

Supplementary Online Content

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This supplementary material has been provided by the authors to give readers additional information about their work.

eMethods. ONLINE METHODS

DNA extraction, quality control and processing (European dataset). Genomic DNA was extracted from blood samples or normal pathologically confirmed tumour-free tissue adjacent to the resection specimen (fresh-frozen or paraffin-embedded sections) using the blackPREP FFPE DNA Kit (845-BP-0020050, analytikjena, Jena, Germany) at AJ Innuscreen (Berlin, Germany). After extraction, the DNA concentration was measured using the pico-green method, and the Illumina FFPE QC Kit (WG-321-1001, Illumina Inc, San Diego, CA, USA) was utilized to evaluate the quality of prospective DNA samples. According to the specifications of the manufacturer, extracted FFPE samples that passed the quality control test were further processed with the Infinium HD FFPE Restore Kit (WG-321-1002, Illumina Inc, San Diego, CA, USA).

Genotyping (European dataset). The genotypes of 2.5M SNPs were determined using the Infinium Omni2.5-8 Kit (20001112- 20001114, Illumina Inc, San Diego, CA, USA) according to the manufacturer's instructions at the Helmholtz Zentrum in Munich, Germany. To ensure a high genotyping quality, only samples with high-confidence genotype calls (SNP call-rate of at least 90%) that did not show extreme departures from Hardy-Weinberg proportions (p-value \geq 0.0001, Hardy-Weinberg Equilibrium test) were included.

TCGA genotype data and imputation. TCGA provides genotype data for matched normal tissue using the Affymetrix Genome-Wide Human SNP Array 6.0 with 906,000 probes. We performed a standard quality control step by considering only genotype calls with at least 95% confidence as reported by the Birdseed genotyping algorithm and SNPs that did not show extreme departures from Hardy-Weinberg proportions (p-value \geq 0.0001). Because genotypes in the European discovery dataset were performed on a different platform, imputation was conducted to generate a common dataset from both cohorts using LDlink¹⁷ (<https://analysistools.nci.nih.gov/LDlink/?tab=help#LDproxy>; National Cancer Institute, NCI) with $r^2 \geq 0.8$ based on the European reference haplotype from Phase 3 (Version 5) of the 1000 Genomes Project¹⁸.

Histopathological protocol. All resected specimens were evaluated uniformly across centres after a standardized protocol for pathologic evaluation in accordance with international norms and regularly updated national guidelines¹⁹⁻²² (<https://www.leitlinienprogramm-onkologie.de/english-language/>). In brief, the pancreatic head, bile duct, stomach, duodenum transection margins, the medial and uncinate (including both superior mesenteric artery and vein) margins, and the circumferential anterior and posterior border were inked. After taking the margin of the bile duct, stomach, duodenum and the pancreatic neck transection margin, the specimen were cut in 0.3 to 0.5 cm slices in an axial plane perpendicular to the duodenal axis for evaluation of circumferential and soft tissue margins. The margin status was reported separately for the pancreatic neck, bile duct, stomach or duodenum transection margin, and combined for the other margins. A margin was reported as positive (R1) only if tumour cells were present at the surface, i.e. if the clearance equalled 0 mm (R0 narrow: \leq 1mm tumour free margin; R0 wide: $>$ 1mm). We provide additional information about the pathological protocol in the Online Methods of the revised manuscript. This can be found on page 7 of the Results section.

Table 1. Cox survival analysis of top SNPs with a cut-off p-value of p<0.001 in the European discovery and the TCGA validation PDAC cohorts (adjusted for AJCC stage and R-status). For each SNP, we report the rsid, minimal allele frequencies (MAFs), the alleles tested as reported in the Illumina HumanOmni 2.5M array annotation. The following models were tested for survival: (i) AA vs AB vs BB (homozygous genotypes being used as baseline), (ii) AA/AB vs BB and (iii) AA vs AB/BB. We report the estimated differences in tumor-specific survival and the relative p-values. Finally, we report the results for each SNP that is represented either directly or by a proxy SNP with a cut-off rsquare value of r²>0.8 on the TCGA Affymetrix Genome-Wide Human SNP Array 6.0 with the nominal p-values as well as the q-values after correction for multiple testing. SNPs rs684559 and rs353630 are highlighted in red font. The only non-coding missense SNPs (rs1047978) identified in the European discovery cohort is highlighted in brown font.

