Supplemental Online Content

Shoucair S, Chen J, Martinson JR, et al. Association of matrix metalloproteinase 7 expression with pathologic response after neoadjuvant treatment in patients with resected pancreatic ductal adenocarcinoma. *JAMA Surg*. Published online May 25, 2022. doi:10.1001/jamasurg.2022.1362

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

eMethods

Study population and tissue specimens

An exploratory cohort of 60 patients who underwent upfront surgical resection (without any neoadjuvant therapy) was investigated to evaluate the association of MMP-7 protein expression in primary tumor surgical specimens with oncologic outcomes. For this purpose, archived surgical resected primary tumor specimen FFPE blocks were obtained and IHC staining for MMP-7 protein expression was performed.

By CAP score criteria, a score of 0 was assigned for pathologic complete response (pCR), score 1 (near-complete PR), score 2 (partial response) and score 3 (poor or no response).

DNA/RNA extraction and sequencing

Bulk FNA specimens were processed for downstream sequencing per the protocol described in our previous study.[19] Briefly, DNA/RNA were extracted using the QIAamp AllPrep DNA/RNA Micro Kit (Cat No: 80284, Qiagen, Dusseldorf, Germany) and then quantified by Qubit DNA/RNA Assay Kit (Thermo Fisher Scientific, Waltham, MA) in Qubit 3.0 Flurometer.

All next-generation sequencing assays were performed by investigators that were blinded to patient information. Bulk FNA samples containing sufficient tumor cells were confirmed by a DNA sequencing with an Ion AmpliSeq Custom Panel (Thermo Fisher) covering *KRAS*, *GNAS*, *TP53*, *SMAD4*, and *CDKN2A*. A targeted RNAseq Custom Panel (**Table S2**) covering *MMP-7* and an additional 471 genes was performed using the Ion GeneStudio™ S5 System (Thermo Fisher Scientific, Waltham, MA), according to the manufacturer's protocols and our previous studies.[19,20]

Immunohistochemistry staining

FFPE Tissue blocks were sectioned into 5 μm thick slides. De-paraffinization with heat and xylene, followed by immersion in alcohol for re-hydration. Endogenous peroxidase activity was suppressed by a solution of 3% hydrogen peroxide in methanol for 30 minutes. Antigen retrieval was performed using ethylenediamine tetraacetic acid (EDTA) antigen retrieval buffer (pH 8.5, 1.0x) microwaved at 750W and 350W for 10 and 15 minutes, respectively. Primary anti-MMP-7 antibody (Cell Signaling Technology Cat# 3801, RRID:AB_2144465) was added (dilution 1:50) and incubated overnight at 4°C. After washing, the slides were treated with biotinylated anti-rabbit secondary antibody for 30 minutes. The slides were finally developed using AEC chromogen substrate kit. The slides were reviewed on a blinded basis by two independent investigators. The categories of immunohistochemical staining were assigned as negative in cases where < 10% of tumor cells were stained and as a positive result when >10% of tumor cells were stained.

eTable 1. Demographics and tumor characteristics of FNA specimens in initial (discovery) cohort prior to neoadjuvant therapy (N = 23).

Variables, N (%)	Discovery cohort (N = 23)	Validation cohort (N = 80)	*P*
Age (years) Median (IQR)	64.5 (59, 69)	66.3 (58.6, 72.9)	
Sex			0.52
Female	11 (47.9)	40 (50.0)	
Male	12 (52.1)	40 (50.0)	
Race (White)	20 (86.9%)	71 (88.7)	0.45
Baseline Ca19-9, Median (IQR)	315.8 (166.4, 1383.3)	182.2 (20.5, 535.2)	0.32
Type of neoadjuvant			0.12
5-FU based	10 (43.5)	57 (71.3)	
Gem-based	5 (21.7)	17 (21.3)	
Other	8 (34.8)	6 (7.5)	
Chemo-Radiation	13 (56.5)	59 (73.8)	0.13
Chemotherapy alone	10 (43.5)	21 (26.3)	
pT-stage ≥ T3	8 (34.8)	23 (28.7)	0.38
pN-stage ≥ N1	13 (56.5)	31 (38.8)	0.10
Grade of differentiation			0.93
Well	2 (8.6)	6 (7.5)	
Moderate	15 (65.2)	50 (62.5)	
Poor	6 (26.2)	24 (30)	
Lymphovascular Invasion	9 (39.1)	20 (25)	0.14
Perineural Invasion	17 (73.9)	44 (55)	0.08
Margin Class (R0)	19 (82.6)	71 (88.7)	0.32
Pathologic Response (CAP Score)			**0.008**
0	0(0)	8 (10)	
1	3 (13)	34 (42.5)	
2	5 (21.7)	13 (16.2)	
3	15 (65.2)	25 (31.2)	
Resectability			0.24
Resectable	9 (39.1)	21 (26.3)	
Borderline resectable	5 (21.7)	32 (40)	
Locally advanced	9 (39.1)	27 (33.8)	

IQR, Interquartile range

eTable 2: Targeted RNA NGS list (N=472).

ID	Target Region Start	Target Region End	AmpliSeqID	Gene	AmpliSeq Version	Workflow
NM_004302	1323	1425	AMPL27122439	ACVR1B	6.0	RNAseq
NM_001616	206	308	AMPL11796878	ACVR2A	6.0	RNAseq
NM_001077401	1349	1457	AMPL3042073	ACVRL1	6.0	RNAseq
NM_001110	466	572	AMPL5356454	ADAM10	6.0	RNAseq
NM_003183	2037	2142	AMPL9492210	ADAM17	6.0	RNAseq
NM_001115	1592	1699	AMPL1610329	ADCY8	6.0	RNAseq
NM_001353	176	286	AMPL9888159	AKR1C1	6.0	RNAseq
NM_205845	463	571	AMPL22530899	AKR1C2	6.0	RNAseq
NM_003739	231	329	AMPL9583583	AKR1C3	6.0	RNAseq
NM_001818	48	154	AMPL7374404	AKR1C4	6.0	RNAseq
NM_001014431	358	467	AMPL2201548	AKT1	6.0	RNAseq
NM_001626	1043	1151	AMPL11229610	AKT2	6.0	RNAseq
NM_004304	3264	3373	AMPL27472922	ALK	6.0	RNAseq
NM_000699	162	262	AMPL5487990	AMY2A	6.0	RNAseq
NM_181861	1015	1117	AMPL21790783	APAF1	6.0	RNAseq
NM_000038	1676	1781	AMPL1716059	APC	6.0	RNAseq
NM_001077628	796	900	AMPL1108511	APH1A	6.0	RNAseq
NM_031301	144	244	AMPL15100366	APH1B	6.0	RNAseq
NM_000044	2927	3032	AMPL1447304	AR	6.0	RNAseq
NM_006015	3744	3851	AMPL28474827	ARID1A	6.0	RNAseq
NM_152641	4844	4944	AMPL34284744	ARID2	6.0	RNAseq
NM_138450	237	345	AMPL34675105	ARL11	6.0	RNAseq
NM_152285	484	589	AMPL35508520	ARRDC1	6.0	RNAseq
NM_000051	5631	5733	AMPL2143252	ATM	6.0	RNAseq
NM_001184	322	429	AMPL6698211	ATR	6.0	RNAseq
NM_003600	1194	1297	AMPL11289894	AURKA	6.0	RNAseq
NM_032989	444	548	AMPL15908493	BAD	6.0	RNAseq
NM_001704	3031	3134	AMPL7540943	BAI3	6.0	RNAseq
NM_004656	756	862	AMPL29370657	BAP1	6.0	RNAseq
NM_000055	108	210	AMPL5334700	BCHE	6.0	RNAseq
NM_000633	154	251	AMPL2837802	BCL2	6.0	RNAseq
NM_138578	223	332	AMPL34227316	BCL2L1	6.0	RNAseq
NM_021946	4299	4401	AMPL36541905	BCORL1	6.0	RNAseq
NM_004333	1411	1520	AMPL28552085	BRAF	6.0	RNAseq
NM_007300	4880	49853	AMPL13649775	BRCA1	6.0	RNAseq

