and

R^4 and R^5 are independently hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, O C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR, cyano or phenyl.

7. The compound of claim **6**, having the structure of Formula (IIIb):

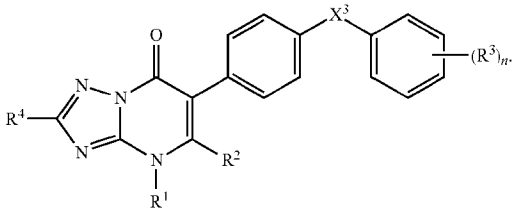
(IIIb)



8. The compound of claim **7**, wherein R^4 is hydrogen or C_{1-3}alkyl.

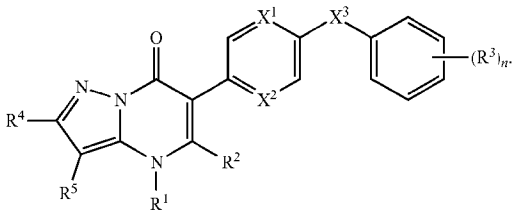
9. The compound of claim 7, having the structure of Formula (IIIb-1):

(IIIb-1)



10. The compound of claim 6, having the structure of Formula (IIIc):

(IIIc)



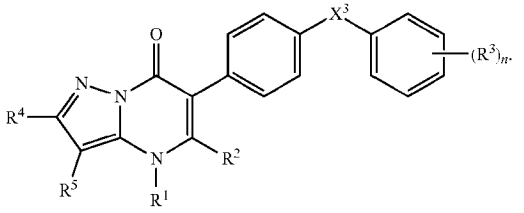
11. The compound of claim 10, wherein

R^4 is hydrogen or C_{1-3}alkylor phenyl; and

R^5 is hydrogen or cyano.

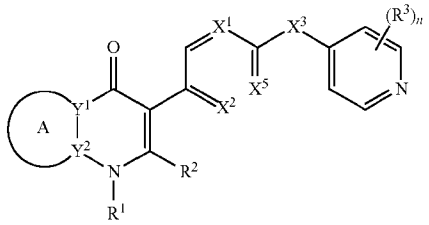
12. The compound of claim 10, having the structure of Formula (IIIc-1):

(IIIc-1)



13. The compound of claim 1, having the structure of Formula (IV):

(IV)



or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof,

wherein

ring A combines with Y^1 and Y_2 to form a C_{3-7}cycloalkenyl or heteroaryl ring,

wherein the C_{3-7}cycloalkenyl or heteroaryl is optionally substituted by halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR, cyano or phenyl;

Y^1 is N;

Y^2 is C or N;

X^1 is $C(R^{x1})$ or N,

wherein R^{x1} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^2 is $C(R^{x2})$ or N,

wherein R^{x2} is hydrogen, halogen, C_{1-3}alkyl, C_{1-3}alkoxy or C_{1-3}haloalkyl;

X^3 is O, N(R), S or C_{1-3}alkyl;

X^5 is C or N;

R^1 is hydrogen or C_{1-3}alkyl;

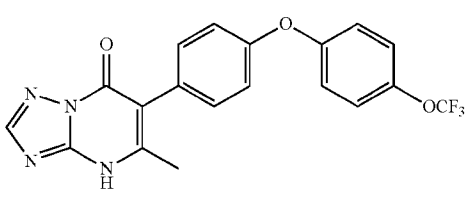
R^2 is hydrogen, C_{1-3}alkyl, C_{1-3}haloalkyl, —CH_2OH, —CH_2OR or —C(O)OR;

n is 0, 1, 2, 3 or 4;

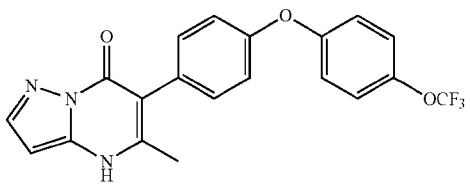
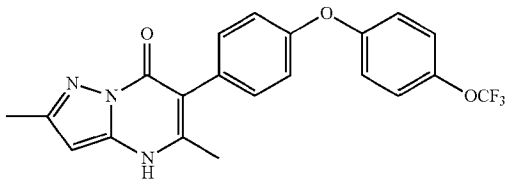
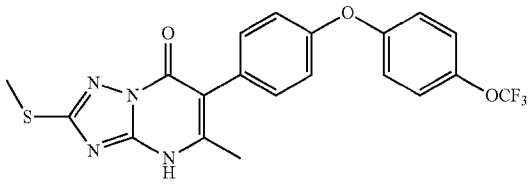
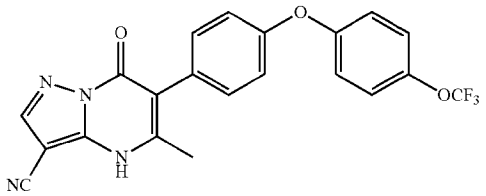
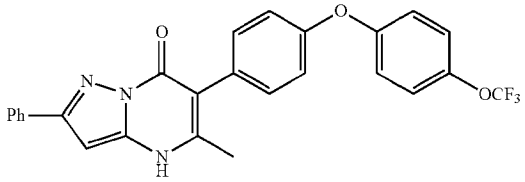
each R^3 is independently halogen, C_{1-3}alkyl, C_{1-3}alkoxy, C_{1-3}haloalkyl, —O—C_{1-3}haloalkyl, —S—C_{1-3}haloalkyl, —C(O)OR or SF_5; and

each R is independently hydrogen or C_{1-3}alkyl.

