

Batch Certificate For Research Use Only

PRODUCT INFORMATION AND QUALITY CONTROL

NAME OF PRODUCT	cfDNA (human) AF: 0% Ashkenazim Son
DESCRIPTION	cfDNA (human) AF: 0% Ashkenazim Son in highly characterized human DNA from cell lines.
CATALOG NUMBER	SID-000003
BATCH NUMBER	00042
MANUFACTURING CONDITIONS	<ul style="list-style-type: none"> • Manufactured and sealed in class 2 safety cabinet • At room temperature
PACKAGE SIZE AND TYPE	<ul style="list-style-type: none"> • 2D barcoded tube with screw cap • Material: Polypropylen (PP)
DATE OF MANUFACTURE	26.02.2020
EXPIRY DATE	25.02.2022
CONCENTRATION	20 ng/µl (dsDNA)
QUANTITY	400 ng (dsDNA)
NOMINAL VOLUME	25 µl; (488 ng)
MUTATION	AKT1 p.E17K (COSM33765*, COSV62571334*, substitution, c.49G>A, Exon 2) BRAF p.V600E (COSM476*, COSV56056643*, substitution, c.1799T>A, Exon 15) ERBB2 p.E770_A771insAYVM (new: p.Y772_A775dup) (COSM20959*/ COSM404915*, COSV54062409*, insertion, c.2313_2324dup/ c.2310_2311ins12, Exon 19) KRAS p.G12D (COSM521*, COSV55497369*, substitution, c.35G>A, Exon 1) KRAS p.Q61K (COSM549*, COSV55502066*, substitution, c.181C>A, Exon 2) KRAS p.A146T (COSM19404*, COSV55501778*, substitution, c.436G>A, Exon 3) PIK3CA p.C420R (COSM757*, COSV55874020* substitution, c.1258T>C, Exon 7) PIK3CA p.E542K (COSM760*, COSV55873227*, substitution, c.1624G>A, Exon 9) PIK3CA p.E545A (COSM12458*, COSV55873209*, substitution, c.1634A>C, Exon 9) PIK3CA p.E545D (COSM765*, COSV55874040*, substitution, c.1635G>T, Exon 9) PIK3CA p.E545G (COSM764*, COSV55873220*, substitution, c.1634A>G, Exon 9) PIK3CA p.E545K (COSM763*, COSV55873239* substitution, c.1633G>A, Exon 9) PIK3CA p.Q546E (COSM6147*, COSV55882350* substitution, c.1636C>G, Exon 9) PIK3CA p.Q546R (COSM12459*, COSV55876869* substitution, c.1637A>G, Exon 9) PIK3CA p.H1047L (COSM776*, COSV55873401* substitution, c.3140A>T, Exon 20) PIK3CA p.H1047R (COSM775*, COSV55873195*, substitution, c.3140A>G, Exon 20) PIK3CA p.H1047Y (COSM774*, COSV55876499* substitution, c.3139C>T, Exon 20) <small>* GRCh38 COSMIC v90</small>
ALLELE FREQUENCY	0%

