

Supplementary Tables:

Supplementary Table 1: TFPI2 Primers used for MSP, BSS, RT and Stool DNA methylation assay

	Sense	Antisense	Size	Strand
TFPI2				
MSP1 U	GTGGTGGGGTGATAGTTTTTG	CCAAACAACCCAAATACCCACTTTATACA	109	1
MSP1 M	CGGCGGGGTGATAGTTTTC	CAAACGACCCGAATACCCGCTTTATACG	106	1
MSP2* U	CCCACATAAAACAACACCCAAACCA	TGGTTTGTGGGTAAGGTGTTTG	93	2
MSP2* M	GTTTCGTTGGGTAAGGCGTTC	CATAAAACGAACACCCGAACCG	86	2
* Primer set used for MSP in FFPE and frozen samples.				
MSP3 U	AAGTTGTGATTTTTTTTTGTTAGGTGTTTTTTTG	AACAAACAAAATCCATAATACAAAAATCAAACA	88	1
MSP3 M	CGTATTTTTTTTCGTTAGGCGTTTTTTTC	CGAAATCCATAATACAAAAATCGAACG	75	1
MSP4 U	GGGTTTTGTGTAGGTGTTTTTAAGTTTTG	CCCCCTACCAACAAAACACA	70	1
MSP4 M	GGGTTTCGTGTAGGCGTTTTTTAAGTTTC	CCCTACCAACGAAACGCG	67	1
BSS	GGTTTATGGTGTAGGGG	CAATCACTAACAAATCATTTC		2
RT TFPI2	GGGCCCTACTTCTCCGTTAC	CACACTGGTCGTCCCACTC	183	
ACTB	CATCCACGAAACTACCTTCAACTCC	GAGCCGCCGATCCACACG	213	
HPRT	CTTTGCTGACCTGCTGGATT	GTTGAGAGATCATCTCCACC	150	

Stool DNA methylation assay

Nested primers used for pilot feasibility study

#Flank MSP 5	GTGTATGAATTAGTTATTTTTTAGGTTT	CTAAACAAAACRTCCRAAAAAAC	144	1
MSP5# U	TTAGTTATTTTTAGGTTTTGTTTTGGT	AAAAACACCTAACAAAAAAAATACACA	118	1
MSP5# M	ATTTTTTAGGTTTCGTTTCGGC	GCCTAACGAAAAAAAATACGCG	106	1

Primers for quantitative MSP (qMSP)

TFPI2_qMSP	GTTTCGTTGGGTAAGGCGTTC	CATAAAACGAACACCCGAACCG	87	2
Molecular beacon	FAM—CGACATGCACCGCGCACCTCCTCCCGCCAAGCATGTCTG—DABCYL			

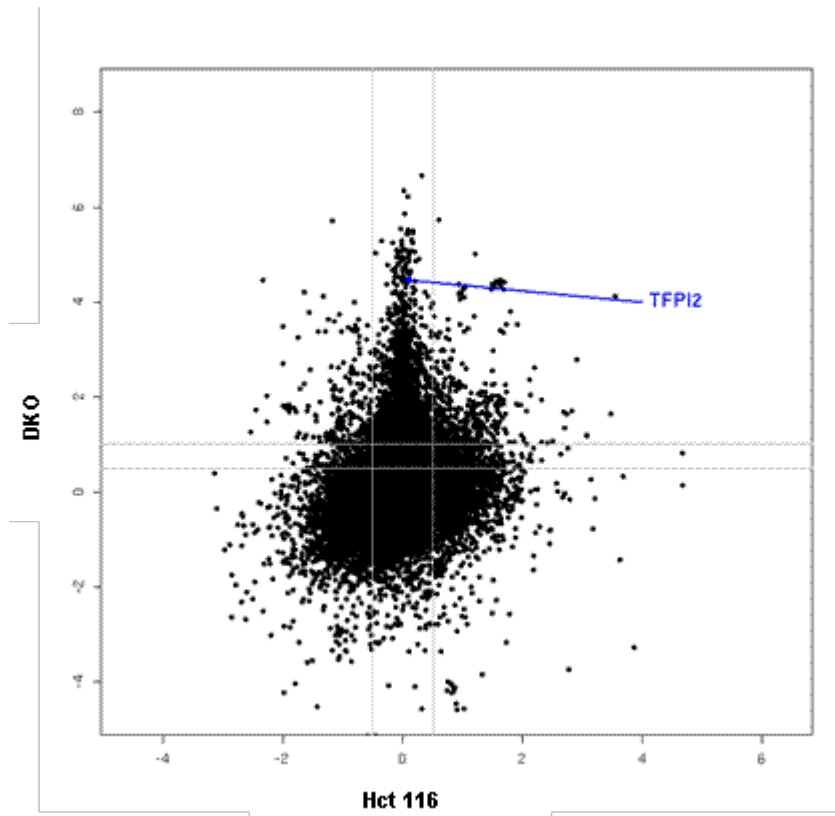
Supplementary Figure Legends:

Supplementary Figure 1: Epigenetic inactivation of TFPI2 in DKO. Gene expression changes are plotted by fold change (log scale) in DKO (Y-axis) compared to its parental cell line Hct116 (X-axis) treatment. TFPI2 expression increases 4.47 in log₂ ratio (=22.12 fold) in DKO compared to HCT116 Mock. □ indicates the location of TFPI2 in the scatter plot.