NM_000059	9817	9922	AMPL6315908	BRCA2	6.0	RNAseq
NM_032043	719	823	AMPL14267024	BRIP1	6.0	RNAseq
NM_004336	701	803	AMPL29290680	BUB1	6.0	RNAseq
NM_004725	566	669	AMPL27323746	BUB3	6.0	RNAseq
NM_021098	4018	4125	AMPL35326025	CACNA1H	6.0	RNAseq
NM_032977	396	502	AMPL17568853	CASP10	6.0	RNAseq
NM_032991	679	783	AMPL18385871	CASP3	6.0	RNAseq
NM_001226	673	778	AMPL9136589	CASP6	6.0	RNAseq
NM_033338	644	738	AMPL17730367	CASP7	6.0	RNAseq
NM_001080125	740	836	AMPL5125235	CASP8	6.0	RNAseq
NM_001229	1337	1440	AMPL8355632	CASP9	6.0	RNAseq
NM_001236	470	577	AMPL9279705	CBR3	6.0	RNAseq
NM_003914	1596	1701	AMPL28572122	CCNA1	6.0	RNAseq
NM_031966	1412	1518	AMPL14157888	CCNB1	6.0	RNAseq
NM_053056	351	458	AMPL16186640	CCND1	6.0	RNAseq
NM_001759	844	953	AMPL11854004	CCND2	6.0	RNAseq
NM_001238	1004	1106	AMPL7555808	CCNE1	6.0	RNAseq
NM_001785	279	388	AMPL8437699	CDA	6.0	RNAseq
NM_001789	1201	1304	AMPL7718089	CDC25A	6.0	RNAseq
NM_024529	1364	1468	AMPL36259069	CDC73	6.0	RNAseq
NM_004360	1610	1714	AMPL28185772	CDH1	6.0	RNAseq
NM_001786	241	344	AMPL7721438	CDK1	6.0	RNAseq
NM_001798	930	1038	AMPL11968322	CDK2	6.0	RNAseq
NM_000075	418	528	AMPL4090056	CDK4	6.0	RNAseq
NM_001145306	1261	1365	AMPL5549170	CDK6	6.0	RNAseq
NM_001260	1257	1362	AMPL8999838	CDK8	6.0	RNAseq
NM_078467	421	530	AMPL18336733	CDKN1A	6.0	RNAseq
NM_004064	928	1036	AMPL30170055	CDKN1B	6.0	RNAseq
NM_000077	666	772	AMPL4089136	CDKN2A	6.0	RNAseq
NM_004936	491	594	AMPL18141488	CDKN2B	6.0	RNAseq
NM_078626	514	617	AMPL16833223	CDKN2C	6.0	RNAseq
NM_001807	341	446	AMPL8603667	CEL	6.0	RNAseq
NM_033440	219	326	AMPL18166601	CELA2A	6.0	RNAseq
NM_005747	229	338	AMPL29440374	CELA3A	6.0	RNAseq
NM_007352	452	559	AMPL13122485	CELA3B	6.0	RNAseq
NM_000492	2544	2648	AMPL6465189	CFTR	6.0	RNAseq
NM_001274	1159	1263	AMPL7796632	CHEK1	6.0	RNAseq
NM_001005735	1537	1639	AMPL2657479	CHEK2	6.0	RNAseq
NM_001278	1948	2055	AMPL7797843	CHUK	6.0	RNAseq

NM_004882	555	654	AMPL28739194	CIR1	6.0	RNAseq
NM_007056	337	443	AMPL12698021	CLASRP	6.0	RNAseq
NM_001832	177	283	AMPL10530961	CLPS	6.0	RNAseq
NM_005140	358	463	AMPL27862431	CNGA2	6.0	RNAseq
NM_080629	4681	4789	AMPL17432958	COL11A1	6.0	RNAseq
NM_000093	5251	5355	AMPL2831737	COL5A1	6.0	RNAseq
NM_004369	3277	3384	AMPL27933904	COL6A3	6.0	RNAseq
NM_153264	7936	8038	AMPL17060297	COL6A5	6.0	RNAseq
NM_001102608	5709	5816	AMPL1545666	COL6A6	6.0	RNAseq
NM_001868	1010	1119	AMPL10917633	CPA1	6.0	RNAseq
NM_015692	1861	1973	AMPL33783463	CPAMD8	6.0	RNAseq
NM_001871	1080	1192	AMPL8245762	CPB1	6.0	RNAseq
NM_000757	806	902	AMPL4769976	CSF1	6.0	RNAseq
NM_005211	1961	2067	AMPL27286436	CSF1R	6.0	RNAseq
NM_033225	10174	10280	AMPL16512831	CSMD1	6.0	RNAseq
NM_052896	6282	6388	AMPL15996854	CSMD2	6.0	RNAseq
NM_198123	862	968	AMPL21626780	CSMD3	6.0	RNAseq
NM_001328	923	1032	AMPL7974282	CTBP1	6.0	RNAseq
NM_022802	2746	2849	AMPL34972921	CTBP2	6.0	RNAseq
NM_001906	19	118	AMPL7364175	CTRB1	6.0	RNAseq
NM_001025200	211	315	AMPL964867	CTRB2	6.0	RNAseq
NM_007272	307	411	AMPL12504527	CTRC	6.0	RNAseq
NM_003592	2558	2664	AMPL8209755	CUL1	6.0	RNAseq
NM_000634	56	156	AMPL3400001	CXCR1	6.0	RNAseq
NM_001557	379	486	AMPL11107598	CXCR2	6.0	RNAseq
NM_020311	96	204	AMPL31908292	CXCR7	6.0	RNAseq
NM_148923	239	346	AMPL34651448	CYB5A	6.0	RNAseq
NM_000781	579	686	AMPL3192936	CYP11A1	6.0	RNAseq
NM_000497	591	701	AMPL1428925	CYP11B1	6.0	RNAseq
NM_000498	864	973	AMPL1673129	CYP11B2	6.0	RNAseq
NM_000102	593	694	AMPL1382000	CYP17A1	6.0	RNAseq
NM_000103	1131	1237	AMPL4602847	CYP19A1	6.0	RNAseq
NM_000500	687	788	AMPL2620377	CYP21A2	6.0	RNAseq
NM_000106	1173	1281	AMPL5936620	CYP2D6	6.0	RNAseq
NM_017460	391	491	AMPL31771047	CYP3A4	6.0	RNAseq
NM_022820	1108	1209	AMPL36782782	CYP3A43	6.0	RNAseq
NM_000777	200	291	AMPL3569906	CYP3A5	6.0	RNAseq
NM_000765	1501	1600	AMPL4295408	CYP3A7	6.0	RNAseq
NM_001343	2802	2906	AMPL7260755	DAB2	6.0	RNAseq

