Supplementary materials

Western blot

Cell lysate samples were collected from cells and subjected to SDS-PAGE, then transferred to polyvinylidene difluoride membrane (Millipore, Billerica, MA, USA). The membrane was blocked with 5% fat-free milk and incubated with specific primary antibodies overnights at 4°C. And this was followed by incubation with HRP-conjugated secondary antibodies for 1 h at room temperature. The expression levels of proteins were detected with an enhanced chemiluminescence system (Thermo Fisher Scientific, Waltham, USA).

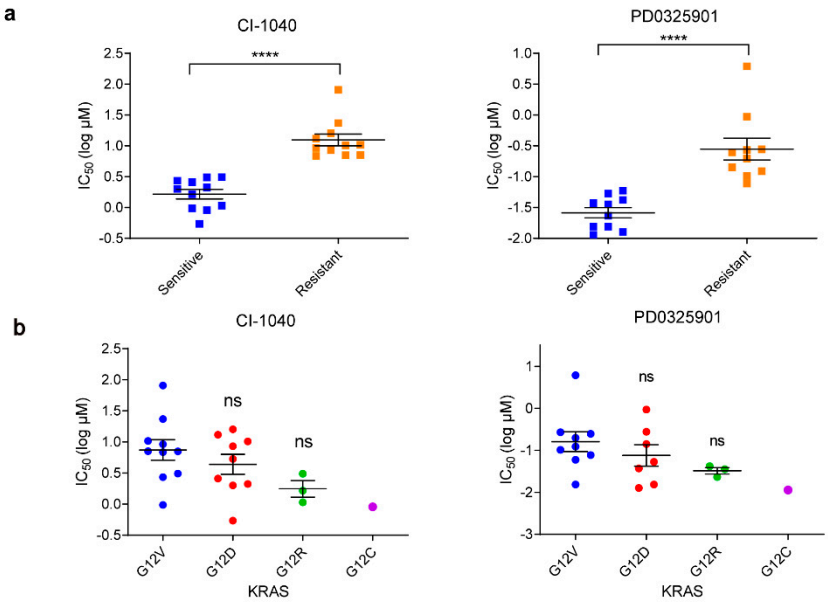
Immunohistochemical (IHC) assay

The tumor tissues of mice were deparaffinized and treated with 3% hydrogen peroxide for 10 min. 10 mmol/l sodium citrate buffer was used to retrieval antigen by heating for 15 min. Then the tissues were blocked with 10% goat serum and were incubated with primary antibody (1:200) at 4°C overnight. Then, the slides were incubated with HRP-conjugated secondary antibody for 1 h at room temperature.