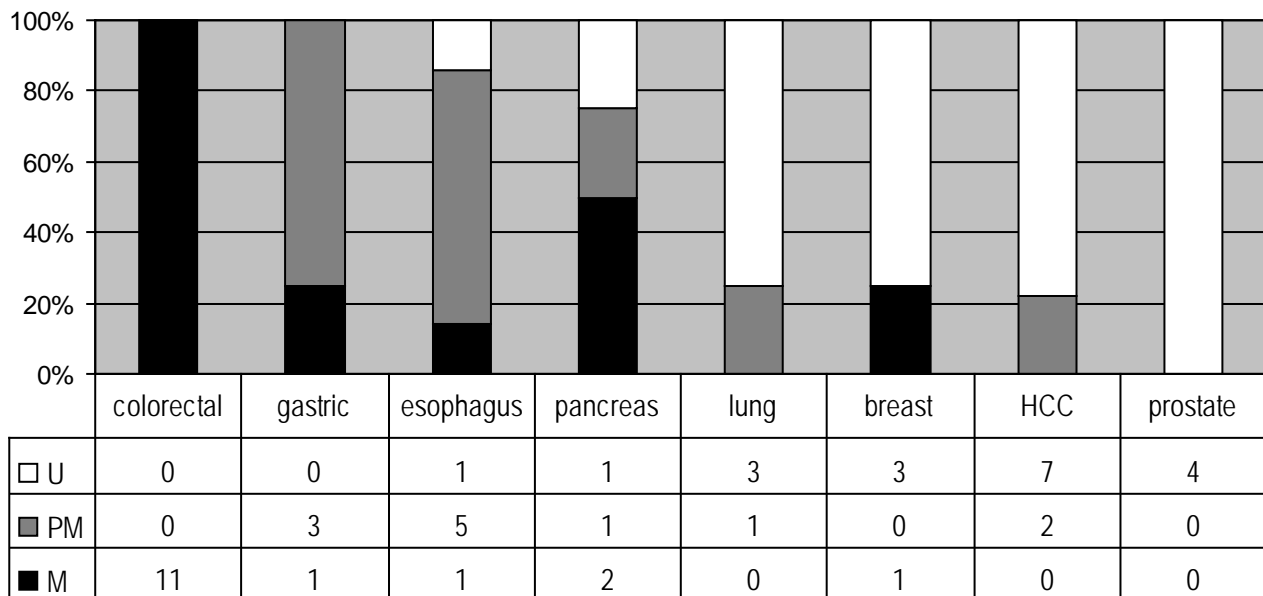
Supplementary Figure 2: Bar graph showing methylation frequency of TFPI2 in various cultured cancer cell lines. □U=unmethylated; PM = partially methylated; □M=methylated

Supplementary Figure 3: Line diagram (A) and bar graph (B) showing cell growth in mock, empty vector and *TFPI2* transfected HCT116 cells. There is no difference in cell survival between mock and empty vector transfected cells. *TFPI2* transfected cells show a 20% decrease in cell survival.

Supplementary Figure 4: Graphic representation of overlap between hypermethylation of HCT116 as determined by Schuebel *et. al.* (10) with PcG-targeted genes according to Bracken *et. al.* (22) in gene pool represented on Agilent 4x44k array platform.

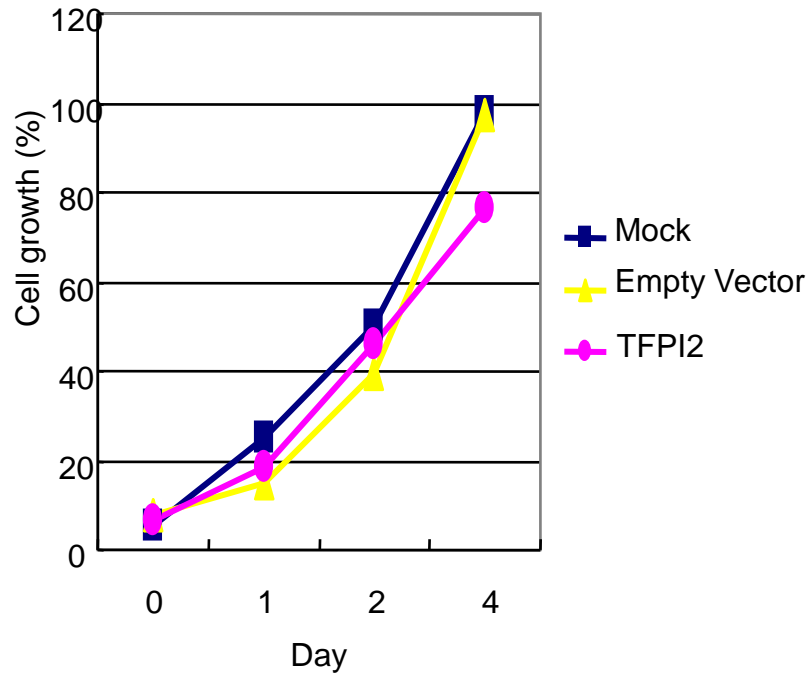


Supplementary Figure 1
Glockner et al.



Supplementary Figure 2
Glockner et al.

3A



3B

