Immunogenicity of SEREX-identified antigens and disease outcome in pancreatic cancer

Cancer Immunology, Immunotherapy

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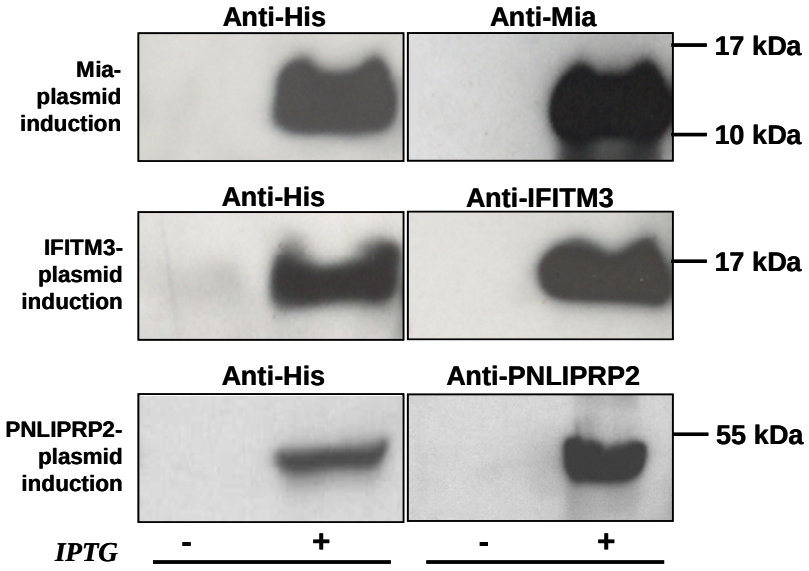
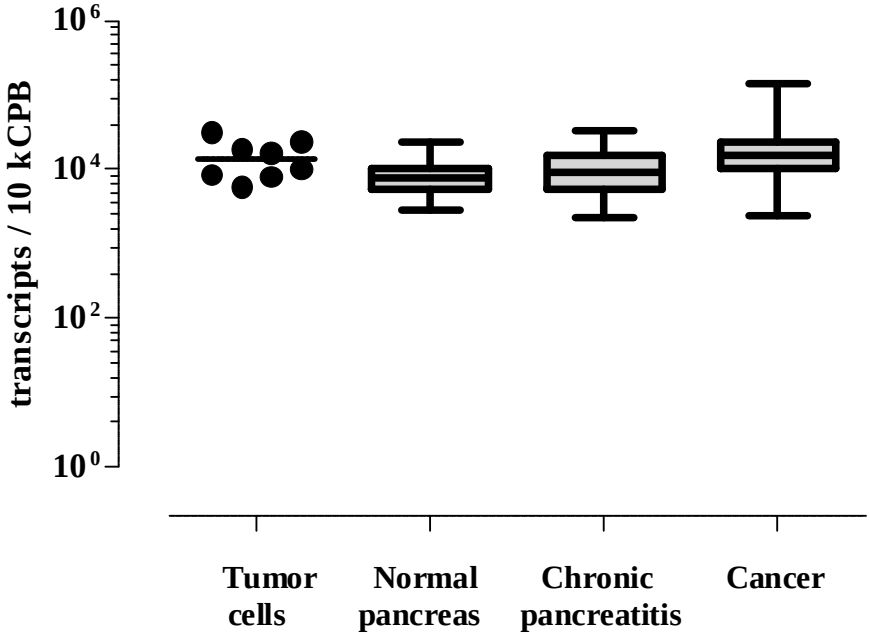
Supplementary material



Supplementary Figure 1: Western blot analysis of induced Mia-, IFITM3- and PNLIPRP2-His-tagged proteins. E. coli M15 bacteria were transformed with a pQE30-His-tag plasmids. Bacteria were lysed in urea buffer, sonicated and the lysates were loaded onto SDS-gels. The IPTG-induced proteins were detected by anti-His (Qiagen), anti-Mia (Santa Cruz), anti-IFITM3 (Abnova) or anti-PNLIPRP2 (Abcam) antibodies. After addition of HRP-conjugated secondary antibodies the signals were visualized by the ECL method. The purity was >80%, determined by SDS-PAGE.



Supplementary Figure 2. QRT-PCR of IFITM3 mRNA expression. QRT-PCR showed an ubiquitous mRNA expression of IFITM3 in cultured pancreatic tumor cells, normal and inflammatory pancreas tissues and cancerous pancreatic lesions.