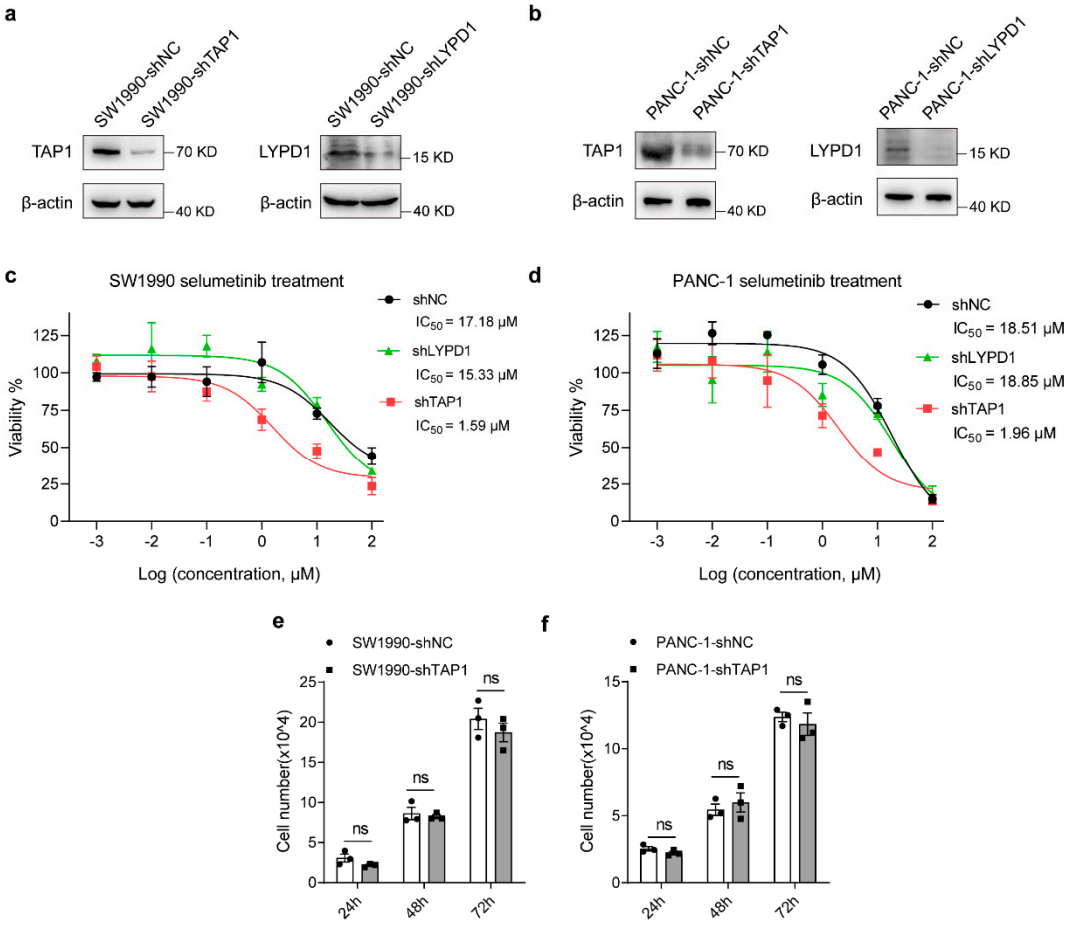
Immunofluorescence (IF)

To stain the TAP1 protein at cell surface membrane, cells cultured on 96-well plates were blocked with 10% goat serum and stained with anti-TAP1 antibody overnight at 4°C. Then cells were fixed in 4% formaldehyde for 5 min. Binding of corresponding fluorescent-conjugated secondary antibodies was carried out at room temperature in dark for 1 h. Cells were subsequently stained for nucleus with DAPI. Images were acquired with Opera Phenix™ High Content Analysis System (PerkinElmer, Waltham, USA).