SNP	MAF*	Allele A	Allele B	Significant models**	Hazard ratio(s)	P-value(s)	SNP	rsquare	Allele A	Allele B	Hazard ratio(s)	P-value(s)	Q-value(s)	Significant models***
rs75961101	0.4141	T	C	AA vs BX; AA vs AB; AA vs BB	0.3154; 0.3342; 0.2981	0.0000133; 9.29e-05; 1.68e-05	N/A							
rs10416513	0.3232	A	G	AA vs BX; AA vs AB; AA vs BB	0.2941; 0.2995; 0.2894	0.0000249; 0.000109; 5.44e-05	N/A							
rs2257906	0.3384	T	C	AX vs BB; AA vs AB	0.4507; 0.4476	0.000031; 4.94e-05	rs2824493	1	A	G	0.8659; 0.5919	0.5842; 0.1991	0.907; 0.4692	
rs1997702	0.4596	T	C	AA vs BX; AA vs AB	0.4116; 0.3910	0.0000387; 5.1e-05	rs9505573	0.9601	A	G	0.8253; 0.6456	0.5141; 0.1552	0.907; 0.4692	
rs11155639	0.4899	T	C	AX vs BB; AA vs AB	0.4554; 0.4646	0.0000413; 0.000169	rs1446556	1	A	C	0.9131; 0.7402	0.74211; 0.4563	0.907; 0.7096	
rs2440	0.3737	A	G	AA vs BX; AA vs AB; AA vs BB	0.3558; 0.3360; 0.3864	0.0000414; 3.94e-05; 0.000567	rs207942	0.8269	C	G	0.9498; 0.7986; 0.9224	0.8852; 0.4273; 0.8361	0.9896; 0.7096; 0.878	
rs9857914	0.2828	A	G	AA vs BX; AA vs AB; AA vs BB	0.2931; 0.3172; 0.2802	0.0000505; 0.000419; 4.35e-05	rs3732477	1	A	G	0.9623; 1.1481; 0.7407	0.9337; 0.767; 0.538	0.9896; 0.8692; 0.8005	
rs1047978	0.3384	G	C	AX vs BB; AA vs AB	2.1183; 2.1446	0.0000863; 0.000117	rs2824493	1	A	G	0.8659; 0.5919	0.5842; 0.1991	0.907; 0.4692	
rs3786721	0.4596	C	T	AA vs BX; AA vs AB	0.3892; 0.3729	0.0000932; 0.000135	rs3786721	1	C	T	0.8227; 0.6844	0.5487; 0.20755	0.907; 0.4692	
rs2834493	0.4293	T	C	AA vs BX; AA vs AB	0.3957; 0.3732	0.0000947; 9.43e-05	N/A							
rs718607	0.4242	A	G	AX vs BB; AA vs AB; AA vs BB	2.1985; 2.0244; 2.9255	0.000111; 0.000808; 5.94e-05	rs718607	1	A	G	1.1986; 1.2535; 1.1047	0.5418; 0.4784; 0.7878	0.907; 0.7096; 0.8753	
rs2658554	0.4798	T	C	AX vs BB; AA vs AB	0.4677; 0.3807	0.000113; 0.000241	rs2658554	1	C	T	0.5498; 0.6808	0.0388; 0.2863	0.4271; 0.8005	
rs2033724	0.3586	T	C	AX vs BB; AA vs AB	0.4824; 0.4617	0.000124; 0.000105	rs2033727	0.8155	C	G	0.7264; 1.0585	0.2131; 0.9032	0.8787; 0.9499	
rs684559	0.3788	A	G	AA vs BX; AA vs AB	0.4115; 0.3975	0.000175; 0.000289	rs684559	1	A	G	0.2705; 0.2746	0.0008; 0.00159	0.0257; 0.0270	AA vs BX; AA vs AB
rs229135	0.4242	T	C	AX vs BB; AA vs AB	2.241; 2.2961	0.000181; 0.000198	rs229136	0.9191	C	T	0.8791; 0.8783	0.6592; 0.6753	0.907; 0.82	
rs10172861	0.4091	T	C	AX vs BB; AA vs AB	2.5299; 2.5455	0.000185; 0.000248	rs2121355	0.8398	A	T	1.372; 1.5041	0.272; 0.2208	0.8977; 0.4692	
rs6496463	0.4646	T	C	AA vs BX; AA vs AB	0.4279; 0.4394	0.000208; 0.000587	N/A							
rs2981485	0.4444	C	T	AA vs BX; AA vs AB	0.4471; 0.4052	0.000214; 0.000111	rs2981485	1	C	T	0.5683; 0.5176	0.0609; 0.0453	0.5027; 0.2567	
rs13418488	0.4899	T	C	AA vs BX; AA vs BB	0.4267; 0.3738	0.000253; 0.000415	N/A							
rs561165	0.4293	A	G	AX vs BB; AA vs AB	0.4975; 0.4465	0.000273; 7.98e-05	rs2039589	1	C	G	1.3894; 1.2240	0.2397; 0.5009	0.8787; 0.7096	
rs4366492	0.4848	T	C	AA vs BX; AA vs BB	2.2271; 2.4833	0.000338; 0.000293	rs1079109	1	G	T	1.2204; 1.2264	0.5855; 0.6332	0.907; 0.8005	
