

**Table S1. Sensitivity of the IDH1/2 PCR assay for the 12 detected and 3 identified IDH1/2 mutations**

<b>Assay</b>	<b>Mutation</b>	<b>LOD</b>	<b>Assay cutoff *</b>	<b>Sensitivity (%)</b>
R132HMut	R132H	6.87	6.87	0.78
R132CMut	R132C	7.14	7.14	1.19
R172KMut	R172K	8.49	8.49	0.61
R132Mut	R132H	5.50	5.34	2.32
	R132C	5.34		4.35
	R132L	5.42		2.30
	R132G	5.61		2.23
	R132S	5.42		2.75
	R132V	5.56		2.24
R172Mut	R172K	6.42	6.42	1.06
	R172G	6.58		3.00
	R172M	6.66		3.31
	R172S	6.42		14.93
	R172W	6.68		2.36
R100Mut	R100Q	4.65	4.65	3.45

LOD: Limit of Detection.

\*A mutation is detected if  $\Delta Ct$  is less or equal to the LOD

**Table S 2. Precision results**

Assay	Sample	Mean $\Delta C_T$	SD <sub>R</sub> <sup>*</sup>	SD <sub>Run</sub> <sup>†</sup>	SD <sub>Total</sub> <sup>‡</sup>	CV <sub>Total</sub> (%) <sup>‡</sup>	Correct calls rate % (Frequency)
<b>R132C MUT</b>	WT	11.58	1.08	0.00	1.11	10	100% (78/78)
	5%	5.19	0.26	0.23	0.46	9	100% (76/76)
	10%	4.37	0.27	0.14	0.48	11	100% (78/78)
	30%	2.62	0.20	0.21	0.46	18	100% (78/78)
<b>R132H MUT</b>	WT	10.87	1.48	0.00	1.48	14	100% (78/78)
	5%	4.46	0.27	0.05	0.31	7	100% (78/78)
	10%	3.57	0.28	0.14	0.31	9	100% (76/76)
	30%	1.86	0.21	0.20	0.30	16	100% (72/72)
<b>R172K MUT</b>	WT	12.20	0.31	0.17	0.39	3	100% (66/66)
	5%	6.19	0.50	0.00	0.63	10	100% (76/76)
	10%	5.23	0.32	0.20	0.48	9	100% (76/76)
	30%	3.68	0.18	0.11	0.36	10	100% (76/76)
<b>R100 MUT</b>	WT	7.21	0.41	0.27	0.52	7	100% (70/70)
	5%	3.68	0.27	0.16	0.33	9	100% (76/76)
	10%	2.93	0.24	0.15	0.32	11	100% (76/76)
	30%	1.56	0.25	0.07	0.26	17	100% (76/76)
<b>R132 MUT</b>	WT	8.01	0.76	0.00	0.78	10	100% (152/152)
	R132H 5%	4.29	0.30	0.15	0.48	11	
	R132C 5%	4.44	0.30	0.00	0.56	13	
<b>R132 MUT</b>	R132H 10%	3.49	0.27	0.22	0.46	13	99% (151/152)
	R132C 10%	3.69	0.27	0.23	0.53	14	
	R132H 30%	1.87	0.21	0.02	0.33	18	100% (152/152)
	R132C 30%	2.00	0.26	0.28	0.59	29	
<b>R172 MUT</b>	WT	9.47	0.91	0.87	1.45	15	100% (66/66)
	5%	4.45	0.35	0.12	0.56	13	100% (76/76)
	10%	3.55	0.29	0.02	0.53	15	100% (76/76)
	30%	2.05	0.18	0.15	0.47	23	100% (76/76)

WT: Wild-type; SD: Standard deviation; CV: Coefficient of variation.

\* R: Repeatability; † Run: Between-run reproducibility; ‡ Total: Total precision (including inter-instrument, inter-operator, and inter-lot).

**Table S 3. Analysis of Agreement between the IDH1/2 PCR assay and IDH1 R132H-IHC and Sanger sequencing**

	<b>Measure of agreement</b>	<b>% (Frequency)</b>	<b>95% CI</b>
<b>IDH1/2 PCR vs IHC</b>	Overall percent agreement	99.0% (102/103)	[95.9 - 99.9]
	Positive percent agreement	98.4% (60/61)	[91.3 - 99.7]
	Negative percent agreement	100% (72/72)	[94.9 - 100]
<b>IDH1/2 PCR vs Sanger Sequencing</b>	Overall percent agreement	96.2% (128/133)	[91.5 - 98.4]
	Positive percent agreement	100% (67/67)	[94.6 - 100]
	Negative percent agreement	92.4% (61/66)	[83.5 - 96.7]

IHC: Immunohistochemistry; PCR: Polymerase chain reaction; CI: Confidence interval.