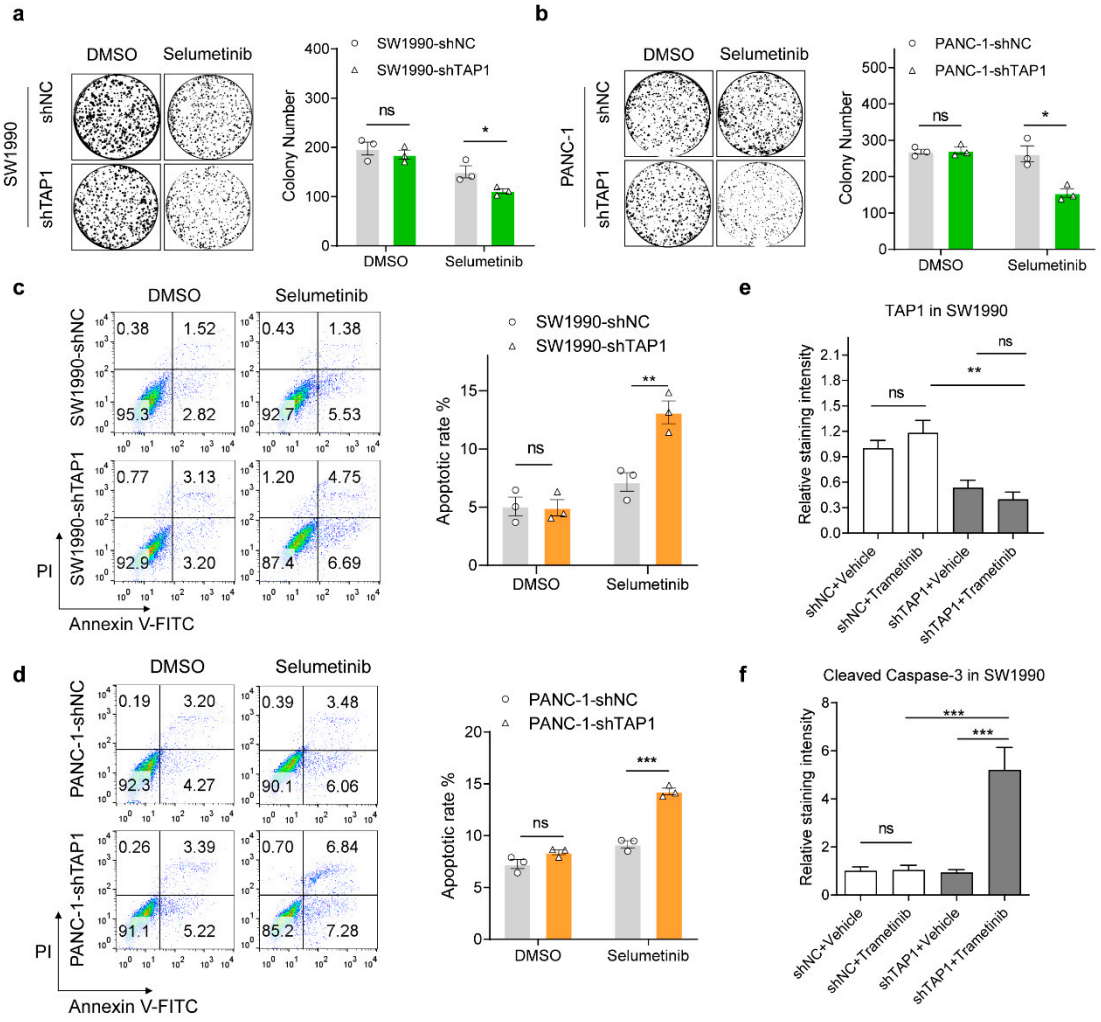
Supplementary figures



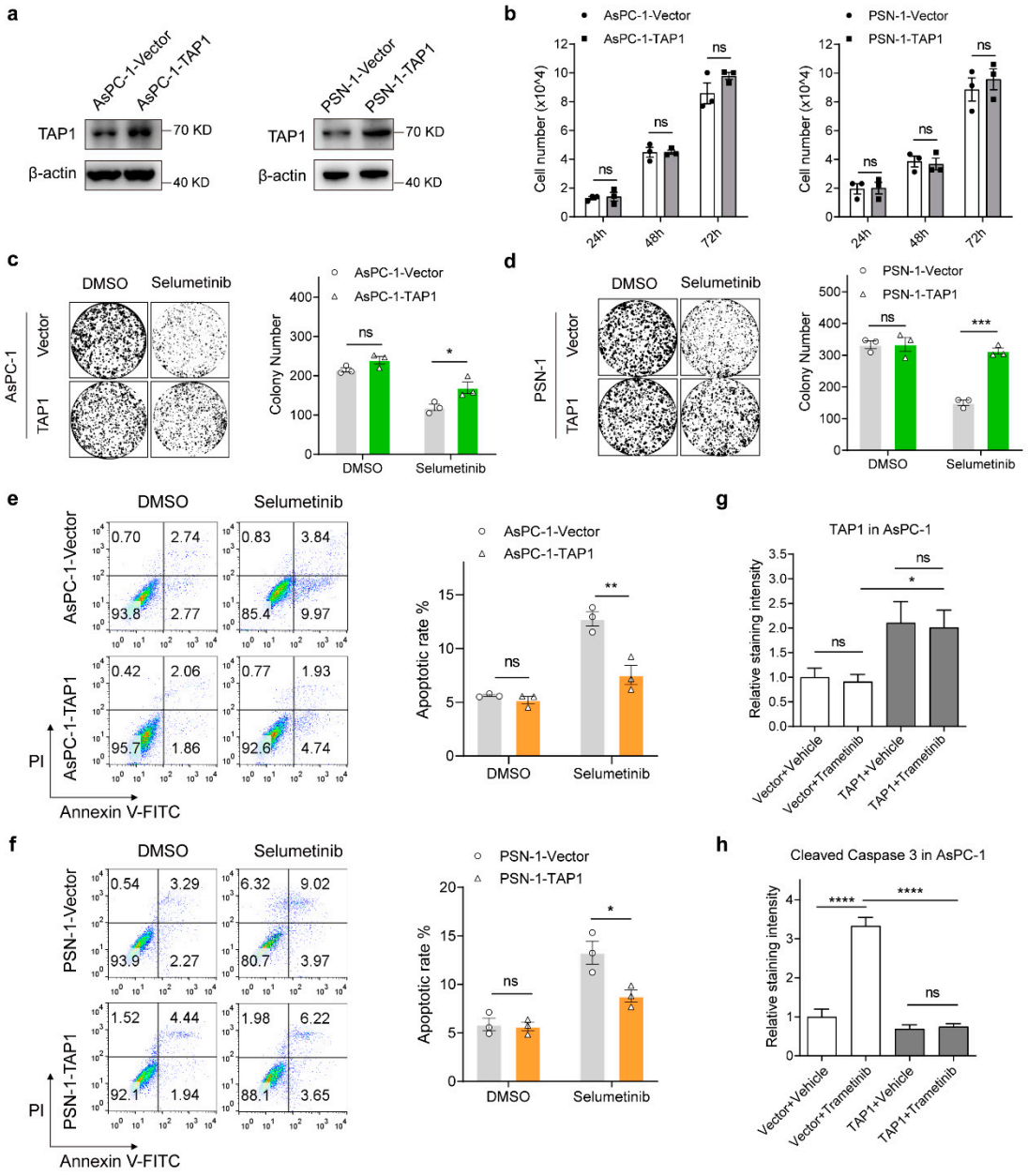
Supplementary Figure S1. *KRAS*-driven pancreatic cancer cells have different sensitivities to MEK inhibitors. (**a**) The response of *KRAS* mutation pancreatic cancer cell lines to CI-1040 and PD0315901. ****P < 0.0001. Data of logIC50 was from the Genomics of Drug Sensitivity in Cancer (GDSC) database. (**b**) IC50 values of cell lines with specific *KRAS* mutational subtypes were compared with cell lines with *KRAS* (G12V) to determine the P-value. Mann-Whitney test. ns, not significant.



