

Supplementary data to:

Impact of *TP53* Loss-of-Function Alterations on the Response to PSMA Radioligand Therapy in Metastatic Castration-Resistant Prostate Cancer Patients

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Supplementary Table 1. Individual data on activity and interval per cycle of PSMA-RLT.

Study ID	TP53 status	Number of cycles	Mean weeks in between cycles	Total ¹⁷⁷ Lu activity (GBq)	Mean ¹⁷⁷ Lu activity per cycle (GBq)	Total ²²⁵ Ac activity (MBq)	Mean ²²⁵ Ac activity per cycle (MBq)
1	TP53wt	3	5,0			24,0	8,0
2	TP53wt	1				8,0	8,0
3	TP53wt	4	8,4	15,0	3,8	17,5	4,4
4	TP53m	2	5,5	13,9	7,0	2,2	1,1
5	TP53wt	2	4,6	14,9	7,5		
6	TP53m	2	3,9			14,0	7,0
7	TP53wt	6	10,4	24,0	4,0	26,0	4,3
8	TP53m	3	3,4	21,1	7,0	4,1	1,4
9	TP53wt	6	5,7	44,4	7,4		
10	TP53m	2	2,4	16,7	8,4		
11	TP53m	2	3,6	14,7	7,4		
12	TP53m	6	7,3	36,2	6,0		
13	TP53wt	3	5,7	18,5	6,2		
14	TP53m	3	5,7	14,0	4,7	8,0	2,7
15	TP53wt	3	5,2			20,1	6,7
16	TP53wt	5	5,9	30,2	6,0		
17	TP53m	2	3,5	11,4	5,7		
18	TP53wt	7	31,0	13,4	1,9	35,0	5,0
19	TP53wt	4	8,1			31,4	7,9
20	TP53wt	2	4,0	12,0	6,0		
21	TP53wt	2	3,4	10,7	5,4		
22	TP53wt	2	4,6	5,0	2,5	12,4	6,2
23	TP53m	3	3,8	19,8	6,6		
24	TP53m	4	4,8	29,6	7,4	2,2	0,6
25	TP53wt	4	5,6	24,0	6,0		
26	TP53m	2	4,0	16,5	8,3	2,0	1,0
27	TP53m	3	7,0			18,8	6,3
28	TP53wt	2	3,9	10,0	5,0		
29	TP53wt	4	4,8	24,4	6,1		
30	TP53m	2	3,5	12,0	6,0		
31	TP53wt	3	6,4			20,0	6,7
32	TP53m	3	13,3	12,2	4,1		
33	TP53wt	4	5,0	24,1	6,0		
34	TP53wt	4	5,6	24,5	6,1		
35	TP53wt	6	6,9	35,8	6,0		
36	TP53wt	4	4,0	28,2	7,1		
37	TP53wt	3	7,0	18,3	6,1		
38	TP53wt	4	5,3	24,0	6,0		
39	TP53m	2	4,9	15,4	7,7		
40	TP53wt	2	4,5	14,0	7,0	14,5	7,3
41	TP53m	6	5,0	44,4	7,4		
42	TP53wt	6	5,0	44,4	7,4		
43	TP53wt	6	5,0	44,4	7,4		
44	TP53m	3	4,0	22,2	7,4		
45	TP53wt	6	5,6	44,4	7,4		
46	TP53wt	6	5,2	44,4	7,4		
47	TP53m	6	5,0	44,4	7,4		
48	TP53wt	6	5,1	44,4	7,4		
49	TP53wt	3	4,0	22,2	7,4		
50	TP53wt	6	5,0	44,4	7,4		
52	TP53wt	3	4,1	22,2	7,4		
53	TP53m	4	4,4	29,6	7,4		
54	TP53wt	2	3,0	14,8	7,4		
55	TP53wt	1		7,4	7,4		
56	TP53wt	3	20,6	22,2	7,4		
57	TP53wt	6	4,9	44,4	7,4		
59	TP53m	6	5,7	44,4	7,4		