rs6517234	0.3535	A	G	AA vs BX; AA vs BB	0.3961; 0.3636	0.000339; 0.000306	N/A							
rs349030	0.4394	A	G	AA vs BX; AA vs AB; AA vs BB	2.3578; 2.2936; 2.4817	0.000344; 0.000936; 0.000920	N/A							
rs946055	0.4141	A	G	AX vs BB	1.935	0.000366	N/A							
rs2720117	0.2929	A	G	AX vs BB	0.527	0.000396	rs2600737	1	G	T	0.8521	0.5425	0.907	
rs2600726	0.3131	A	G	AX vs BB	1.892	0.000487	rs2600731	0.9769	A	C	1.0987	0.7999	0.9427	
rs1879543	0.4899	T	C	AX vs BB	0.4973	0.000494	rs4669895	0.98	A	C	1.0105	0.9718	0.9896	
rs4927129	0.3485	T	C	AA vs BX; AA vs AB	0.3759; 0.3700	0.000515; 0.000699	N/A							
rs1997401	0.4293	A	G	AX vs BB; AA vs BB	0.5004; 0.3603	0.000533; 0.000346	rs1997401	1	A	G	0.8848; 0.8386	0.6756; 0.6317	0.907; 0.8005	
rs2670217	0.3939	A	G	AA vs BX; AA vs AB	0.3664; 0.3543	0.000539; 0.000704	rs2559537	1	A	G	1.274; 1.2632	0.6088; 0.6337	0.907; 0.820	
rs4130820	0.4899	A	G	AX vs BB	2.0291	0.000541	rs4130820	1	A	G	1.6977	0.0888	0.5858	
rs12019147	0.4545	A	G	AA vs BX	0.4801	0.00056	rs1201914	1	A	G	1.3465	0.4008	0.907	
rs33412	0.4141	A	G	AA vs BX; AA vs AB; AB vs BB	0.3655; 0.2599; 0.5095	0.000618; 1.57e-05; 0.000605	rs33412	1	A	G	1.0044; 0.9780; 0.9354	0.9896; 0.9499; 0.8187	0.9896; 0.9499; 0.8187	
rs13432615	0.3131	A	G	AA vs BX; AA vs BB	0.4233; 0.3836	0.000619; 0.000336	rs1343261	1	A	G	0.7961; 0.4993	0.5706; 0.11620	0.907; 0.8005	
rs962066	0.399	T	G	AA vs BX; AA vs BB	0.3773; 0.3448	0.000631; 0.00049	5	1	A	G	0.7961; 0.4993	0.5706; 0.11620	0.907; 0.8005	
rs56319045	0.4848	A	G	AA vs BX; AA vs BB	0.4466; 0.3514	0.000636; 9.82e-05	rs1004512	0.8035	A	C	1.5116; 1.5550	0.3942; 0.382	0.907; 0.8005	
rs6020324	0.3636	A	G	AX vs BB	0.5365	0.000646	6	1	A	C	1.5116; 1.5550	0.3942; 0.382	0.907; 0.8005	
rs7598275	0.2626	A	G	AA vs BX;	0.4283	0.000667	rs4591400	1	A	G	1.3387	0.7384	0.907	
rs803742	0.4192	A	G	AA vs BX; AA vs BB	0.4636; 0.4219	0.000731; 0.000894	rs7598275	1	A	G	1.0072	0.987	0.9896	
rs12909013	0.3485	A	G	AX vs BB	0.5155	0.000735	N/A							
rs246770	0.3283	A	G	AA vs BX; AA vs AB	0.4066; 0.3725	0.000743; 0.00044	N/A							
rs10793269	0.3788	C	T	AA vs BX; AA vs BB	0.4544; 0.3775	0.000807; 0.000261	rs1079326	1	C	T	0.5962; 0.5490	0.1337; 0.1098	0.7354; 0.8005	
rs4757940	0.4747	T	C	AX vs BB	0.5339	0.000842	9	1	C	T	0.5962; 0.5490	0.1337; 0.1098	0.7354; 0.8005	
rs353630	0.2727	A	G	AA vs BX; AA vs AB	0.3814; 0.3491	0.000876; 0.000847	rs353630	0.8834	A	G	1.164	0.6027	0.907	
rs935412	0.3687	T	C	AX vs BB	0.4931	0.000884	4	1	A	G	1.164	0.6027	0.907	
rs6664	0.3232	C	T	AA vs BX	0.3963	0.000951	rs353630	0.8834	A	G	0.1908; 0.2270	0.0027; 0.00981	0.0452; 0.083	AA vs BX
rs2160046	0.404	A	G	AX vs BB	0.5421	0.000959	N/A							
rs1332182	0.4848	A	C	AX vs BB	0.5346	0.000981	rs6664	1	C	T	1.1952	0.6405	0.907	
rs3107	0.3182	T	G	AX vs BB	0.5396	0.000982	rs2160046	1	A	G	1.4417	0.1604	0.7564	
rs879742	0.3333	T	G	AA vs BB; AB vs BB	0.2561; 0.2033	0.0000989; 0.00000747	rs1332182	1	A	C	0.91	0.7377	0.907	