NM_000790	899	1001	AMPL3751222	DDC	6.0	RNAseq
NM_004675	217	319	AMPL27553848	DIRAS3	6.0	RNAseq
NM_007337	1415	1522	AMPL12691995	DLEC1	6.0	RNAseq
NM_005618	431	539	AMPL27827828	DLL1	6.0	RNAseq
NM_016941	1678	1787	AMPL32704364	DLL3	6.0	RNAseq
NM_019074	2182	2281	AMPL31396774	DLL4	6.0	RNAseq
NM_003777	4908	5011	AMPL11272646	DNAH11	6.0	RNAseq
NM_001369	287	394	AMPL7777818	DNAH5	6.0	RNAseq
NM_001372	11290	11393	AMPL10593530	DNAH9	6.0	RNAseq
NM_001380	3787	3892	AMPL11742501	DOCK1	6.0	RNAseq
NM_004946	4006	4118	AMPL27535684	DOCK2	6.0	RNAseq
NM_004947	2323	2427	AMPL27417255	DOCK3	6.0	RNAseq
NM_014705	5075	5181	AMPL24723128	DOCK4	6.0	RNAseq
NM_024940	5011	5117	AMPL13845021	DOCK5	6.0	RNAseq
NM_001383	293	400	AMPL9168667	DPH1	6.0	RNAseq
NM_000110	419	519	AMPL2093445	DPYD	6.0	RNAseq
NM_020693	2953	3056	AMPL31003419	DSCAML1	6.0	RNAseq
NM_015548	3170	3272	AMPL31722246	DST	6.0	RNAseq
NM_004416	582	686	AMPL30097098	DTX1	6.0	RNAseq
NM_001102595	1895	2001	AMPL1357097	DTX2	6.0	RNAseq
NM_178502	1056	1163	AMPL15715094	DTX3	6.0	RNAseq
NM_138287	1960	2060	AMPL16254296	DTX3L	6.0	RNAseq
NM_015177	463	573	AMPL25368900	DTX4	6.0	RNAseq
NM_001130987	2983	3087	AMPL2561700	DYSF	6.0	RNAseq
NM_005225	894	1004	AMPL29253382	E2F1	6.0	RNAseq
NM_004091	1191	1296	AMPL30036601	E2F2	6.0	RNAseq
NM_001949	1186	1292	AMPL10340053	E2F3	6.0	RNAseq
NM_001950	148	256	AMPL7531768	E2F4	6.0	RNAseq
NM_001951	627	735	AMPL9976395	E2F5	6.0	RNAseq
NM_198256	601	706	AMPL21241121	E2F6	6.0	RNAseq
NM_203394	532	639	AMPL22268883	E2F7	6.0	RNAseq
NM_024680	1444	1547	AMPL14009997	E2F8	6.0	RNAseq
NM_005228	438	543	AMPL28541017	EGFR	6.0	RNAseq
NM_001130678	233	339	AMPL4132220	EIF4E	6.0	RNAseq
NM_004095	305	409	AMPL27717688	EIF4EBP1	6.0	RNAseq
NM_020390	439	542	AMPL32456506	EIF5A2	6.0	RNAseq
NM_001127615	464	567	AMPL4408658	ENOX1	6.0	RNAseq
NM_182314	889	994	AMPL21591023	ENOX2	6.0	RNAseq
NM_001429	4967	5071	AMPL7241858	EP300	6.0	RNAseq

NM_025209	577	684	AMPL13895048	EPC1	6.0	RNAseq
NM_004439	2845	2949	AMPL28063841	EPHA5	6.0	RNAseq
NM_004448	1532	1637	AMPL28173828	ERBB2	6.0	RNAseq
NM_001982	413	521	AMPL11448617	ERBB3	6.0	RNAseq
NM_005235	229	336	AMPL27284273	ERBB4	6.0	RNAseq
NM_202001	190	295	AMPL21410533	ERCC1	6.0	RNAseq
NM_000400	96	201	AMPL2905825	ERCC2	6.0	RNAseq
NM_005236	2016	2118	AMPL28737581	ERCC4	6.0	RNAseq
NM_004456	781	884	AMPL27926041	EZH2	6.0	RNAseq
NM_000135	502	608	AMPL4937401	FANCA	6.0	RNAseq
NM_000136	1368	1475	AMPL1843962	FANCC	6.0	RNAseq
NM_022725	757	865	AMPL37319244	FANCF	6.0	RNAseq
NM_004629	1314	1421	AMPL28022492	FANCG	6.0	RNAseq
NM_000043	301	410	AMPL6420985	FAS	6.0	RNAseq
NM_005245	3793	3891	AMPL27071211	FAT1	6.0	RNAseq
NM_001447	10481	10591	AMPL10043234	FAT2	6.0	RNAseq
NM_001008781	3542	3648	AMPL919279	FAT3	6.0	RNAseq
NM_024582	5169	5275	AMPL15455383	FAT4	6.0	RNAseq
NM_033632	1523	1625	AMPL16316797	FBXW7	6.0	RNAseq
NM_000569	575	682	AMPL3610335	FCGR3A	6.0	RNAseq
NM_000800	696	804	AMPL5789556	FGF1	6.0	RNAseq
NM_001174067	1138	1246	AMPL8453388	FGFR1	6.0	RNAseq
NM_022970	2682	2789	AMPL36377917	FGFR2	6.0	RNAseq
NM_001163213	1884	1987	AMPL10721356	FGFR3	6.0	RNAseq
NM_002016	157	253	AMPL8411679	FLG	6.0	RNAseq
NM_004119	2108	2209	AMPL27128024	FLT3	6.0	RNAseq
NM_016725	288	397	AMPL31000380	FOLR1	6.0	RNAseq
NM_000804	324	425	AMPL4063208	FOLR3	6.0	RNAseq
NM_001199206	289	396	AMPL9002590	FOLR4	6.0	RNAseq
NM_004496	197	306	AMPL29995295	FOXA1	6.0	RNAseq
NM_002015	976	1081	AMPL8755973	FOXO1	6.0	RNAseq
NM_001455	884	992	AMPL9536179	FOXO3	6.0	RNAseq
NM_005269	552	662	AMPL27734562	GLI1	6.0	RNAseq
NM_002067	561	669	AMPL7673320	GNA11	6.0	RNAseq
NM_002072	867	975	AMPL11684262	GNAQ	6.0	RNAseq
NM_032119	18230	18334	AMPL15359727	GPR98	6.0	RNAseq
NM_002093	2159	2263	AMPL7670984	GSK3B	6.0	RNAseq
NM_000852	559	666	AMPL4426508	GSTP1	6.0	RNAseq
NM_002104	178	286	AMPL9245840	GZMK	6.0	RNAseq