60	TP53m	2	3,0	14,8	7,4		
61	TP53wt	3	5,7	25,5	8,5		
62	TP53m	1		6,0	6,0		
63	TP53wt	1		6,7	6,7		
64	TP53wt	3	4,7	18,0	6,0		
65	TP53m	2	3,0	13,7	6,9		
66	TP53wt	2	7,3	11,4	5,7	4,0	2,0
67	TP53m	2	3,9	13,6	6,8		
68	TP53wt	2	3,0	14,8	7,4		
69	TP53m	6	5,1	44,4	7,4		
101	TP53wt	5	10,1				
102	TP53m	2	3,0	14,8	7,4		
103	TP53wt	2	3,4	14,9	7,5		
104	TP53m	3	4,1	22,2	7,4		
105	TP53wt	1		7,4	7,4		
106	TP53wt	6	6,0	44,4	7,4		
107	TP53wt	3	4,0	22,2	7,4		
108	TP53wt	6	5,7	44,4	7,4		
109	TP53m	2	1,0				
110	TP53wt	6	6,0	44,4	7,4		
111	TP53wt	4	4,5				
112	TP53wt	1		7,4	7,4		
113	TP53wt	2	3,0	14,8	7,4		
114	TP53wt	6	6,2	44,4	7,4		
115	TP53m	3	6,0	14,0	4,7	10,0	3,3
116	TP53wt	6	6,2	44,4	7,4		
117	TP53wt	5	5,8	36,6	7,3		
118	TP53m	5	7,0	38,4	7,7	22,8	4,6
119	TP53m	3	6,3	20,4	6,8		
120	TP53wt	4	6,5	27,2	6,8		
121	TP53wt	5	4,4	37,0	7,4		
123	TP53wt	4	6,2	27,2	6,8		
124	TP53wt	6	5,5	44,4	7,4		
125	TP53wt	2	4,0	17,8	8,9		
126	TP53m	2	3,9	13,6	6,8		
127	TP53m	1					
128	TP53wt	2	1,0	14,8	7,4		
129	TP53wt	4	4,5	29,6	7,4		
130	TP53wt	5	1,2	37,0	7,4		

¹⁷⁷Lu, ¹⁷⁷Lutetium; ²²⁵Ac, ²²⁵Actinium; GBq, gigaBecquerel; MBq, megaBecquerel; TP53m, *TP53* mutated; TP53wt, *TP53* wildtype.

Supplementary Table 2. Multivariable analysis of *TP53*-mutational status and known prognostic factors for progression-free survival on PSMA-RLT.

Variables	HR [95%CI]	<i>P</i> -value
TP53m	2.27 [1.27-4.05]	0.005
Line of therapy	1.09 [0.93-1.28]	0.298
Time from initial diagnosis to castration-resistance	1.00 [0.99-1.01]	0.999
SUVmax of highest lesions on baseline PSMA-PET scan	1.00 [0.99-1.01]	0.703
Presence of visceral metastasis on baseline PSMA-PET scan	0.85 [0.56-1.58]	0.607
Type of radioligand therapy	0.76 [0.52-1.11]	0.151
Laboratory variables at baseline:		
PSA	1.00 [1.00-1.00]	0.133
LDH	1.00 [1.00-1.01]	0.001
HB	0.92 [0.76-1.12]	0.414
ALP	1.00 [1.00-1.00]	0.163

ALP, alkaline phosphatase; CI, confidence interval; HB, haemoglobin; HR, hazard ratio; LDH, lactate dehydrogenase; PSA, prostate specific antigen; SUVmax, maximum standardised uptake values.

Supplementary Table 3. Biochemical response per mutational status of canonical oncogenic or tumour-suppressive pathways.

