

Technical Product Report

For Research Use Only; Not for use in Diagnostic Procedures

Product Description: Seraseq® ctDNA Reference Material v2 AF0.5%

Material Number: 0710-0205 Batch Number: 10410228

Material Description: A ctDNA-like mixture of human genomic DNA from the reference cell line, GM24385, and synthetic DNA constructs

Concentration
(Qubit dsDNA BR Assay): 33.8 ng/mL

Volume: 5.0 mL

Date of Manufacture: 03 Jun 2019 Expiration Date: 03 Jun 2022

Storage: 2-8°C

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Digital PCR testing using
BioRad QX200™ Droplet
Digital™ PCR System:

| Gene ID | COSMIC Identifier | AF% |
|-----------|------------------------|-------------------------|
| AKT1 | COSM33765 | 0.53 |
| APC | COSM13127 | 0.50 |
| APC | COSM18561 | 0.43 |
| ATM | COSM21924 | 0.59 |
| BRAF | COSM476 | 0.54 |
| CTNNB1 | COSM5664 | 0.63 |
| EGFR | COSM6224 | 0.63 |
| EGFR | COSM12378 | 0.54 |
| EGFR | COSM6225 | 0.56 |
| EGFR | COSM6240 | 0.54 |
| ERBB2 | COSM20959 | 0.53 |
| FGFR3 | COSM715 | 0.62 |
| FLT3 | COSM783 | 0.51 |
| FOXL2 | COSM33661 | 0.54 |
| GNA11 | COSM52969 | 0.69 |
| GNAQ | COSM28758 | 0.52 |
| GNAS | COSM27887 | 0.58 |
| IDH1 | COSM28747 | 0.75 |
| JAK2 | COSM12600 | 0.48 |
| KIT | COSM1314 | 0.60 |
| KRAS | COSM521 | 0.53 |
| MPL | COSM18918 | 0.59 |
| NCOA4-RET | N/A | 0.54 |
| NPM1 | COSM17559 | 0.38 |
| NRAS | COSM584 | 0.54 |
| PDGFRA | COSM736 | 0.53 |
| PDGFRA | COSM28053 | 0.55 |
| PIK3CA | COSM763 | 0.45 |
| PIK3CA | COSM12464 ² | 0.47 |
| PIK3CA | COSM775 | 0.60 |
| PTEN | COSM4986 | 0.34 |
| PTEN | COSM5809 | 0.34 |
| RET | COSM965 | 0.55 |
| SMAD4 | COSM14105 | 0.53 |
| TP53 | COSM10648 | 0.39 |
| TP53 | COSM10660 | 0.56 |
| TP53 | COSM10662 | 0.59 |
| TP53 | COSM6530 | 0.42 |
| TP53 | COSM18610 | Not Tested ¹ |
| TPR-ALK | N/A | 0.56 |

¹ NGS testing alone is used for QC of variant COSM18610.

² As of June 2019 this mutation is no longer listed in the COSMIC database.

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| Gene | COSMIC Identifier | AF% |
|--------|------------------------|------|
| AKT1 | COSM33765 | 0.75 |
| BRAF | COSM476 | 0.36 |
| CTNNB1 | COSM5664 | 0.35 |
| EGFR | COSM6225 | 0.63 |
| EGFR | COSM12378 | 0.37 |
| EGFR | COSM6240 | 0.52 |
| EGFR | COSM6224 | 0.47 |
| ERBB2 | COSM20959 | 0.38 |
| IDH1 | COSM28747 | 0.34 |
| KIT | COSM1314 | 0.62 |
| KRAS | COSM521 | 0.58 |
| NRAS | COSM584 | 0.83 |
| PDGFRA | COSM28053 | 0.57 |
| PDGFRA | COSM736 | 0.57 |
| PIK3CA | COSM763 | 0.61 |
| PIK3CA | COSM775 | 0.52 |
| PIK3CA | COSM12464 ¹ | 0.44 |
| RET | COSM965 | 0.62 |
| TP53 | COSM10660 | 0.36 |
| TP53 | COSM10662 | 0.35 |
| TP53 | COSM6530 | 0.28 |
| TP53 | COSM10648 | 0.55 |
| TP53 | COSM18610 | 0.57 |

Next Generation Sequencing testing using Archer® Reveal ctDNA™ 28 Kit run on an Illumina® MiSeq™ using v2 (2x150 bp) PE chemistry reagents:

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NGS was performed as an orthogonal verification step. Parameters used:

NGS Parameters:

DNA input = 50 ng

of samples / flow cell = 3

of total reads / sample = 5.9 M

Average read depth = 6870X

On-target reads = ~94%

Q30 score = ~93%

Analysis = Archer Analysis Suite v5.1.7

Default analysis settings except for the following:

- ERROR_CORRECTION is turned to ON (default=OFF; cfDNA pipeline)
- MAPQ_THRESHOLD_FOR_VARIANT_CALL is set to 10 (default=0)
- MIN_ALLELE_FRACTION_FOR_VARIANT_CALL is set to 0.00025 (default=0.001)
- MIN_BASEQUAL_FOR_VARIANT_CALL is set to 30 (default=20)
- NORMALIZATION_LEVEL is set to 10000000 (default is 3500000; cfDNA pipeline)
- VARIANT_DOWNSTREAM_ROI_SIZE is set to 150 (default=400; cfDNA pipeline)

Please see the poster from NIST for more information about assay sensitivity:

<https://digital.seracare.com/multilab-assessment-reference-materials-ctdna-poster2018>

Approval:



Prepared By

13 Jun 2019

Date