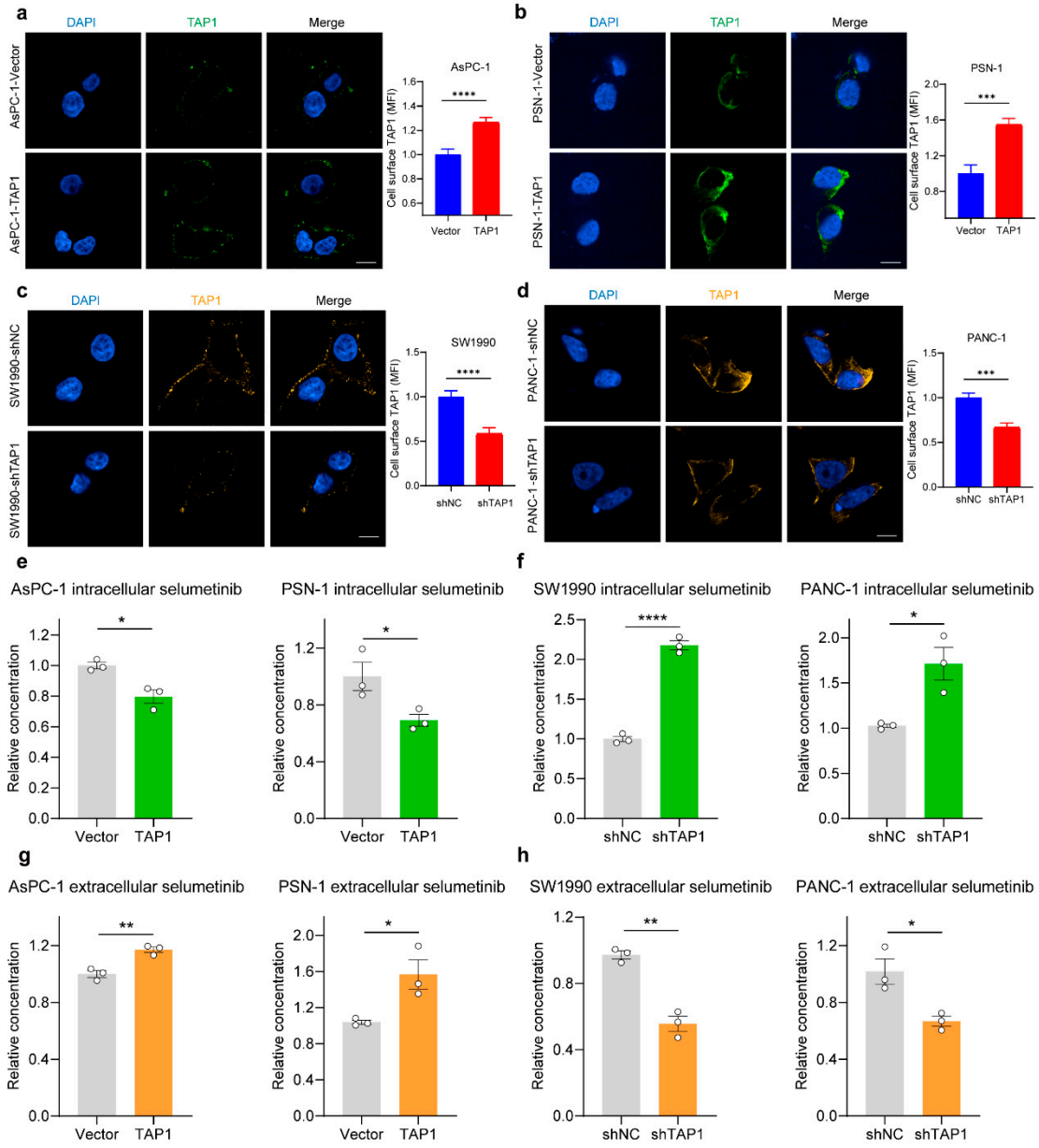
Supplementary Figure S2. The sensitivity of MEKi-resistant cells to selumetinib is increased by TAP1 knockdown. (**a, b**) Western blot of the expression level of TAP1 in SW1990 and PANC-1 cells with TAP1 shRNA or LYPD1 shRNA. Nontargeting shRNA (NC) was used as control. (**c, d**) Selumetinib sensitivities for SW1990 and PANC-1 cells with shTAP1, shLYPD1 and shNC were determined by CellTiter-Lumi™ assays. (**e, f**) The cell numbers of each cell lines were counting by Countstar software. ns, not significant.



Supplementary Figure S3. S**uppression of TAP1 enhances the proliferation inhibitory and pro-apoptotic effect induced by selumetinib**. (**a, b**) Representative wells of the colony formation assay upon knockdown of TAP1 or NC in SW1990 and PANC-1 cells after selumetinib treatment for 12 days. Quantification of colony number was shown at the right. (**c, d**) Representative FCM images of shTAP1 or shNC in SW1990 and PANC-1 cells were treated by selumetinib for 72h. The quantitative data were showed at the right. (**e, f**) The relative staining intensity of TAP1 and cleaved caspase-3 in tumor tissues from the indicated groups of mice in figure 3i. Data were represented as mean ± SEM; **P < 0.01; ***P < 0.001; ns, not significant. Two-tailed t test.