Genetic subgroup	Effect of alteration	Median PSA response		Patients with PSA50	
		Mutated vs. WT	<i>P</i> -value	Mutated vs. WT	<i>P</i> -value
<i>TP53</i>	Inactivation	-55% vs. -75%	0.012	45% vs. 56%	0.569
<i>AR</i>	Hyperactivation	-70% vs. -61%	0.294	69% vs. 57%	0.380
<i>MYC</i>	Hyperactivation	-66% vs. -65%	0.306	57% vs. 61%	1.000
PI3K	Hyperactivation	-64% vs. -68%	0.456	62% vs. 61%	0.980
<i>BRCA1/2</i>	Inactivation	-35% vs. -66%	0.971	45% vs. 63%	0.327
HRR	Inactivation	-69% vs. -64%	0.804	59% vs. 62%	0.825
MAPK	Hyperactivation	-85% vs. -64%	0.214	100% vs. 51%	0.154

HRR, homologous recombination repair; PSA, $\geq 50\%$ PSA decline; PSA, prostate specific antigen; WT, wildtype.

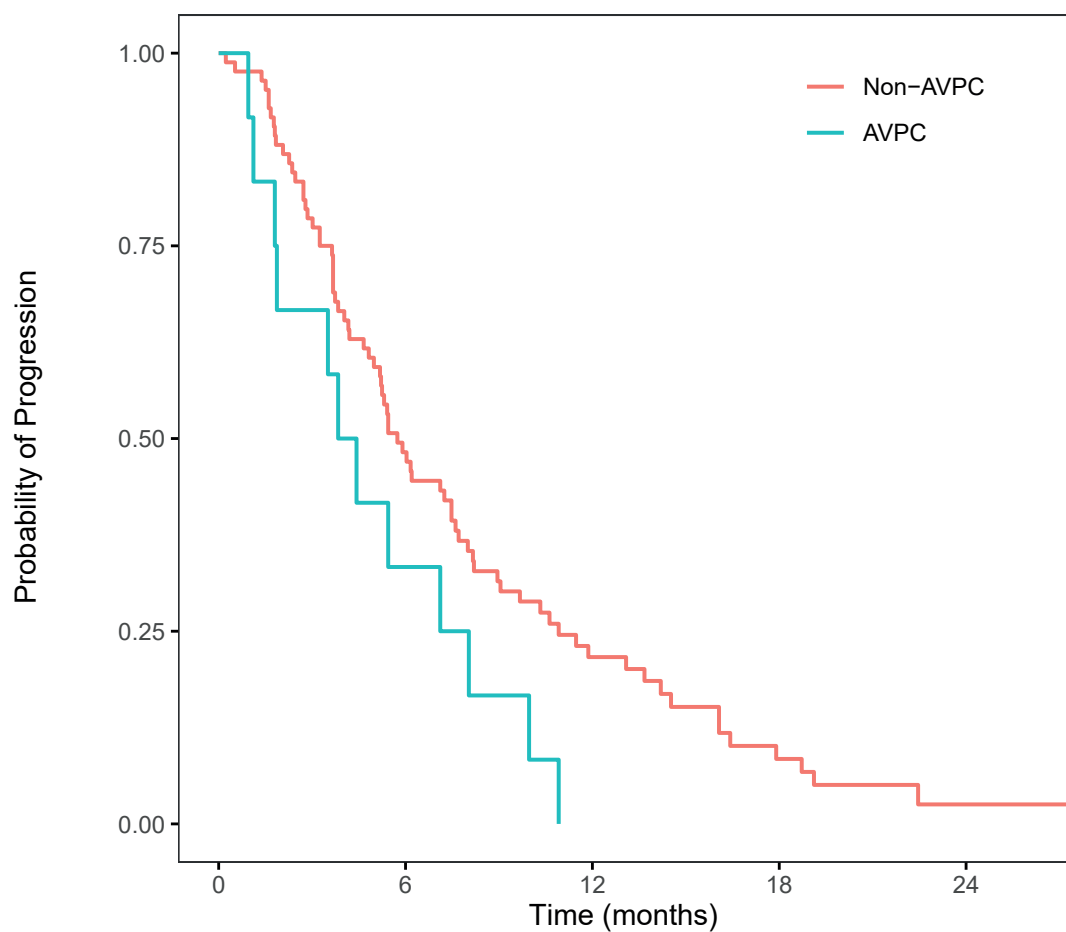
Supplementary Table 4. Multivariable analysis for change of biochemical progression per mutational status of canonical oncogenic or tumour-suppressive pathways.