rs7258937	0.4596	T	C	AA vs BB	0.2492	0.0000223	N/A										
rs2706759	0.298	A	G	AA vs BB	2.945	0.0000684	N/A										
rs10955174	0.4394	T	C	AA vs BB; AB vs BB	2.5872; 2.006	0.000134; 0.000948	rs4388428	0.9797	C	T	1.3105; 0.8218	0.4643; 0.5290	0.8005; 0.7053				
rs1711944	0.3737	C	G	AA vs BB	2.8869	0.000164	rs1711944	1	C	G	0.5569	0.1766	0.8005				
rs7409	0.257	T	C	AA vs BB	2.9092	0.000218	rs2598446	0.9741	C	T	1.376	0.4193	0.8005				
rs921383	0.5	A	G	AA vs BB	0.3975	0.000244	rs7936118	0.8828	C	T	1.1132	0.7625	0.8005				
rs10821968	0.399	T	C	AA vs BB	0.4018	0.000332	rs1082196	1	C	T	1.4828	0.3589	0.8753				
rs379977	0.4343	A	C	AA vs BB	2.829	0.000348	rs1164118	1	G	T	1.2829	0.4846	0.8005				
rs11641184	0.4242	A	C	AA vs BB	0.3904	0.000354	rs2345804	0.98	C	T	0.7361	0.4286	0.8005				
rs4261436	0.4444	T	C	AA vs BB	0.4126	0.000402	N/A										
rs971530	0.4848	A	C	AA vs BB	2.6256	0.000478	N/A										
rs1972455	0.3788	A	G	AA vs BB	2.5224	0.000483	N/A										
rs2426228	0.4646	A	C	AA vs BB	2.5453	0.000601	rs6569937	1	A	G	1.1914	0.6404	0.8005				
rs6569937	0.4899	A	G	AA vs BB	2.6344	0.000622	N/A										
rs4968568	0.4545	A	G	AA vs BB	2.7318	0.000659	N/A										
rs1134767	0.4596	A	G	AA vs BB	0.3742	0.000726	rs2518628	1	A	C	1.267	0.5503	0.8005				
rs7684068	0.4646	T	C	AA vs BB	2.373	0.000746	rs1073942	1	A	T	1.0591; 1.322	0.878; 0.336	0.878; 0.672				
rs1075372	0.4596	A	G	AA vs BB; AB vs BB	2.7731; 2.2704	0.000749; 0.000319	rs1422961	1	A	T	0.5284; 0.6608	0.14619; 0.15465	0.8005; 0.6186				
rs9905663	0.4747	A	C	AA vs BB	2.3365	0.000785	N/A										
rs6892869	0.4091	T	C	AA vs BB; AB vs BB	2.3225; 2.7160	0.000889; 2.56e-05	N/A										
rs910652	0.3687	T	C	AA vs BB	2.7313	0.000899	N/A										
rs7167269	0.4697	A	G	AA vs BB	0.3962	0.000914	N/A										
rs7523821	0.404	A	G	AA vs BB	2.3691	0.000982	N/A										

