**Supplementary Information**

Construction of recombinant target vector pKO2.1-LSL-hK-rasG12D-IRES-HSV1-tk

The recombinant target vector was constructed using in-fusion cloning technology in 4 steps.

Step1: Constructing pLOX-hKrasG12D-iresTK vector. hK-rasG12D was amplified from pcDNA3-*KRAS*G12D and pLOX-gfp-iresTK vector was double digested by XohI/BamHI After removing gfp, the fragment of hK-rasG12D was cloned to form pLOX-hK-rasG12D-iresTK vector, which was structurally confirmed by the digestion analysis of restriction endonuclease (Fig.S1) as well as verified by sequencing.

Step2: Subcloning 3’-homologous arm into pKO2.1. The 7 kb homologous arm fragment was amplified from goat genome DNA and subcloned into pKO2.1. The recombination plasmid pKO2.1-long arm was identified, which was structurally confirmed by the digestion analysis of restriction endonuclease (Fig.S2).

Step3: Amplifying target fragments. Table S1 described the sequences of PCR primers used for target fragment amplification. The primer design was based on the strategy of adding a 15 base sequence homologous to the 15 bases at one end of the DNA fragment to which it will be joined to the 5'-end to facilitate In-Fusion Cloning. The DNA fragments of 5'-homolous arm and splicing acceptor sequence (SA) were amplified from goat genome DNA. The fragments of loxP-Stop-LoxP (LSL) were constructed from PGKneotpAlox2 and the fragment of hK-rasG12D-iresTK was amplified from pLOX-hKrasG12D-iresTK. Fragments of SA and hK-rasG12D-iresTK were cloned into T-vector and SA-hK-rasG12D-iresTK as a whole fragment was amplified. Fig.S3 showed all the amplified fragments and inserted vector.

Step4: Cloning target vector by In-fusion PCR. The linearized vector of Pko2.1-long arm was generated using unique restriction enzymes-AgeI. In-fusion cloning reaction with a total 10 μL volume was carried out, containing 3 fragments obtained from Step 3 and the linearized vector with a molar ratio of 2:2:2:1. Fig. S4 confirmed the sizes of four fragments and the whole target plasmid. After transformation, 29 colonies were screened by colony PCR and 3 positive colonies were identified and the corresponding plasmids were verified by the digestion analysis of restriction endonuclease (Fig.S5). The junction sequences among the fragments were verified by sequencing.