NM_004964	169	271	AMPL27170842	HDAC1	6.0	RNAseq
NM_001527	1230	1331	AMPL11601143	HDAC2	6.0	RNAseq
NM_005524	469	578	AMPL27714869	HES1	6.0	RNAseq
NM_032580	107	199	AMPL18502798	HES7	6.0	RNAseq
NM_012259	386	492	AMPL12272409	HEY2	6.0	RNAseq
NM_014571	304	410	AMPL25840637	HEYL	6.0	RNAseq
NM_001530	1337	1439	AMPL8472036	HIF1A	6.0	RNAseq
NM_002127	483	585	AMPL8216930	HLA-G	6.0	RNAseq
NM_031935	6055	6162	AMPL14102879	HMCN1	6.0	RNAseq
NM_000545	489	597	AMPL2323015	HNF1A	6.0	RNAseq
NM_001130442	214	321	AMPL1124340	HRAS	6.0	RNAseq
NM_000413	1063	1167	AMPL4345976	HSD17B1	6.0	RNAseq
NM_004493	335	445	AMPL27578526	HSD17B10	6.0	RNAseq
NM_016245	355	468	AMPL32947097	HSD17B11	6.0	RNAseq
NM_178135	449	550	AMPL16810933	HSD17B13	6.0	RNAseq
NM_016246	861	970	AMPL33528637	HSD17B14	6.0	RNAseq
NM_002153	614	713	AMPL11677126	HSD17B2	6.0	RNAseq
NM_000197	349	457	AMPL3348088	HSD17B3	6.0	RNAseq
NM_000414	664	770	AMPL2905253	HSD17B4	6.0	RNAseq
NM_003725	410	518	AMPL7693179	HSD17B6	6.0	RNAseq
NM_016371	770	870	AMPL30919304	HSD17B7	6.0	RNAseq
NM_000862	26	132	AMPL2793468	HSD3B1	6.0	RNAseq
NM_000198	425	529	AMPL4724632	HSD3B2	6.0	RNAseq
NM_025193	385	495	AMPL14898030	HSD3B7	6.0	RNAseq
NM_005529	8027	8137	AMPL28063196	HSPG2	6.0	RNAseq
NM_005896	297	402	AMPL27566624	IDH1	6.0	RNAseq
NM_002168	720	825	AMPL11892986	IDH2	6.0	RNAseq
NM_001111285	417	518	AMPL5838528	IGF1	6.0	RNAseq
NM_000875	1229	1338	AMPL1833765	IGF1R	6.0	RNAseq
NM_000584	197	302	AMPL5817994	IL8	6.0	RNAseq
NM_176877	5179	5285	AMPL15901678	INADL	6.0	RNAseq
NM_031483	2503	2605	AMPL15095512	ITCH	6.0	RNAseq
NM_002210	1694	1802	AMPL8031875	ITGAV	6.0	RNAseq
NM_000212	749	853	AMPL3868029	ITGB3	6.0	RNAseq
NM_000214	2156	2264	AMPL1544343	JAG1	6.0	RNAseq
NM_145159	2837	2939	AMPL36559008	JAG2	6.0	RNAseq
NM_002227	3417	3519	AMPL8376933	JAK1	6.0	RNAseq
NM_004972	2772	2871	AMPL29237409	JAK2	6.0	RNAseq
NM_000215	316	426	AMPL2020493	JAK3	6.0	RNAseq

NM_001042603	1224	1327	AMPL1122415	KDM5A	6.0	RNAseq
NM_021140	1828	1934	AMPL37400297	KDM6A	6.0	RNAseq
NM_002253	1485	1586	AMPL7318204	KDR	6.0	RNAseq
NM_014774	2196	2299	AMPL26332646	KIAA0494	6.0	RNAseq
NM_000222	1122	1229	AMPL5414183	KIT	6.0	RNAseq
NM_033360	395	496	AMPL16322052	KRAS	6.0	RNAseq
NM_005559	5419	5520	AMPL28378784	LAMA1	6.0	RNAseq
NM_001040167	772	883	AMPL4591694	LFNG	6.0	RNAseq
NM_002317	1169	1274	AMPL8366296	LOX	6.0	RNAseq
NM_018557	6516	6619	AMPL30817456	LRP1B	6.0	RNAseq
NM_014757	2260	2367	AMPL26244940	MAML1	6.0	RNAseq
NM_032427	3688	3789	AMPL15506417	MAML2	6.0	RNAseq
NM_018717	3124	3231	AMPL30578808	MAML3	6.0	RNAseq
NM_002374	425	533	AMPL10103073	MAP2	6.0	RNAseq
NM_002755	666	773	AMPL10018074	MAP2K1	6.0	RNAseq
NM_030662	856	966	AMPL14285263	MAP2K2	6.0	RNAseq
NM_145109	856	965	AMPL36474922	MAP2K3	6.0	RNAseq
NM_003010	406	508	AMPL9798196	MAP2K4	6.0	RNAseq
NM_145160	866	948	AMPL35261487	MAP2K5	6.0	RNAseq
NM_005921	4584	4692	AMPL27448342	MAP3K1	6.0	RNAseq
NM_006609	1065	1168	AMPL12148967	MAP3K2	6.0	RNAseq
NM_002401	1459	1567	AMPL7474677	MAP3K3	6.0	RNAseq
NM_005922	1753	1859	AMPL28602471	MAP3K4	6.0	RNAseq
NM_005923	804	908	AMPL27107895	MAP3K5	6.0	RNAseq
NM_002745	1177	1285	AMPL8508553	MAPK1	6.0	RNAseq
NM_002969	321	428	AMPL11232233	MAPK12	6.0	RNAseq
NM_139012	641	749	AMPL36223989	MAPK14	6.0	RNAseq
NM_002746	1203	1310	AMPL7175442	MAPK3	6.0	RNAseq
NM_002747	1635	1737	AMPL9034720	MAPK4	6.0	RNAseq
NM_002748	1788	1887	AMPL9842890	MAPK6	6.0	RNAseq
NM_002749	2445	2547	AMPL8508709	MAPK7	6.0	RNAseq
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NM_001039469	1415	1523	AMPL5019307	MARK2	6.0	RNAseq
NM_014641	2535	2638	AMPL26336548	MDC1	6.0	RNAseq
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NM_020774	2712	2813	AMPL37300734	MIB1	6.0	RNAseq
NM_001167617	832	933	AMPL6964984	MLH1	6.0	RNAseq
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NM_170606	14602	14709	AMPL15606787	MLL3	6.0	RNAseq
NM_001199173	1004	1113	AMPL9313754	MLST8	6.0	RNAseq
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NM_002426	431	535	AMPL8177821	MMP12	6.0	RNAseq
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NM_002428	1277	1387	AMPL10443021	MMP15	6.0	RNAseq
NM_005941	1072	1181	AMPL26995318	MMP16	6.0	RNAseq
NM_016155	1464	1575	AMPL33652624	MMP17	6.0	RNAseq
NM_002429	797	900	AMPL8346069	MMP19	6.0	RNAseq
NM_004530	1105	1213	AMPL30209982	MMP2	6.0	RNAseq
NM_147191	1226	1327	AMPL34190725	MMP21	6.0	RNAseq
NM_006690	356	462	AMPL13038272	MMP24	6.0	RNAseq
NM_022468	988	1094	AMPL36519971	MMP25	6.0	RNAseq
NM_021801	530	635	AMPL35772412	MMP26	6.0	RNAseq
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NM_024302	431	536	AMPL37037907	MMP28	6.0	RNAseq
NM_002422	1107	1210	AMPL8346216	MMP3	6.0	RNAseq
NM_002423	516	619	AMPL11759516	MMP7	6.0	RNAseq
NM_004994	1174	1283	AMPL27989111	MMP9	6.0	RNAseq
NM_005373	835	945	AMPL28730730	MPL	6.0	RNAseq
NM_000251	1119	1223	AMPL4542401	MSH2	6.0	RNAseq
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NM_024690	43411	43518	AMPL15019677	MUC16	6.0	RNAseq
NM_002457	5450	5557	AMPL8871912	MUC2	6.0	RNAseq
NM_018406	14139	14247	AMPL31213936	MUC4	6.0	RNAseq
NM_015057	4948	5054	AMPL24379913	MYCBP2	6.0	RNAseq
NM_006540	4183	4291	AMPL13044799	NCOA2	6.0	RNAseq
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NM_006312	7037	7146	AMPL27417266	NCOR2	6.0	RNAseq
NM_015331	498	607	AMPL26118170	NCSTN	6.0	RNAseq
NM_001164508	18031	18138	AMPL7999843	NEB	6.0	RNAseq
NM_001042492	1514	1612	AMPL3996151	NF1	6.0	RNAseq
NM_000268	1115	1214	AMPL6592465	NF2	6.0	RNAseq
NM_003998	2345	2452	AMPL27136949	NFKB1	6.0	RNAseq
NM_001077494	965	1065	AMPL6185903	NFKB2	6.0	RNAseq
NM_017617	5930	6040	AMPL33493342	NOTCH1	6.0	RNAseq
NM_024408	1520	1630	AMPL35823019	NOTCH2	6.0	RNAseq
NM_000435	738	847	AMPL3430435	NOTCH3	6.0	RNAseq
NM_004557	2358	2467	AMPL27569053	NOTCH4	6.0	RNAseq
NM_002520	576	681	AMPL8187368	NPM1	6.0	RNAseq
NM_002524	303	407	AMPL11267387	NRAS	6.0	RNAseq
NM_003489	93	189	AMPL11392168	NRIP1	6.0	RNAseq
NM_004801	4960	5059	AMPL29358066	NRXN1	6.0	RNAseq
NM_015080	4577	4680	AMPL25474143	NRXN2	6.0	RNAseq
NM_004796	3216	3319	AMPL28007436	NRXN3	6.0	RNAseq
NM_001012338	1027	1137	AMPL2580213	NTRK3	6.0	RNAseq
NM_001005743	957	1059	AMPL936056	NUMB	6.0	RNAseq
NM_004756	1129	1236	AMPL27317997	NUMBL	6.0	RNAseq
NM_001098623	1855	1965	AMPL2976103	OBSCN	6.0	RNAseq
NM_002545	504	605	AMPL10048041	OPCML	6.0	RNAseq
NM_024675	3014	3116	AMPL14193671	PALB2	6.0	RNAseq
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NM_018313	271	377	AMPL32182407	PBRM1	6.0	RNAseq
NM_001142763	1917	2019	AMPL6180976	PCDH15	6.0	RNAseq
NM_033026	2109	2217	AMPL15716280	PCLO	6.0	RNAseq
NM_002607	1201	1307	AMPL9054458	PDGFA	6.0	RNAseq
NM_002608	1710	1816	AMPL10511493	PDGFB	6.0	RNAseq
NM_006206	1195	1301	AMPL27093990	PDGFRA	6.0	RNAseq
NM_002609	2793	2901	AMPL6946491	PDGFRB	6.0	RNAseq
NM_002613	962	1071	AMPL10512203	PDPK1	6.0	RNAseq
NM_001146184	1171	1280	AMPL7379639	PEG3	6.0	RNAseq
NM_006218	1278	1384	AMPL27428910	PIK3CA	6.0	RNAseq
NM_181523	2248	2355	AMPL15796294	PIK3R1	6.0	RNAseq
NM_005027	1903	2010	AMPL29342504	PIK3R2	6.0	RNAseq
NM_003629	1036	1142	AMPL6971173	PIK3R3	6.0	RNAseq
NM_000928	256	361	AMPL4412977	PLA2G1B	6.0	RNAseq
NM_001080954	527	630	AMPL1084498	PLAGL1	6.0	RNAseq