* based on the European reference haplotype from Phase 3 (Version 5) of the 1000 Genomes Project

** p-value threshold p<0.001

*** q-value threshold q<0.05

Abbr. MAF, minimal allele frequency

eTable 2. Survival time analysis in the European and TCGA datasets based on Kaplan Meier estimates

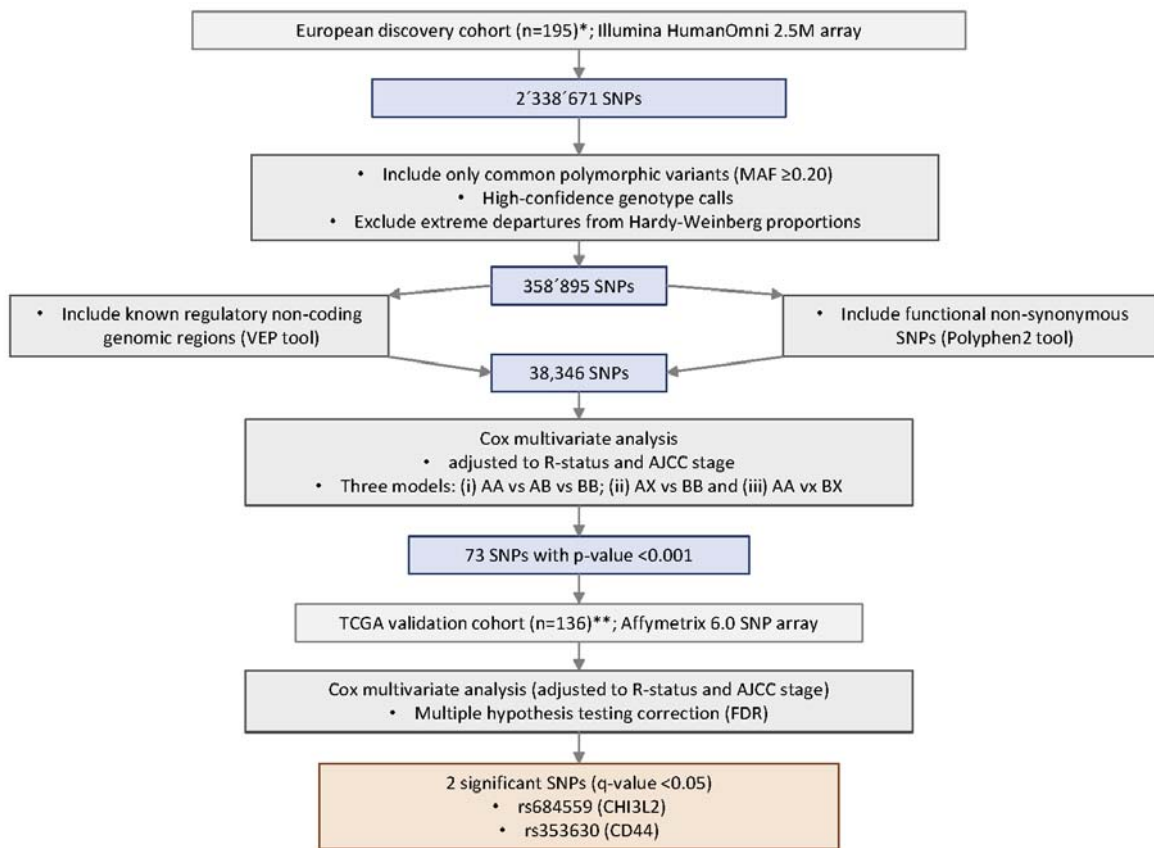
Cohort	SNP	Group	n	Number of tumour-related deaths	Means				Medians				p-value (log-rank)
					Estimate (months)	Std. Error	95% Confidence Interval		Estimate (months)	Std. Error	95% Confidence Interval		
							Lower Bound	Upper Bound			Lower Bound	Upper Bound	
European	rs353630 (CD44)	T/T	20	15	17.7	2.3	13.2	22.2	19.0	6.3	6.7	31.3	0.03
		C/T	67	42	42.0	6.0	30.4	53.7	27.7	5.0	17.9	37.5	
		C/C	105	75	38.6	4.3	30.1	47.0	20.0	2.7	14.6	25.4	
		C-allele	172	117	40.9	3.7	33.6	48.1	23.0	3.4	16.4	29.6	
	rs684559 (CHI3L2)	A/A	30	24	18.1	2.7	12.9	23.3	15.0	1.2	12.6	17.4	0.001
		A/G	84	53	47.0	5.7	35.8	58.3	29.0	4.5	20.1	37.9	
		G/G	79	56	34.6	4.6	25.7	43.6	20.0	3.7	12.7	27.3	
Signature*,**	Risk-indicating*	47	36	18.1	2.1	14.1	22.1	15.0	1.5	12.0	18.0	0.00003	
	Protective**	148	99	43.7	4.1	35.7	51.6	29.0	3.4	22.3	35.7		
TCGA	rs353630 (CD44)	T/T	6	4	11.4	1.8	7.7	15.0	11.0	1.5	8.1	13.9	0.01
		C/T	56	26	23.0	2.4	18.2	27.8	19.8	0.7	18.5	21.1	
		C/C	74	35	31.2	3.6	24.1	38.2	21.4	1.4	18.7	24.2	
		C-allele	130	61	28.5	2.7	23.3	33.8	20.4	0.9	18.5	22.2	
	rs684559 (CHI3L2)	A/A	18	11	13.7	2.1	9.6	17.8	13.0	1.9	9.2	16.7	0.006
		A/G	70	32	29.9	3.8	22.4	37.4	20.2	1.8	16.6	23.8	
		G/G	48	22	27.4	3.5	20.5	34.3	20.8	0.8	19.2	22.5	
Signature*,**	Risk-indicating*	118	54	29.4	2.8	23.9	35.0	20.8	1.3	18.4	23.3	0.001	
	Protective**	23	15	13.0	1.6	9.8	16.2	11.0	1.6	7.8	14.2		0.00001
			113	50	30.3	2.9	24.5	36.0	21.4	1.2	19.1	23.8	