Genetic subgroup	Univariable analysis	
	HR [95%CI]	P-value
<i>TP53</i>	2.53 [1.52-4.22]	<0.001
<i>AR</i>	0.85 [0.69-1.05]	0.130
<i>MYC</i>	0.61 [0.26-1.45]	0.263
<i>PI3K</i>	1.13 [0.70-1.82]	0.628
<i>BRCA1/2</i>	0.90 [0.43-1.89]	0.776
<i>HRR</i>	1.07 [0.61-1.88]	0.811
<i>MAPK</i>	0.99 [0.33-3.02]	0.987

CI, confidence interval; HR, hazard ratio; HRR, homologous recombination repair.

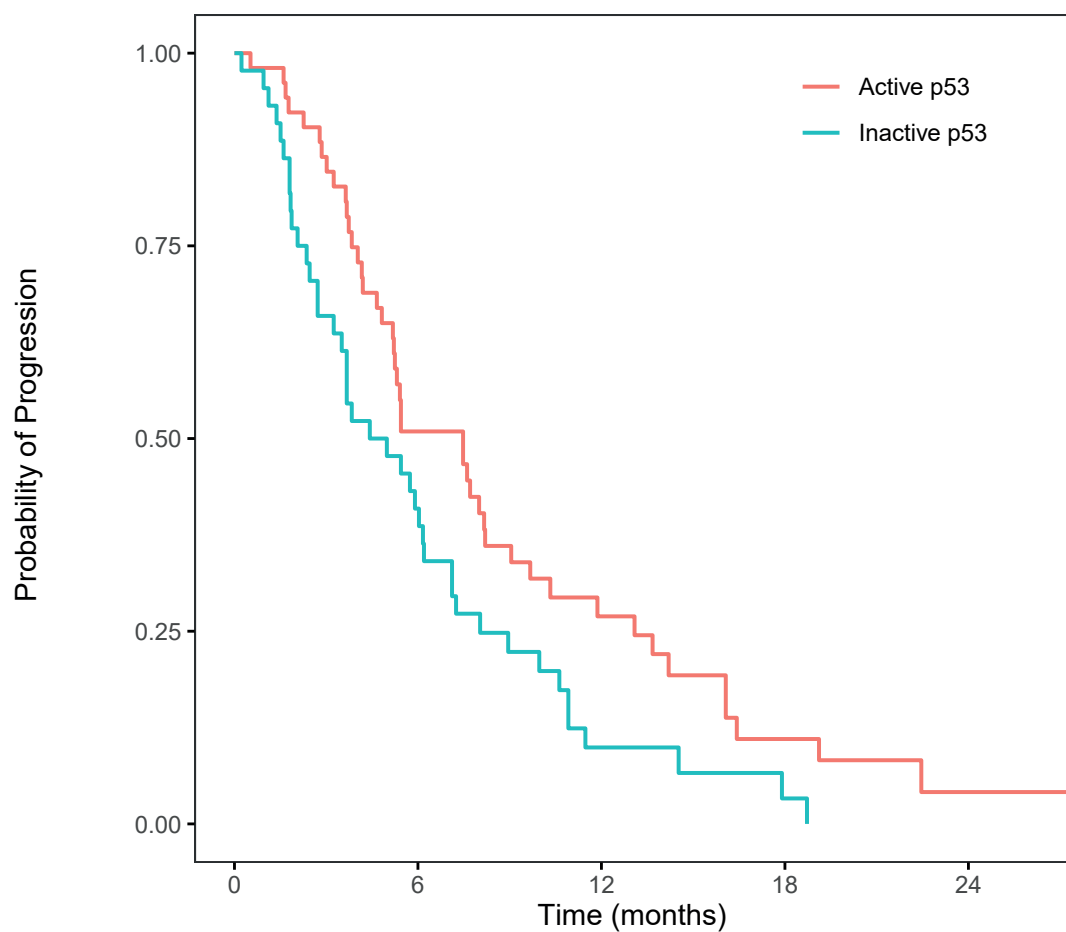
Supplementary Table 5. Genes from the signature HALLMARK P53 PATHWAY driving enrichment in PSA50/No PSA50 and decreased in TP53m/TP53wt.