14. A compound that is:

Structure	Name
	5-methyl-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-[1,2,4]triazolo[1,5-a]pyrimidin-7(4H)-one

-continued

Structure	Name
	5-methyl-6-(4-(4-(trifluoromethoxy)phenoxy) phenyl)pyrazolo[1,5-a] pyrimidin-7(4H)-one
	2,5-dimethyl-6-(4-(4-(trifluoromethoxy)phenoxy) phenyl)pyrazolo[1,5-a] pyrimidin-7(4H)-one
	5-methyl-2-(methylthio)-6-(4-(4-(trifluoromethoxy)phenoxy) phenyl)-[1,2,4]triazolo[1,5-a] pyrimidin-7(4H)-one
	5-methyl-7-oxo-6-(4-(4-(trifluoromethoxy)phenoxy) phenyl)-4,7-dihydropyrazolo[1,5-a] pyrimidine-3-carbonitrile
	5-methyl-2-phenyl-6-(4-(4-(trifluoromethoxy)phenoxy) phenyl)pyrazolo[1,5-a] pyrimidin-7(4H)-one

5-methyl-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-[1,2,4]triazolo[1,5-a]pyrimidin-7(4H)-one;

5-methyl-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl) pyrazolo[1,5-a]pyrimidin-7(4H)-one;

2,5-dimethyl-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl) pyrazolo[1,5-a]pyrimidin-7(4H)-one;

5-methyl-2-(methylthio)-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-[1,2,4]triazolo [1,5-a]pyrimidin-7(4H)-one;

5-methyl-7-oxo-6-(4-(4-(trifluoromethoxy)phenoxy)phenyl)-4,7-dihydropyrazolo[1,5-a]pyrimidine-3-carbonitrile;

5-methyl-2-phenyl-6-(4-(4-(trifluoromethoxy)phenoxy) phenyl)pyrazolo[1,5-a]pyrimidin-7(4H)-one;

or a stereoisomer thereof, or a pharmaceutically acceptable salt thereof.

15. A pharmaceutical composition comprising a compound of claim **1** and a pharmaceutically acceptable diluent, excipient, or carrier.

16. A method for treating an apicomplexan parasitic infection, comprising administering to a subject (such as a human subject) in need thereof an amount effective to treat the infection of the compound or pharmaceutical composition of claim **1**.

17. An invention selected from the group consisting of:

(a) a method for monitoring treatment of an apicomplexan parasitic infection, such as *T. gondii* infection (including but not limited to any of the treatment of claim **16**), comprising monitoring expression, protein in serum or plasma, and/or activity of 1, 2, 3, 4, 5, 6, 7. 8, 9, 10, or all of the markers listed in FIGS. **1-2** in a subject (such as a human subject) being treated for an apicomplexan parasitic infection, wherein a decrease or increase in expression and/or presence and/or activity of the one or more markers indicates that the treatment is effective;

(b) a cell line infected with an apicomplexan parasite, wherein the apicomplexan parasite genome comprises a gene encoding an Apetela 2 IV-4 protein with an M=>I modification at residue 570 ("AP2 IV-4 M570I") compared to its orthologous gene on the reference *T. gondii* ME49 strain (gene ID: TGME49_318470);

(c) a method for treating an apicomplexan parasite infection (such as a *T. gondii* infection), comprising admin-

istering to a subject in need thereof an amount effective to treat the infection of an inhibitor (of up-regulated genes) or an activator (of down-regulated genes) of 1 or more of the up-regulated genes listed in FIG. **1** or FIG. **2**;

(d) a method for identifying test compounds for apicomplexan parasite therapy, comprising identifying test compounds that reduce expression (for up-regulated genes), or increase expression (for down-regulated genes) of 1 or more of the apicomplexan parasite genes in FIGS. **3-5**;

(e) a plurality of isolated probes that in total selectively bind to at least 2, 3, 4, 5, 6, 7, 8, 9, 10, 15, 20, 25, 30, 35, 40, 45, 100, 150, 200, 250, 500, or all of the markers listed in FIGS. **3-5**, complements thereof, or their expression products, or functional equivalents thereof wherein at least 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, or all of the probes in total are selective for markers that are upregulated in the EGS strain of *T. gondii* after infection of human fibroblasts, human neuronal stem cells or human monocytic lineage cells;

(f) a plurality of isolated probes that in total selectively bind to at least 2, 3, 4, 5, 6, 7, 8, 9, 10, 15, 20, 25, 30, 35, 40, 45, 100, 150, 200, 250, 500, or all of the markers listed in FIG. **1-2**, complements thereof, or their expression products, or functional equivalents thereof, wherein at least 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, or all of the probes in total are selective for markers that are upregulated in human fibroblasts, human neuronal stem cells or human monocytic lineage cells after infection with *T. gondii*, including but not limited to infection with the EGS strain of *T. gondi*.

* * * * *