* rs353630 T/T and/or rs684559 A/A genotype
 ** rs353630 C-allele and rs684559 G-allele

eTable 3. Hazard Ratios

Term	HR	95% Confidence Interval low	95% Confidence Interval high	p-value
AJCC - 1	0.7147	0.335	1.525	0.385
AJCC - 3	1.6626	0.7844	3.524	0.185
AJCC - 4	1.7593	1.0405	2.975	0.035
R-status - 1	1.4108	1.0377	1.918	0.028
R-status - 2	2.4212	1.2814	4.575	0.006

eFigure 1. Workflow diagram of the survival screen.

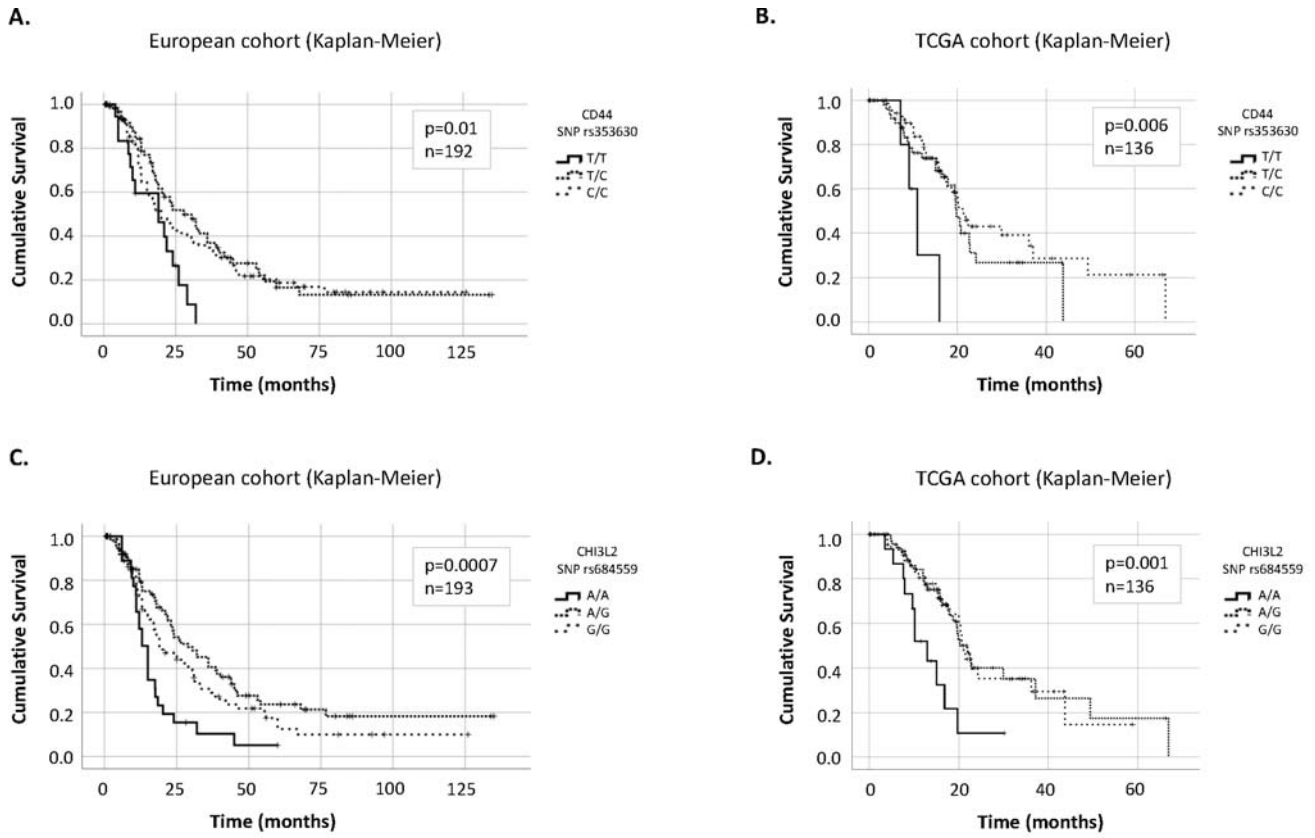


* Cantonal Hospital Winterthur, University Hospital Zurich, University Clinic Ulm