NM_016341	2421	2529	AMPL30694092	PLCE1	6.0	RNAseq
NM_012401	2348	2457	AMPL12107637	PLXNB2	6.0	RNAseq
NM_000535	846	956	AMPL2611585	PMS2	6.0	RNAseq
NM_000936	298	400	AMPL1809225	PNLIP	6.0	RNAseq
NM_005396	206	314	AMPL26905924	PNLIPRP2	6.0	RNAseq
NM_024870	2122	2231	AMPL14424417	PREX2	6.0	RNAseq
NM_002740	1904	2003	AMPL7637660	PRKCI	6.0	RNAseq
NM_002742	2685	2791	AMPL8507412	PRKD1	6.0	RNAseq
NM_002769	443	546	AMPL8328647	PRSS1	6.0	RNAseq
NM_000021	1212	1320	AMPL14470	PSEN1	6.0	RNAseq
NM_000447	54	157	AMPL5186868	PSEN2	6.0	RNAseq
NM_000264	542	645	AMPL2735774	PTCH1	6.0	RNAseq
NM_000314	1973	2078	AMPL1258705	PTEN	6.0	RNAseq
NM_005607	396	498	AMPL27593575	PTK2	6.0	RNAseq
NM_002834	1426	1533	AMPL7836151	PTPN11	6.0	RNAseq
NM_133170	4341	4449	AMPL17962429	PTPRT	6.0	RNAseq
NM_001080855	1495	1600	AMPL5092974	PXN	6.0	RNAseq
NM_020387	565	673	AMPL31006624	RAB25	6.0	RNAseq
NM_002875	775	883	AMPL10470661	RAD51	6.0	RNAseq
NM_133509	504	605	AMPL16253058	RAD51B	6.0	RNAseq
NM_058216	184	288	AMPL15676654	RAD51C	6.0	RNAseq
NM_001142571	1157	1264	AMPL3899153	RAD51D	6.0	RNAseq
NM_002880	584	689	AMPL7972359	RAF1	6.0	RNAseq
NM_006325	477	581	AMPL29167139	RAN	6.0	RNAseq
NM_170714	880	983	AMPL17627221	RASSF1	6.0	RNAseq
NM_000321	2459	2560	AMPL4177352	RB1	6.0	RNAseq
NM_002895	956	1061	AMPL7447897	RBL1	6.0	RNAseq
NM_005611	1420	1528	AMPL27471829	RBL2	6.0	RNAseq
NM_001204468	2818	2929	AMPL8840086	RBM10	6.0	RNAseq
NM_019610	280	386	AMPL33877560	RBMXL1	6.0	RNAseq
NM_015874	530	635	AMPL33316469	RBPJ	6.0	RNAseq
NM_014276	638	749	AMPL25934349	RBPJL	6.0	RNAseq
NM_001199771	841	951	AMPL9773680	RDH5	6.0	RNAseq
NM_004260	3011	3118	AMPL28074626	RECQL4	6.0	RNAseq
NM_002909	393	496	AMPL9367761	REG1A	6.0	RNAseq
NM_020975	3083	3189	AMPL35555716	RET	6.0	RNAseq
NM_005614	848	952	AMPL29765372	RHEB	6.0	RNAseq
NM_152756	511	612	AMPL34230729	RICTOR	6.0	RNAseq
NM_001100117	3580	3692	AMPL5481686	RIMS2	6.0	RNAseq