<p>QUALITY</p> <p>STORAGE CONDITIONS</p> <p>MANUFACTURING AND QUALITY CONTROL SITES</p>	<p>DNA quantity metrologically traceable to internationally certified reference material¹</p> <p>The copy number values are metrologically traceable to the natural units count 1 and ratio 1 and International System of Units (SI) derived units of volume.</p> <p>+ 2-8 °C</p> <p>SensID GmbH</p> <p>Schillingallee 68, 18057 Rostock, Germany</p>																																	
<p>TEST METHOD AND ACCEPTANCE CRITERIA</p>	<p>Quality Control</p>	<p>Test Method</p>	<p>Acceptance criteria</p>																															
	<p>Fragmentation</p>	<p>Fragment Length Analysis Agilent High Sensitivity DNA Kit (Agilent Technologies)</p>	<p>peak size 167 bp ± 10% (151 bp – 181 bp)</p>																															
	<p>Quantification</p>	<p>Total DNA measurement: Spectrophotometry ssDNA [ng/μl] = (A260-A320)*38^{2,3}</p> <p>dsDNA measurement: Qubit dsDNA BR Assay Kit (Invitrogen)</p>	<p>Total DNA: n.a.⁴</p> <p>dsDNA: 17.5 – 22.5 ng/μl</p>																															
	<p>Allele Frequency</p>	<p>ddPCR Analysis using BioRad QX200™ System</p>	<p>AF 0.00% (≤0.03%)⁵</p>																															
<p>RESULTS OF ANALYSIS</p>	<table border="1"> <thead> <tr> <th></th> <th>Result</th> <th>PASS/FAIL</th> </tr> </thead> <tbody> <tr> <td>Fragmentation</td> <td>181 bp</td> <td>PASS</td> </tr> <tr> <td>Quantity</td> <td>29.1 ng/μl (total DNA) 19.5 ng/μl (dsDNA)</td> <td>PASS</td> </tr> <tr> <td rowspan="8">Allele Frequency</td> <td>Mutation</td> <td>AF in %</td> </tr> <tr> <td>AKT1 E17K</td> <td>0.00</td> </tr> <tr> <td>BRAF V600E</td> <td>0.00</td> </tr> <tr> <td>ERBB2 E770_A771insAYVM (Y772_A775dup)</td> <td>0.00</td> </tr> <tr> <td>KRAS G12D</td> <td>0.00</td> </tr> <tr> <td>KRAS Q61K</td> <td>0.00</td> </tr> <tr> <td>KRAS A146T</td> <td>0.00</td> </tr> <tr> <td>PIK3CA C420R</td> <td>0.00</td> </tr> <tr> <td>PIK3CA E542K</td> <td>0.00</td> </tr> <tr> <td></td> <td>PASS/FAIL</td> <td></td> </tr> </tbody> </table>				Result	PASS/FAIL	Fragmentation	181 bp	PASS	Quantity	29.1 ng/μl (total DNA) 19.5 ng/μl (dsDNA)	PASS	Allele Frequency	Mutation	AF in %	AKT1 E17K	0.00	BRAF V600E	0.00	ERBB2 E770_A771insAYVM (Y772_A775dup)	0.00	KRAS G12D	0.00	KRAS Q61K	0.00	KRAS A146T	0.00	PIK3CA C420R	0.00	PIK3CA E542K	0.00		PASS/FAIL	
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¹ ERM_AD442K

² Protocol NK603 – Community Reference Laboratory for GM Food and Feed

³ Measured before filling in product tube

⁴ not applicable

⁵ With the exception of PIK3CA E545A where it is ≤0.7%

		PIK3CA E545A ⁶	0.58	PASS
		PIK3CA E545D	0.00	PASS
		PIK3CA E545G	0.00	PASS
		PIK3CA E545K	0.00	PASS
		PIK3CA Q546E	0.00	PASS
		PIK3CA Q546R	0.00	PASS
		PIK3CA H1047L	0.00	PASS
		PIK3CA H1047R	0.00	PASS
		PIK3CA H1047Y	0.03	PASS

COMMENTS/REMARKS

Additional information:

Copy numbers (CN) of the respective measurements

Mutation	CN wt ⁷ /μl	CN mut ⁸ /μl
AKT1 E17K	2059	0
BRAF V600E	2030	0
ERBB2 E770_A771insAYVM (Y772_A775dup)	3047	0
KRAS G12D	2782	0
KRAS Q61K	3206	0
KRAS A146T	3638	0
PIK3CA C420R	1749	0
PIK3CA E542K	4033	0
PIK3CA E545A ⁶	3964	23
PIK3CA E545D	2717	0
PIK3CA E545G	4118	0
PIK3CA E545K	2366	0
PIK3CA Q546E	4801	0
PIK3CA Q546R	4494	0
PIK3CA H1047L	3294	0
PIK3CA H1047R	3550	0
PIK3CA H1047Y	3646	1

Table 1 indicates the values of the QC assays performed by SensID GmbH with an DNA input of ~20 ng. The value for the respective mutation results from the mean value of five-eight measured replicates (CN values are rounded). CN concentration values per microliter (μl), are based on droplet digital (ddPCR) assay counts dilution factors, and droplet volume measurements. The detection of the amount of CNs may vary depending on the assay used. Therefore, due to assay properties, there may be deviations in the observed number of copies and allele frequencies compared to the values given here.

Name and position/title of Person authorising the batch release:

Mr. Björn Nowack

Date of batch release: 24.03.2020

Signature batch release: Björn Nowack

This document was created electronically and is valid without a signature.

⁶ A BLAST sequence analysis shows 98% homology of PIK3CA E545A mutation sequence to genome locus Homo sapiens chromosome 22, GRCh38.p13. Therefore, a higher false positive rate is expected and measured, most likely due to a cross reaction of gene probe to genome locus Homo sapiens chromosome 22, GRCh38.p13.

⁷ Wild Type

⁸ Mutation