** TCGA, The Cancer Genome Atlas

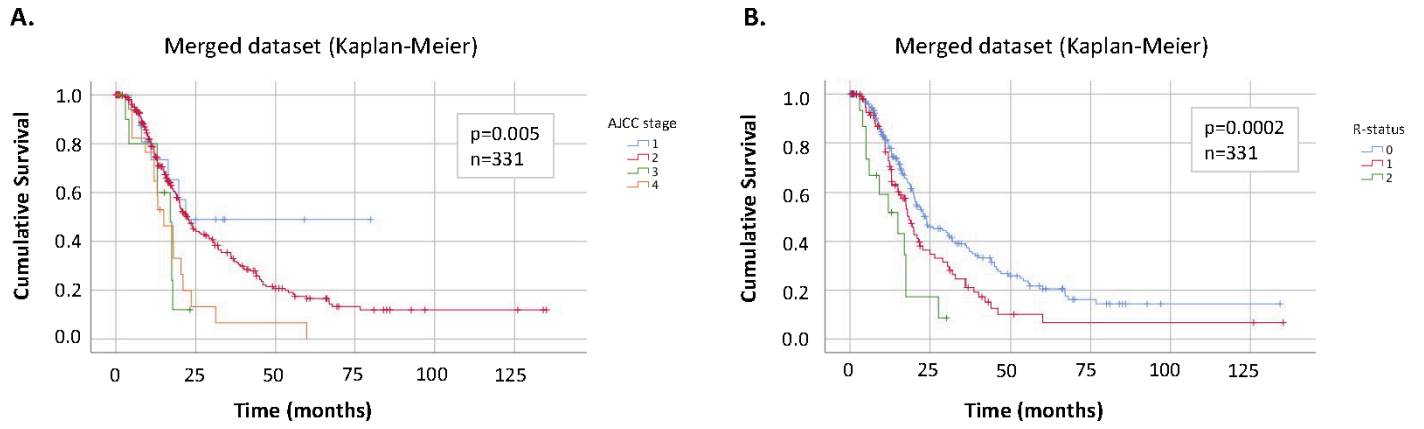
*** Resectable stages of PDAC; Consecutive patients (full clinical follow-up and non-tumour tissue available for DNA analysis)

eFigure 2. Survival analysis of tumour-specific survival after resection of PDAC. (A) and (B) Kaplan-Meier survival curves for SNP rs353630 in the European and TCGA cohort, respectively. (C) and (D) Kaplan-Meier survival curves for SNP rs684559 in the European and TCGA cohort, respectively.



