

**Supplementary Table 1** – Results of the internal validation phase from 2X parallel runs for 4 different NGS panels for 9 tested cell lines.

Cell line	Illumina		ThermoFisher		Qiagen		Foundation Medicine	
	RUN1	RUN2	RUN1	RUN2	RUN1	RUN2	RUN1	RUN2
<b>NCI-H2009</b>	18.801	18.147	19.31	18.41	13.69	12.92	15.13	15.13
<b>NCI-H2126</b>	17.864	16.479	16.81	16.74	8.38	8.38	13.87	13.87
<b>NCI-H2171</b>	4.248	3.92	9.24	9.19	1.52	1.52	10.09	10.09
<b>NCI-H1437</b>	6.282	7.058	7.54	7.56	5.33	5.33	5.04	5.04
<b>NCI-H23</b>	15.667	13.298	23.5	24.26	9.11	9.88	27.74	27.74
<b>NCI-H322</b>	8.651	8.632	10.9	10.9	6.09	5.32	25.22	25.22
<b>SKMEL2</b>	18.003	25.174	17.56	17.53	16.71	16.72	34.04	44.13
<b>C33A</b>	66.942	64.147	36.23	37.49	25.08	25.08	69.35	69.35
<b>IGROV1</b>	112.715	113.429	71.6	61.73	51.66	51.66	71.87	76.91

**Supplementary Table 2 – Description of the participating NGS methods at the pilot EQA scheme**

<b>Company</b>	<b>Panel Name</b>	<b>Type of NGS approach</b>	<b>Type of variants used to infer TMB</b>	<b>Number of gene</b>
<b>IDT</b>	Custom panel	Gene targated	Non synonymous mutations, Indels	423
<b>Twist Bioscience</b>	Custom panel	Gene targated	Non-synonymous mutations, Splice variants, Indels	350
<b>Twist Bioscience</b>	Custom panel	Gene targated	Synonymous and non-synonymous mutations, Indels	645
<b>Agilent</b>	Custom panel	Gene targated	Non synonymous mutations, Indels	431
<b>Illumina</b>	TruSight Oncology 500	Gene targated	Non synonymous mutations, Indels	523
<b>Thermo Fisher Scientific</b>	Oncomine Tumor Mutation Load	Gene targated	Non synonymous and synonymous mutations, Indels	409
<b>Thermo Fisher Scientific</b>	Oncomine Tumor Mutation Load	Gene targated	Non synonymous mutations, Indels	409
<b>Thermo Fisher Scientific</b>	Oncomine Comprehensive Assay Plus	Gene targated	Non synonymous mutations, Indels	514
<b>IDT</b>	IDT xGen Pan-Cancer Panel v2.4	Clinical exome	Non synonymous mutations	523
<b>Agilent</b>	Custom Panel	Clinical exome	Non synonymous mutations, Indels	431
<b>Twist Bioscience</b>	Twist human Exome plus	Whole exome sequencing	Non-synonymous mutations, Splice variants, Indels	22000
<b>Roche</b>	Nimblegen (Roche) SeqCap EZ MedExome	Whole exome sequencing	Synonymous and non-synonymous mutations, Indels	22334