Supplementary Table 1. Sequences of primers used for RT-PCR

Gene	Primer
ANXA2	FOR: 5'-gattttgggcctattgaagacacc-3', REV: 5'-tacagccgatcagcaaaatacagg-3'
ATP6V0E1	FOR: 5'-tctggggcttcgtcggcttct-3', REV: 5'-atttttcaattgcggtccaaagag-3'
ß-Actin	FOR: 5´-ggcatcgtgatggactccg-3´, REV: 5´-gctggaaggtggacagcga-3´
COX3	FOR: 5'-tggcgcgatgtaacacgagaaag-3', REV:5'-tggcggatgaagcagatagtgagg-3'
DYNC1I2	FOR: 5'-ggctcctaaaccacctattga-3', REV: 5'-gttctttgcactggagttcttttattg-3'
GNB2L1	FOR: 5'-gatctcacaacgggcaccaccac-3', REV: 5'-tgaagcacagggcgttgatga-3'
HDGFRP3	FOR: 5'-gatgcgggcaacgacacaagaaac-3', REV: 5'-gtggggtcaaggcaggaaaacgat-3'
IFITM3	FOR: 5'-ggccccacaaccctgctc-3', REV: 5'-gtcacgtcgccaaccatcttc-3'
KALRN	FOR: 5'-acccgcaaagatgtggctgtgaaa-3', REV: 5'-ggagacggggatgccttgaatgac-3'
KEL	FOR: 5'-tggccccctgcaccgacttcttc-3', REV: 5'-ccctgtgcccgccgctgctc-3'
LUC7L	FOR: 5'-gctgcgtgtctgcgaggtctgttc-3', REV: 5'-ctcgccggcctccttctcttctga-3'
MIA	FOR: 5'-atggcccggtccctggtgtg-3', REV: 5'-acttggccccggtgaatggtc-3'
PNLIPRP2	FOR: 5'-ggcacggaaccagacaccattga-3', REV:5'-tccgggcatttcctttcctccatt-3'
POSTN	FOR: 5'-ttgccctggttatatgagaatgga-3', REV: 5'-tgggagcaaagagtgtgaagtgac-3'
RPL13	FOR: 5'-ccagccggaatggcatggtcttgaagc-3', REV: 5'-ccttggctctttttgcccgtatgccgaa-3'
S100A6	FOR: 5'-ccggcagggagggtgacaag-3', REV: 5'-aggcccccaggaaggtgacatact-3'
SMARCA5	FOR: 5'-acgcgtgctggtggtcttg-3', REV: 5'-cggcatagttggcttttctttctc-3'
TAGLN	FOR: 5'-gagccgcgaagtgcagtccaaaat-3', REV: 5'-tgatctgccgaggtcgtccgtagc-3'
TUBA6	FOR: 5'-acccccgcatccacttccctctg-3', REV: 5'-actcgccttcctccatcccctcac-3'

Supplementary Table 2. mRNA expression of SEREX-identified genes in human tissues (RT-PCR)

Gene \ Tissue	Pancreas		Other Tissues																						
	Cancer Pancreas	Donor Pancreas	Average level	Extrapancreatic expression [%]	No. of tissues (out of n=21)	Bone Marrow	Brain	Colon	Heart	Kidney	Liver	Lung	Mammary Gland	Melanocytes	Muscle Skeletal	Ovary	Placenta	Prostate	Skin	Small Intestine	Spleen	Stomach	Testis	Thymus
ANXA2	++	++	++	100	21	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
ATP6V0E1	+	+	+	100	21	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
COX3	++	++	++	100	21	++	++	++	++	++	++	++	+	++	++	+	++	+	++	+	+	+	+	+
DYNC1I2	++	+	+	100	21	+	++	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
GNB2L1	++	++	++	100	21	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
HDGFRP3	++	+	+	100	21	+	++	+	++	+	+	+	+	+	+	+	+	+	+	+	+	+	++	+
IFITM3	+	+	+	100	21	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
KALRN	++	+	+	95	20	+	++	+	++	+	++	++	++	-	++	+	+	+	+	+	++	++	++	+
KEL	++	-	+	52	11	++	+	+	-	-	-	+	+	+	-	-	+	+	-	-	+	-	++	+
LUC7L	++	+	+	95	20	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
MIA	**+**	**-**	**+**	**43**	**9**	**-**	**-**	**-**	**-**	**-**	**-**	**+**	**++**	**+**	**-**	**-**	**-**	**+**	**-**	**+**	**-**	**+**	**+**	**+**
PNLIPRP2	**+**	**++**	**+**	**14**	**3**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**+**	**+**	**-**	**+**	**-**
POSTN	++	+	+	80	17	-	+	+	+	+	+	+	+	-	+	+	+	+	++	-	-	+	+	+
RPL13	+	+	+	100	21	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
S100A6	+	+	+	95	20	+	+	++	++	++	+	++	+	+	+	++	+	+	+	+	+	+	-	+
SMARCA	++	+	+	100	21	+	+	+	+	+	+	+	+	++	+	+	+	+	+	+	+	+	+	+
TAGLN	++	+	+	100	21	+	+	++	++	+	+	+	+	+	+	+	+	+	++	+	+	+	+	+
TUBA6	++	+	+	95	20	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+