NM_020954	1902	2003	AMPL35562486	RNF213	6.0	RNAseq
NM_017763	1824	1931	AMPL31087747	RNF43	6.0	RNAseq
NM_002941	852	958	AMPL8294357	ROBO1	6.0	RNAseq
NM_002942	957	1066	AMPL8289722	ROBO2	6.0	RNAseq
NM_002944	5968	6075	AMPL8656446	ROS1	6.0	RNAseq
NM_001006665	1321	1425	AMPL348625	RPS6KA1	6.0	RNAseq
NM_001006932	544	651	AMPL137058	RPS6KA2	6.0	RNAseq
NM_003161	1295	1401	AMPL7785472	RPS6KB1	6.0	RNAseq
NM_003952	367	472	AMPL28449225	RPS6KB2	6.0	RNAseq
NM_020761	4111	4215	AMPL35157367	RPTOR	6.0	RNAseq
NM_001003699	1034	1141	AMPL1968710	RREB1	6.0	RNAseq
NM_001033	936	1042	AMPL1611364	RRM1	6.0	RNAseq
NM_000540	11872	11977	AMPL3225561	RYR1	6.0	RNAseq
NM_001035	9945	10047	AMPL1733055	RYR2	6.0	RNAseq
NM_001036	1723	1828	AMPL1520839	RYR3	6.0	RNAseq
NM_014363	2640	2745	AMPL24834319	SACS	6.0	RNAseq
NM_001159576	758	863	AMPL10039407	SCNN1A	6.0	RNAseq
NM_002999	471	577	AMPL7301130	SDC4	6.0	RNAseq
NM_001122752	387	484	AMPL5662405	SERPINI1	6.0	RNAseq
NM_012433	153	259	AMPL12373410	SF3B1	6.0	RNAseq
NM_001040	185	290	AMPL3074490	SHBG	6.0	RNAseq
NM_194255	1280	1388	AMPL21930828	SLC19A1	6.0	RNAseq
NM_004213	162	269	AMPL27356353	SLC28A1	6.0	RNAseq
NM_004212	1382	1489	AMPL27258113	SLC28A2	6.0	RNAseq
NM_001199633	597	704	AMPL7027115	SLC28A3	6.0	RNAseq
NM_001078176	1188	1298	AMPL1105895	SLC29A1	6.0	RNAseq
NM_004787	3519	3625	AMPL28261068	SLIT2	6.0	RNAseq
NM_005902	1079	1187	AMPL27676678	SMAD3	6.0	RNAseq
NM_005359	1745	1851	AMPL29138004	SMAD4	6.0	RNAseq
NM_003072	564	674	AMPL7948109	SMARCA4	6.0	RNAseq
NM_003073	523	631	AMPL7593839	SMARCB1	6.0	RNAseq
NM_005631	1856	1959	AMPL29337409	SMO	6.0	RNAseq
NM_012245	303	412	AMPL13062838	SNW1	6.0	RNAseq
NM_014758	3119	3224	AMPL26165780	SNX19	6.0	RNAseq
NM_001024465	587	696	AMPL1734190	SOD2	6.0	RNAseq
NM_000346	986	1093	AMPL5370025	SOX9	6.0	RNAseq
NM_015001	271	380	AMPL25046245	SPEN	6.0	RNAseq
NM_003122	242	344	AMPL9338361	SPINK1	6.0	RNAseq
NM_198291	856	962	AMPL22168733	SRC	6.0	RNAseq

NM_001047	804	904	AMPL5562052	SRD5A1	6.0	RNAseq
NM_024592	803	908	AMPL14248816	SRD5A3	6.0	RNAseq
NM_021908	1218	1321	AMPL35771376	ST7	6.0	RNAseq
NM_000349	351	457	AMPL4163525	STAR	6.0	RNAseq
NM_007315	1887	1996	AMPL12063649	STAT1	6.0	RNAseq
NM_005419	973	1083	AMPL27615998	STAT2	6.0	RNAseq
NM_139276	1751	1856	AMPL34373921	STAT3	6.0	RNAseq
NM_003152	1850	1954	AMPL8257905	STAT5A	6.0	RNAseq
NM_012448	1887	1989	AMPL12105990	STAT5B	6.0	RNAseq
NM_000455	1309	1416	AMPL5683663	STK11	6.0	RNAseq
NM_182961	19512	19616	AMPL20997674	SYNE1	6.0	RNAseq
NM_001163278	3297	3405	AMPL6770728	TENM1	6.0	RNAseq
NM_001122679	7304	7408	AMPL1166021	TENM2	6.0	RNAseq
NM_001080477	3577	3688	AMPL1805634	TENM3	6.0	RNAseq
NM_198253	2498	2605	AMPL20680601	TERT	6.0	RNAseq
NM_000660	1368	1474	AMPL4304112	TGFB1	6.0	RNAseq
NM_004612	1024	1131	AMPL28513658	TGFBR1	6.0	RNAseq
NM_001024847	1806	1912	AMPL686015	TGFBR2	6.0	RNAseq
NM_001130475	354	449	AMPL6206261	THAP5	6.0	RNAseq
NM_001080427	3087	3194	AMPL1658138	THSD7B	6.0	RNAseq
NM_001010938	1304	1410	AMPL119735	TNK2	6.0	RNAseq
NM_003286	890	997	AMPL7567651	TOP1	6.0	RNAseq
NM_000546	1249	1356	AMPL3223250	TP53	6.0	RNAseq
NM_001141980	3695	3802	AMPL4088949	TP53BP1	6.0	RNAseq
NM_001031685	1080	1184	AMPL1671575	TP53BP2	6.0	RNAseq
NM_000368	663	766	AMPL2671559	TSC1	6.0	RNAseq
NM_000548	1161	1266	AMPL3612152	TSC2	6.0	RNAseq
NM_003318	1487	1591	AMPL10426758	TTK	6.0	RNAseq
NM_133378	4985	5092	AMPL17955240	TTN	6.0	RNAseq
NM_020245	1678	1786	AMPL32912710	TULP4	6.0	RNAseq
NM_001071	673	780	AMPL4563892	TYMS	6.0	RNAseq
NM_006758	361	453	AMPL12030759	U2AF1	6.0	RNAseq
NM_000463	800	899	AMPL4494755	UGT1A1	6.0	RNAseq
NM_001076	1422	1530	AMPL1839870	UGT2B15	6.0	RNAseq
NM_001077	740	837	AMPL4973457	UGT2B17	6.0	RNAseq
NM_001074	1338	1442	AMPL2443325	UGT2B7	6.0	RNAseq
NM_001171623	1280	1389	AMPL8112284	VEGFA	6.0	RNAseq
NM_003377	862	953	AMPL11734697	VEGFB	6.0	RNAseq
NM_000551	453	559	AMPL6148676	VHL	6.0	RNAseq

NM_020945	7773	7882	AMPL34915315	WDFY4	6.0	RNAseq
NM_006103	391	500	AMPL26931127	WFDC2	6.0	RNAseq
NM_016373	558	664	AMPL31669480	WWOX	6.0	RNAseq
NM_006297	1115	1223	AMPL27997545	XRCC1	6.0	RNAseq
NM_005431	146	251	AMPL28183476	XRCC2	6.0	RNAseq
NM_001100118	298	395	AMPL3903442	XRCC3	6.0	RNAseq
NM_024721	3974	4079	AMPL14007565	ZFHX4	6.0	RNAseq
NM_014717	2415	2524	AMPL26170911	ZNF536	6.0	RNAseq

eTable 3. Univariable and multivariable logistic regression analyzing the association of preoperative demographic and clinical factors with favorable PR (score 0 or 1) showing that MMP-7 expression profile on FNA specimens prior to neoadjuvant therapy is an independent predictor of PR.