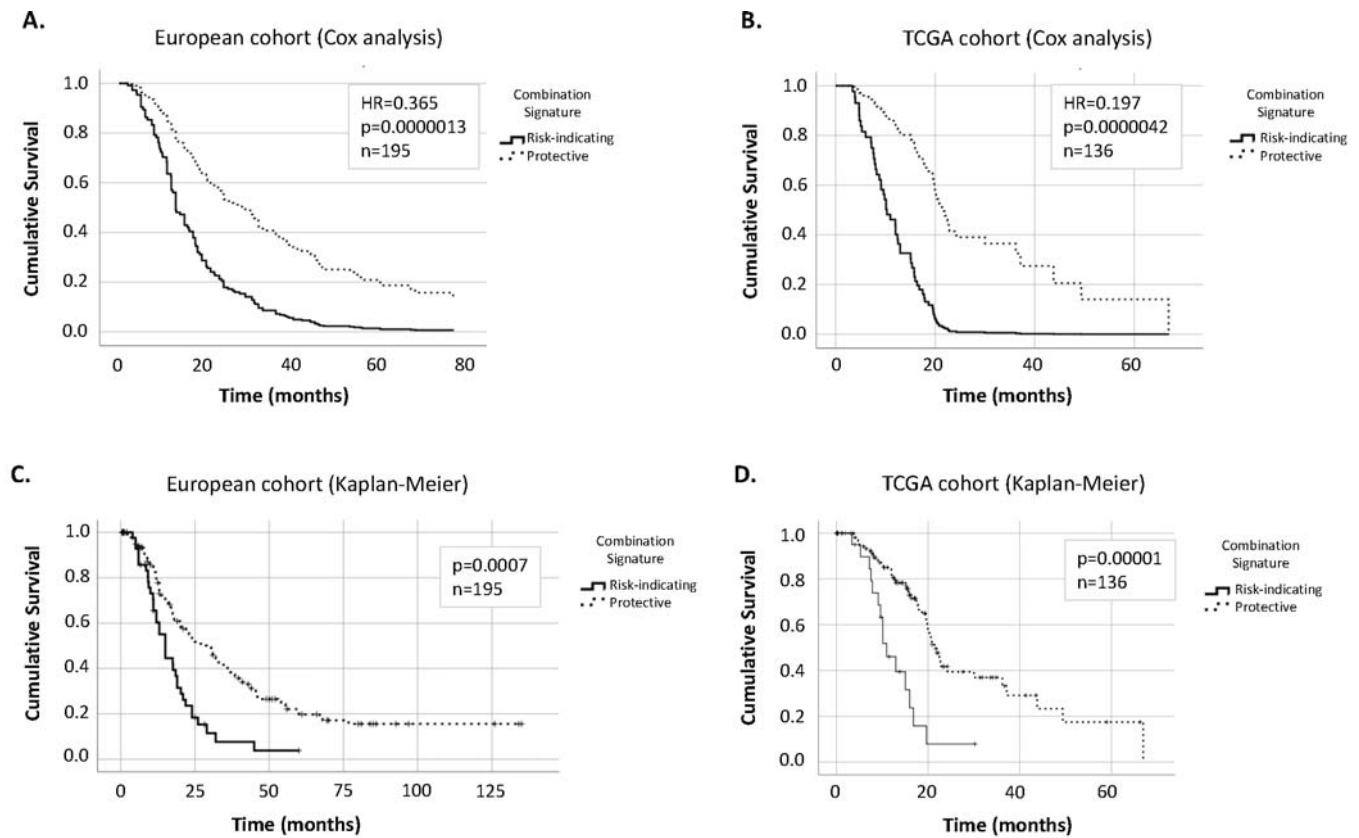
The p-values were determined with the log-rank test.

eFigure 3. Kaplan-Meier survival curves for the two independent prognostic factors AJCC stage (A) and resection margin status (B) in the merged dataset.



The p-values were determined with the log-rank test.

eFigure 4. Analysis of tumour-related survival for the biomarker signature that combines both SNP loci (rs684559 and rs353630). (A) and (B) Cox multivariate regression survival curves for the combination signature in the European and TCGA cohort, respectively (adjusted to the known independent prognostic factors tumour stage and resection status). (C) and (D) Kaplan-Meier survival curves for the combination signature in the European and TCGA cohort, respectively.



The p-values were determined with the log-rank test.