TP53m/TP53wt		PSA50/No PSA50		Intersect
<i>ALOX15B</i>	<i>RAD9A</i>	<i>TNNI1</i>	<i>LIF</i>	FOS
<i>FOS</i>	<i>RAD51C</i>	<i>FOS</i>	<i>IRAG2</i>	SESN1
<i>SESN1</i>	<i>H2AJ</i>	<i>CTSF</i>	<i>HDAC3</i>	TXNIP
<i>PHLDA3</i>	<i>CSRNP2</i>	<i>TP63</i>	<i>TGFA</i>	RPS27L
<i>TXNIP</i>	<i>SERTAD3</i>	<i>INHBB</i>	<i>RPL18</i>	TGFA
<i>FAM162A</i>	<i>RPL36</i>	<i>SESN1</i>	<i>RB1</i>	TSPYL2
<i>KLF4</i>	<i>HDAC3</i>	<i>HBEGF</i>	<i>TSC22D1</i>	MAPKAPK3
<i>JUN</i>	<i>CYFIP2</i>	<i>H2AJ</i>	<i>TSPYL2</i>	LIF
<i>RPS27L</i>	<i>BAX</i>	<i>BTG2</i>	<i>VDR</i>	ATF3
<i>TGFA</i>	<i>HRAS</i>	<i>LDHB</i>	<i>RAP2B</i>	IRAG2
<i>TSPYL2</i>	<i>POM121</i>	<i>CYFIP2</i>	<i>ACVR1B</i>	CCNG1
<i>DDB2</i>	<i>FOXO3</i>	<i>RACK1</i>	<i>VWA5A</i>	HBEGF
<i>MAPKAPK3</i>	<i>FDXR</i>	<i>TAP1</i>	<i>COQ8A</i>	FUCA1
<i>CDKN1A</i>	<i>ZMAT3</i>	<i>GM2A</i>	<i>ABCC5</i>	H2AJ
<i>LIF</i>	<i>RAP2B</i>	<i>RRAD</i>	<i>ITGB4</i>	HDAC3
<i>DCXR</i>	<i>TNFSF9</i>	<i>MAPKAPK3</i>	<i>EPHX1</i>	CYFIP2
<i>MDM2</i>	<i>ANKRA2</i>	<i>STEAP3</i>	<i>ATF3</i>	RAP2B
<i>ATF3</i>	<i>CGRRF1</i>	<i>TGFB1</i>	<i>NUDT15</i>	ANKRA2
<i>BAIAP2</i>	<i>MXD1</i>	<i>CCNG1</i>	<i>TXNIP</i>	
<i>IRAG2</i>	<i>XPC</i>	<i>FUCA1</i>	<i>ANKRA2</i>	
<i>DGKA</i>		<i>S100A4</i>	<i>PTPRE</i>	
<i>CCNG1</i>		<i>SOCS1</i>	<i>SPHK1</i>	
<i>HBEGF</i>		<i>RPS27L</i>	<i>KRT17</i>	
<i>AEN</i>		<i>EPHA2</i>	<i>RPS12</i>	
<i>FUCA1</i>		<i>EI24</i>	<i>HEXIM1</i>	
<i>HMOX1</i>		<i>SAT1</i>	<i>PVT1</i>	
<i>SFN</i>		<i>BMP2</i>		