	Univariable Logistic Regression			Multivariable Logistic Regression		
	OR	95% CI	*P*	OR	95% CI	*P*
Age ≥ 65 (years)	0.88	[0.36 - 2.13]	0.77			
Sex (Male)	0.99	[0.41 - 2.41]	0.99			
Race (White)	1.43	[0.35 - 5.81]	0.61			
Type of Neoadjuvant (5FU-based)	0.92	[0.31 - 2.65]	0.88			
Chemo-Radiation (vs. Chemotherapy only)	4.82	[1.54 - 15.06]	**0.007**	7.42	[1.55 - 35.38]	**0.01**
Baseline Ca19-9 ≥200	0.90	[0.37 - 2.16]	0.81	1.36	[0.37 - 4.96]	0.63
Size (largest dimension) ≥ 3.5 cm	0.65	[0.26 - 1.58]	0.35	0.36	[0.10 -1.32]	0.13
Interval decrease in size	1.21	[0.40 -3.63]	0.73			
Grade (Poor)	0.86	[0.33 - 2.25]	0.76	0.85	[0.21 - 3.31]	0.81
MMP-7 (Negative)	21.25	[6.19 - 72.95]	**0.001**	32.85	[7.22 - 149.41]	**0.001**
Resectability Status			0.21			
Resectable	1 [Ref]					
Borderline resectable	0.83	[0.28 - 2.47]	0.74			
Locally advanced	0.39	[1.12 - 1.19]	0.10			

OR, Odds ratio; CI, Confidence Interval.
Neoadjuvant chemoradiation, baseline of CA 19-9 ≥ 200, tumor size ≥ 3.5 cm, poorly differentiated tumor grade, and MMP-7 status were applied for multivariable logistic regression analysis.

eTable 4. Demographics and tumor characteristics of FFPE surgical specimens in chemo-naïve cohort (n=60).

Variables, N (%)	MMP-7 Negative (N = 28)	MMP-7 Positive (N = 32)	*P*
Age (years) Median (IQR)	71 (60.2, 78.7)	71 (62.2, 76.7)	0.56
Sex (Male)	16 (57.1)	7 (21.9)	**0.008**
Race (White)	23 (82.1)	26 (81.3)	0.99
Ca19-9 Median (IQR)	96.1 (30.7, 277.1)	107 (35.2, 338.5)	0.79
Size (largest dimension in cm) Median (IQR)	3.1 (2.5, 4.8)	3.5 (2.5, 4.4)	0.79
Tumor location (Head/neck)	21 (75)	21 (65.6)	0.57
pT-stage			0.46
pT1	1 (3.6)	2 (6.3)	
pT2	14 (50)	11 (34.4)	
pT3	13 (46.4)	19 (59.4)	
pN-stage			0.99
pN0	7 (25)	8 (25)	
pN1	21 (75)	24 (75)	
Grade of differentiation			0.24
Well	2 (7.1)	0 (0)	
Moderate	14 (50)	20 (62.5)	
Poor	12 (42.9)	12 (37.5)	
Lymphovascular Invasion	17 (60.7)	23 (71.9)	0.41
Perineural Invasion	25 (89.3)	29 (90.6)	0.99
Margin Class (R0)	23 (82.1)	27 (84.4)	0.99
Adjuvant Chemotherapy	19 (70.4)	26 (83.8)	0.34

IQR, Interquartile range.

eTable 5. Area Under Curve (AUC) obtained from ROC curves (Figure S2) based on multivariable regression modelling with and without MMP-7 after stratification by resectability status at the time of diagnosis showing role of MMP-7 protein expression profile on FNA specimens in predicting favorable PR.

	Without MMP-7					With MMP-7			
	AUC [95% CI]	Hosmer-Lemeshow				AUC [95% CI]	Hosmer-Lemeshow		
		Chi-Square	*df*	*P*			Chi-Square	*df*	*P*
Resectable	0.688 [0.539 - 0.828]	8.14	6	0.419		0.902 [0.812 - 0.991]	6.37	7	0.606
Borderline resectable	0.800 [0.631 - 0.969]	8.53	7	0.287		0.881 [0.750 - 0.992]	5.45	6	0.604
Locally advanced	0.682 [0.470 - 0.895]	2.01	7	0.959		0.927 [0.833 - 0.998]	3.43	7	0.842

CI, Confidence Interval

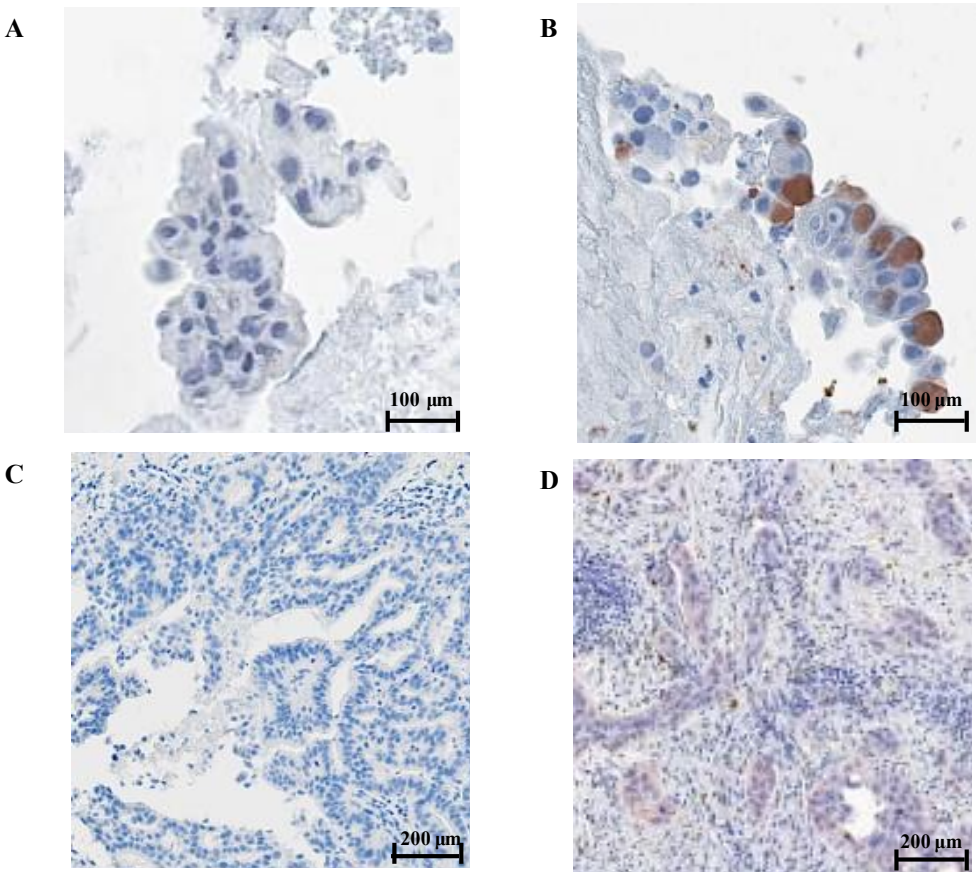
eTable 6. Univariable and multivariable Cox regression (backward-entry method) on FFPE surgical specimens in exploratory (chemo-naïve) cohort.

