

Product Insert

Set Cat No: HD825

Batch: 28695

Template: 6135-03 USR-01 (V-02)

EGFR Multiplex cfDNA Reference Standard Set

Product Handling:

Horizon Diagnostics recommends quantifying the material using preferred laboratory method and instrumentation prior to use.

Origin of Material:

- Horizon Diagnostics uses a proprietary genome engineering platform to precisely engineer mutations within human cancer cell lines.
- The cell line DNA is fragmented to an average fragment length of 160 bp which resembles cfDNA extracted from plasma.
- Additional information can be found on Product Specification Sheet (6135 PSS) on the website for this catalogue number.

Allelic Frequency Verification Data:

- Data is provided in Table 1-4. These values represent only the QC assays performed by Horizon using an input of **100 ng** and are an average of 4 replicates (WT and Mutant copy values are rounded to whole numbers, with the exception of EGFR Multiplex wild-type Mutant copies which are reported to 1 decimal place).

Table 1: 5% EGFR Multiplex cfDNA Reference Standard (Part No.: HD821)

Gene	Variant	Amplicon size (bp)	Expected AF (%)	WT Conc (copies/μl)	Mutant Conc (copies/μl)	Total Conc (copies/μl)
EGFR	T790M	94	5.0	2140	130	2270
EGFR	L858R	78	5.0	2824	158	2982
EGFR	ΔE746-A750	95†/95	5.0	2036	122	2158
EGFR	L861Q	78	5.0	2776	158	2934
EGFR	G719S	95	5.0	1900	114	2014
EGFR	C797S	62	5.0	3416	179	3595
EGFR	S464L	105	5.0	1668	98	1766
EGFR	G465R	96	5.0	2068	126	2194
EGFR	V769-D770insASV	122/131	5.0	1312	74	1386
EGFR	S768I	64	5.0	3232	198	3430

Table 2: 1% EGFR Multiplex cfDNA Reference Standard (Part No.: HD822)

Gene	Variant	Amplicon size (bp)	Expected AF (%)	WT Conc (copies/μl)	Mutant Conc (copies/μl)	Total Conc (copies/μl)
EGFR	T790M	94	1.0	2180	28	2208
EGFR	L858R	78	1.0	2980	34	3014
EGFR	ΔE746-A750	95†/95	1.0	2155	25	2180
EGFR	L861Q	78	1.0	3036	38	3074
EGFR	G719S	95	1.0	1992	26	2018
EGFR	C797S	62	1.0	3520	41	3561
EGFR	S464L	105	1.0	1868	20	1888
EGFR	G465R	96	1.0	2292	28	2320
EGFR	V769-D770insASV	122/131	1.0	1548	18	1566
EGFR	S768I	64	1.0	3388	42	3430

Table 3: 0.1% EGFR Multiplex cfDNA Reference Standard (Part No.: HD823)

Gene	Variant	Amplicon size (bp)	Expected AF (%)	WT Conc (copies/μl)	Mutant Conc (copies/μl)	Total Conc (copies/μl)
EGFR	T790M	94	0.1	2284	3	2287
EGFR	L858R	78	0.1	2804	2	2806
EGFR	ΔE746-A750	110/95	0.1	1760	2	1762
EGFR	L861Q	78	0.1	2860	3	2863
EGFR	G719S	95	0.1	1984	2	1986
EGFR	C797S	62	0.1	3480	4	3484
EGFR	S464L	105	0.1	1756	2	1758
EGFR	G465R	96	0.1	2236	2	2238
EGFR	V769-D770insASV	122/131	0.1	1524	1	1525
EGFR	S768I	64	0.1	3292	4	3296

Table 4: EGFR Multiplex wild type cfDNA Reference Standard (Part No.: HD824)

Gene	Variant	Amplicon size (bp)	Expected AF (%)	WT Conc (copies/μl)	Mutant Conc (copies/μl)	Total Conc (copies/μl)
EGFR	T790M	94	0	2356	0.6	2357
EGFR	L858R	78	0	2832	0.0	2832
EGFR	ΔE746-A750	110/95	0	1788	0.0	1788
EGFR	L861Q	78	0	2940	0.0	2940
EGFR	G719S	95	0	2008	0.2	2008
EGFR	C797S	62	0	3488	0.0	3488
EGFR	S464L	105	0	1820	0.1	1820
EGFR	G465R	96	0	2328	0.1	2328
EGFR	V769-D770insASV	122/131	0	1560	0.0	1560
EGFR	S768I	64	0	3392	0.1	3392

† EGFR wild-type concentration was calculated using a similar amplicon-size WT probe in this particular assay

Table 1-4: Allelic frequency and copy number data collected by Droplet Digital™ PCR. Other endogenous variants are present, however only the data presented is tested within each batch.