Supplementary Figure S4. Overexpression of TAP1 increases the resistance of MEKi-sensitive cells to selumetinib treatment. (**a**) Expression of TAP1 in TAP1-overexpression AsPC-1 and PSN-1 cells were examined by western blotting, compared to their repective vector control cells. (**b**) The cell numbers of each cell lines were counting by Countstar software. (**c, d**) Representative wells of the colony formation assay in TAP1-overexpressing AsPC-1 and PSN-1 cells under selumetinib treatment for 12 days. AsPC-1-Vector and PSN-1-Vector cells were as control. Quantification of colony number was shown at the right. (**e, f**) Representative FCM images of TAP1-ovexpressing AsPC-1 cells after treatment of selumetinib for 72 h. TAP1-Vector and PSN-1-Vector cells were as control. The quantitative data were showed at the right. (**g, h**) The relative staining intensity of TAP1 and cleaved caspase-3 in tumor tissues from the indicated groups of mice in figure 4k. Data were represented as mean ± SEM; *P < 0.05; ***P < 0.001; ****P < 0.0001; ns, not significant. Two-tailed t test.



Supplementary Figure S5. TAP1 overexpression increased the cell surface TAP1 expression and drug efflux in PDAC cells. (**a, b**) Confocal microscopy showed the cell surface expression of TAP1 in AsPC-1 and PSN-1 cells with TAP1 overexpression and vector control cells. TAP1, green; DAPI, blue. Scale bars, 10 μm. Quantification data of was shown at the right. MFI, mean fluorescence intensity. (**c, d**) Cell surface expression of TAP1 in SW1990 and PANC-1 cells with TAP1 knockdown and control cells (shNC) were visualized by confocal microscope. TAP1, yellow; DAPI, blue. Scale bars, 10 μm. Quantification data of MFI was shown at the right. (**e, f**) Intracellular drug accumulation after incubation with 2 μM selumetinib in TAP1-overexpressing and TAP1-knockdown cells were determined by LC-MS/MS, compared to their respective control cells. (**g, h**) Measurement of extracellular selumetinib in the cell culture medium of TAP1-overexpressing and TAP1-knockdown cells by LC-MS/MS, compared to their respective control cells. Data were represented as mean ± SEM; *P < 0.05; **P < 0.01; ***P < 0.001; ****P < 0.0001; two-tailed t test.

Supplementary tables

Table S1. Pancreatic cancer cells used for analyzed

Cell line	TCGA classification	Tissue	G12
AsPC-1	PAAD	pancreas	D
SUIT-2	PAAD	pancreas	D
PSN1	PAAD	pancreas	R
PA-TU-8902	PAAD	pancreas	V
KP-2	PAAD	pancreas	R
PANC-04-03	PAAD	pancreas	D
PANC-03-27	PAAD	pancreas	V
Capan-2	PAAD	pancreas	V
PANC-10-05	PAAD	pancreas	D
MZ1-PC	PAAD	pancreas	V
CFPAC-1	PAAD	pancreas	V
DAN-G	PAAD	pancreas	V
MIA-PaCa-2	PAAD	pancreas	C
KP-1N	PAAD	pancreas	D
HuP-T4	PAAD	pancreas	V
YAPC	PAAD	pancreas	V
PANC-02-03	PAAD	pancreas	D
HuP-T3	PAAD	pancreas	R
CAPAN-1	PAAD	pancreas	V
PL4	PAAD	pancreas	D
SW1990	PAAD	pancreas	D
KP-4	PAAD	pancreas	D
SU8686	PAAD	pancreas	D
PANC-08-13	PAAD	pancreas	D
PA-TU-8988T	PAAD	pancreas	V
HPAF-II	PAAD	pancreas	D