	Recurrence-Free Survival (RFS)							Overall Survival (OS)					
	Univariable Cox Regression			Multivariable Cox Regression				Univariable Cox Regression			Multivariable Cox Regression		
	HR	95% CI	*P*	HR	95% CI	*P*		HR	95% CI	*P*	HR	95% CI	*P*
Age ≥ 65 years	1.01	[0.97 - 1.02]	0.92					1.62	[0.84 - 3.09]	0.14			
Sex (Male)	0.63	[0.34 - 1.17]	0.15					0.93	[0.50 - 1.72]	0.84			
Race (White)	1.26	[0.62 - 2.55]	0.52					1.81	[0.76 - 4.29]	0.17			
Baseline Ca19-9 ≥200	1.61	[0.87 - 2.95]	0.12					1.26	[0.68 - 2.31]	0.46			
Grade (Poor)	2.18	[1.20 - 3.97]	**0.01**					2.16	[1.19 - 3.93]	**0.01**			
T-stage ≥ T3	2.26	[1.22 - 4.18]	**0.009**					2.88	[1.52 - 5.47]	**0.001**			
N-stage ≥ N1	1.12	[0.58 - 2.19]	0.72					2.29	[1.09 - 4.80]	**0.03**	3.20	[1.43 - 7.42]	**0.005**
Lymphovascular Invasion	1.62	[0.86 - 3.04]	0.13					1.86	[0.96 - 3.63]	0.07			
Perineural Invasion	2.07	[0.72 - 5.91]	0.17					2.24	[0.69 - 7.26]	0.18			
Margin Class (R0)	0.97	[0.41 - 2.31]	0.98					0.71	[0.32 - 1.52]	0.38			
MMP-7 (Negative)	0.35	[0.18 - 0.68]	**0.002**	0.23	[0.11 - 0.49]	**0.001**		0.53	[0.28 - 0.79]	**0.03**	0.44	[0.23 - 0.85]	**0.02**
Adjuvant Chemotherapy	0.43	[0.20 - 0.91]	**0.03**	0.28	[0.11 - 0.70]	**0.07**		0.27	[0.13 - 0.53]	**0.001**	0.19	[0.09 - 0.42]	**0.001**

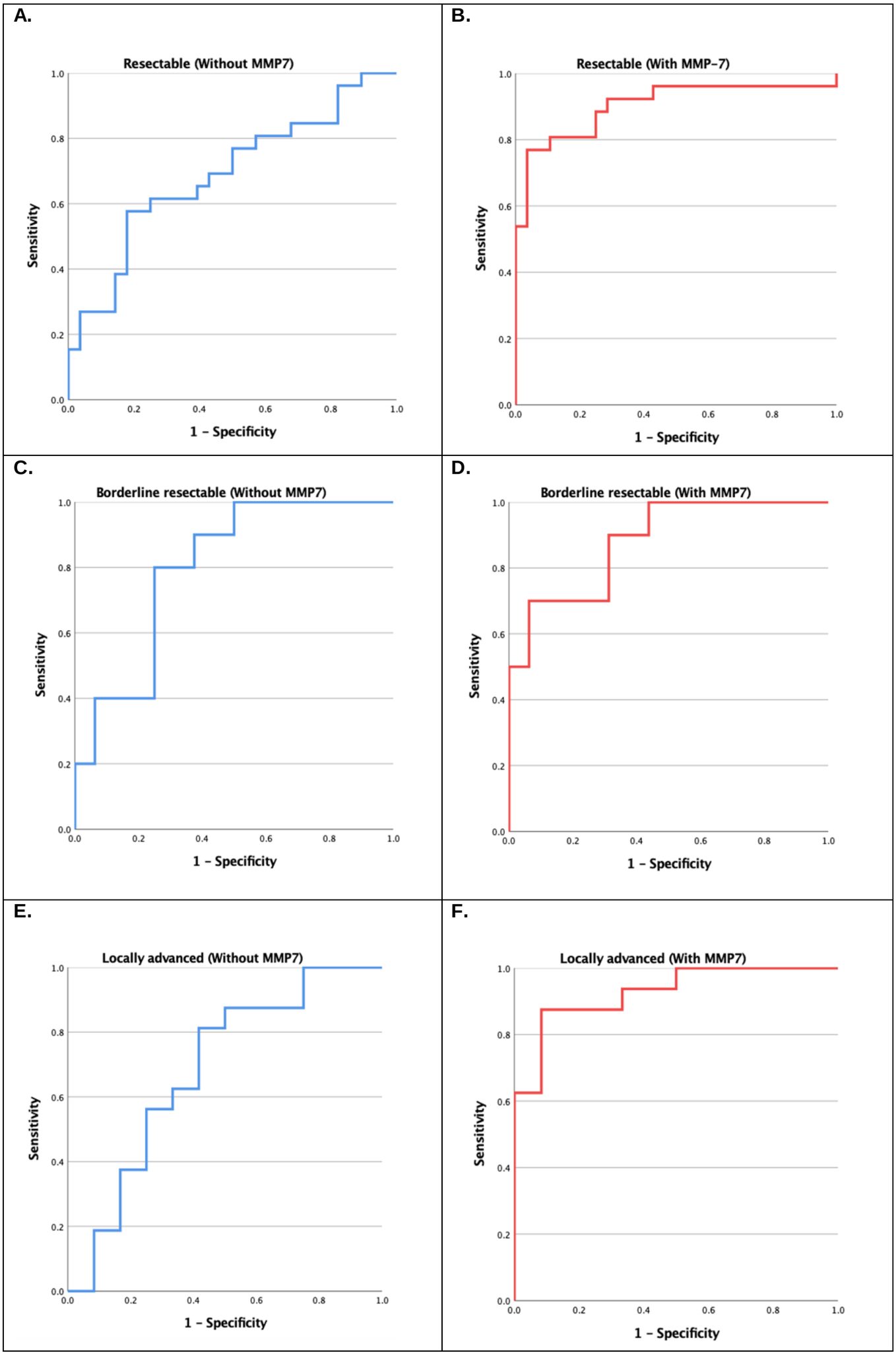
HR, hazard ratio; CI, confidence interval.

Poorly differentiated tumor grade, T-stage ≥ T3, N-stage ≥ N1, MMP-7 status, and adjuvant chemotherapy were applied multivariable Cox regression analysis (backward-entry method).

eFigure 1. IHC staining of tissue biopsy prior to neoadjuvant therapy (validation cohort) representing negative (A) and positive (B) MMP-7 protein expression; IHC staining of primary tumor surgical specimens from chemo-naïve cohort (exploratory cohort) showing negative (C) and positive (D) MMP-7 protein expression.



eFigure 2. ROC curves based on multivariable regression modelling with and without MMP-7 after stratification by resectability status at the time of diagnosis showing role of MMP-7 protein expression profile on FNA specimens in predicting favorable PR.



eFigure 3. Kaplan-Meier Overall Survival and Recurrence-Free Survival Curves comparing MMP-7 positive and negative expression in FNA specimens from cohort undergoing neoadjuvant therapy (A, B) and FFPE surgical specimens from chemo-naïve cohort (C, D).