Number at risk

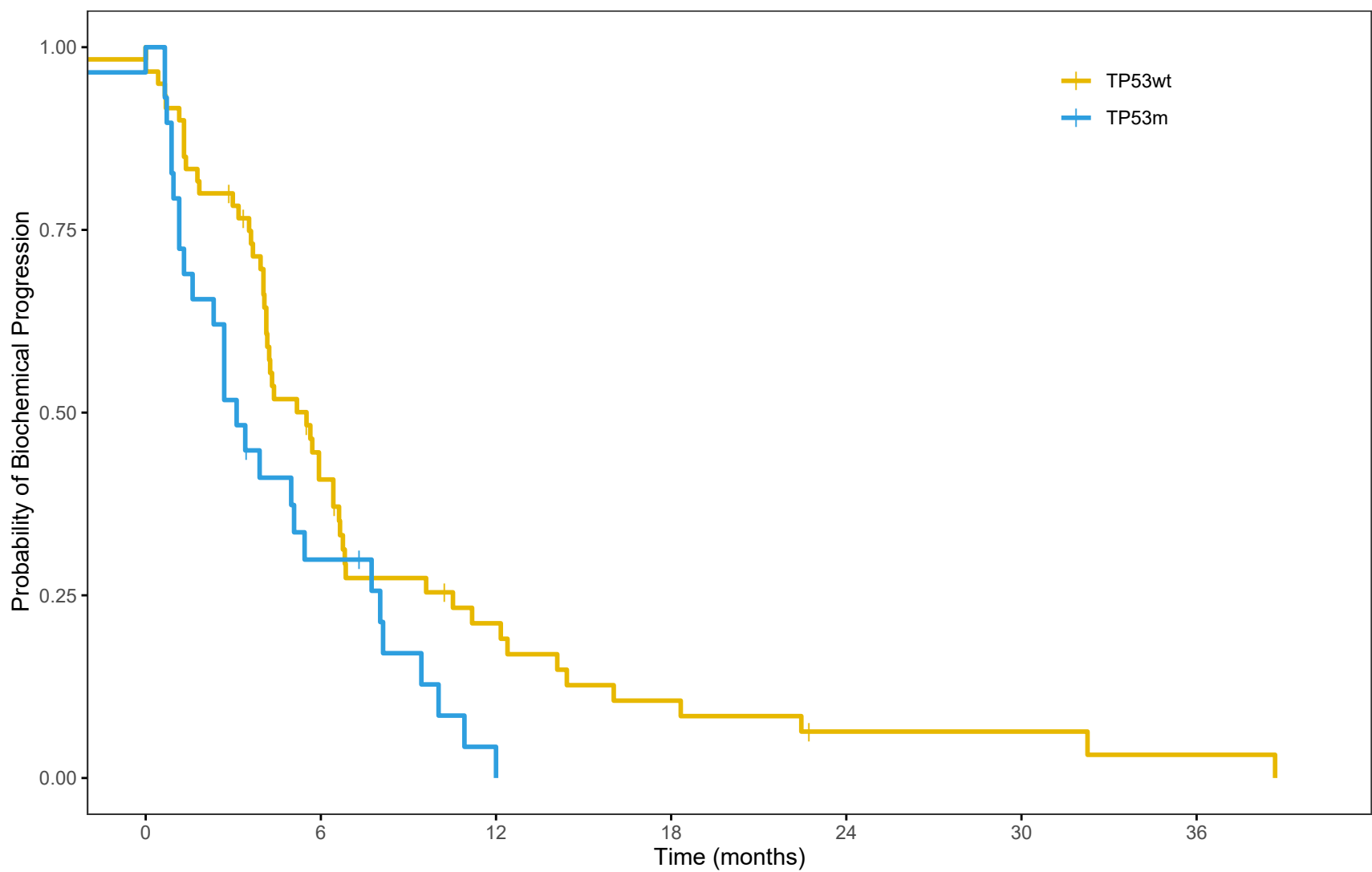
Non-AVPC	84	39	15	5	1
AVPC	12	4	0	0	0
	0	6	12	18	24