Table S2. Primers used for qPCR

Primers used for qPCR		
Gene		Sequence
β-actin	F 5'-3'	ACTCTTCCAGCCTTCCTTCC
	R 5'-3'	CGTCATACTCCTGCTTGCTG
IFI44L	F 5'-3'	ACAGAGCCAAATGATTCCCTATG
	R 5'-3'	TCGATAAACGACACACCAGTTG
KRT17	F 5'-3'	GGTGGGTGGTGAGATCAATGT
	R 5'-3'	CGCGGTTCAGTTCCTCTGTC
IFI27	F 5'-3'	TGCTCTCACCTCATCAGCAGT
	R 5'-3'	CACAACTCCTCCAATCACAACT
IL8	F 5'-3'	TTTTGCCAAGGAGTGCTAAAGA
	R 5'-3'	AACCCTCTGCACCCAGTTTTC
RBP1	F 5'-3'	TCCAGTCACTCCCCGAAATG
	R 5'-3'	AGGTACTCCTCGAAATTCTCGTT
AADAT	F 5'-3'	AATTACGCACGGTTCATCACG
	R 5'-3'	TCCTCTGCTCAATATGTCAGTCA
TAP1	F 5'-3'	TGCCCCGCATATTCTCCCT
	R 5'-3'	CACCTGCGTTTTCGCTCTTG
SLFN5	F 5'-3'	GAGTGTGTTGTAGATGCAGGAA
	R 5'-3'	ACTGCTCGCAGGATGATTTCA
TMEM163	F 5'-3'	CCGCCTGAAACCTCACGAA
	R 5'-3'	TCATAACGGAGACAGTAAAGGCA
INHBB	F 5'-3'	GTGAAGCGGCACATCTTGAG
	R 5'-3'	GCGAAGCTGATGATTTCGGAAAC
SCG5	F 5'-3'	ATGCTATCTGGCCTACTGTTTTG
	R 5'-3'	GGCCCACAAGATTCATGGC
TCEA3	F 5'-3'	CCCCAAAACACCTAGCAGC
	R 5'-3'	CTTCATGTCCGTGCTCTTGAG
AJUBA	F 5'-3'	ATGGGGAAGTCCTATCATCCAG
	R 5'-3'	TGGTAGTCGGTGACACAGTAT
MFI2	F 5'-3'	ACCTCCTATTACGCCGTGG
	R 5'-3'	AGGGACTCAGAGTAACTGGTC
COL9A3	F 5'-3'	GTGGATGGTCTGACTGGACG
	R 5'-3'	GGGCAGATACTTGGGCACTG
IL7R	F 5'-3'	TGTCGTCTATCGGGAAGGAG
	R 5'-3'	CGGTAAGCTACATCGTGCATTA
LYPD1	F 5'-3'	GGCAACTTTTTGCGGATTGTT
	R 5'-3'	CGTTCACCGTGCAATTCACA
ID2	F 5'-3'	GCTATACAACATGAACGACTGCT
	R 5'-3'	AATAGTGGGATGCGAGTCCAG
C10orf116	F 5'-3'	CAGGAAACCATCGACAAGACTG

	R 5'-3'	TCATTTCAGGAGGCCGAATTTTT
SLFN12	F 5'-3'	TGAGTGACCTCGATGACTTAGAA
	R 5'-3'	ACACGGTTATCTTTCACATGCC
TMC8	F 5'-3'	AGAACTACCCTCCCAACACG
	R 5'-3'	TTGTAGCCGTAGGACTCACAG
LCP1	F 5'-3'	GATCAGTGTCCGATGAGGAAATG
	R 5'-3'	CCAGATCACCTGTAGCCATCA
GUCY1B3	F 5'-3'	TGCTGGTGATCCGCAATTAC
	R 5'-3'	CCAGGACACGCAAGATTGTATC
ALDH1A1	F 5'-3'	GCACGCCAGACTTACCTGTC
	R 5'-3'	CCTCCTCAGTTGCAGGATTAAAG
BMI1	F 5'-3'	CCACCTGATGTGTGTGCTTTG
	R 5'-3'	TTCAGTAGTGGTCTGGTCTTGT
CXCR4	F 5'-3'	ACGCCACCAACAGTCAGAG
	R 5'-3'	AGTCGGGAATAGTCAGCAGGA
EpCAM	F 5'-3'	TGATCCTGACTGCGATGAGAG
	R 5'-3'	CTTGTCTGTTCTTCTGACCCC
OCT4	F 5'-3'	CTTGAATCCCGAATGGAAAGGG
	R 5'-3'	GTGTATATCCCAGGGTGATCCTC
c-Myc	F 5'-3'	CGACTCTGAGGAGGAACAAG
	R 5'-3'	GTGATCCAGACTCTGACCTT
NESTIN	F 5'-3'	GGCGCACCTCAAGATGTCC
	R 5'-3'	CTTGGGGTCCTGAAAGCT