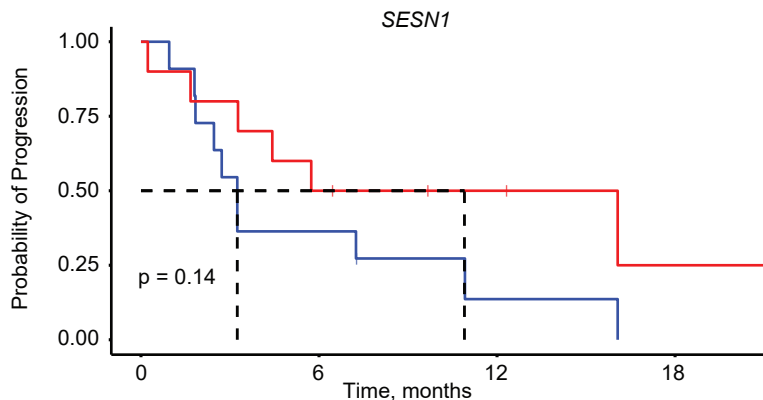
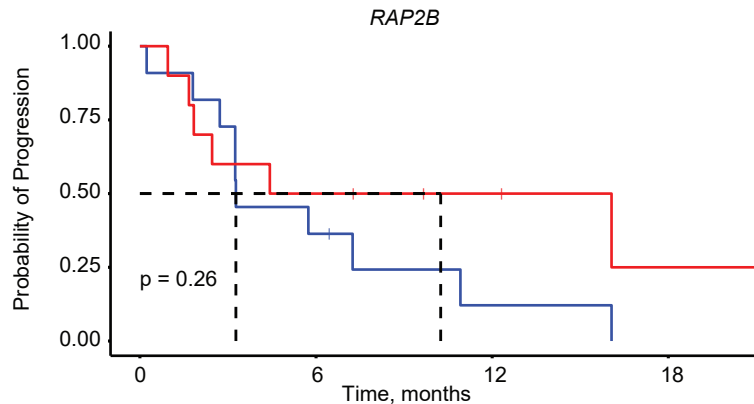
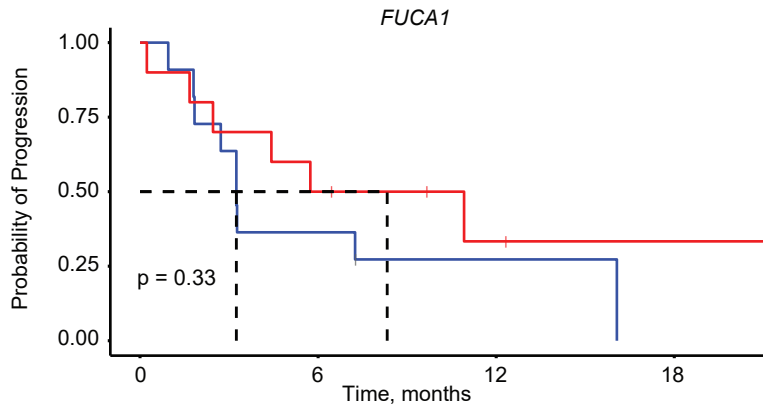
Number at risk

Active P53	52	25	11	4	1
Inactive P53	44	18	4	1	0
	0	6	12	18	24

Kaplan-Meier curves for the progression-free survival, including a table presenting the number of patients at risk for: **(Left)** patients with and without an aggressive variant prostate cancer (AVPC) molecular signature and **(Right)** patients with and without an impaired p53 signalling based on loss-of-function alterations in TP53, ATM, CHEK1, or CHEK2.



Kaplan-Meier curves for the biochemical progression-free survival probability per TP53-mutational status.



Expression levels:

+ Highest 50%

+ Lowest 50%

Kaplan-Meier curves for the progression-free survival based on the expression of *FUCA1*, *RAP2B*, and *SESN1*.