

## 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 is highly characterized human DNA from cell line
CATALOG NUMBER	SID-000003
BATCH NUMBER	00145
MANUFACTURING CONDITIONS	<ul style="list-style-type: none"> <li>· Manufactured und sealed in class 2 safety cabinet</li> <li>· At room temperature</li> <li>· Manufactured according to DIN EN ISO 13485:2016</li> </ul>
PACKAGE SIZE AND TYPE	<ul style="list-style-type: none"> <li>· 2D barcoded tube with screw cap</li> <li>· Material: Polypropylen (PP)</li> </ul>
DATE OF MANUFACTURE	20.01.2021
EXPIRY DATE	19.01.2023
TARGET CONCENTRATION	20 ng/µl (dsDNA)
TARGET QUANTITY	400 ng (dsDNA)
NOMINAL VOLUME	25 µl
MUTATION * GRCh38 COSMIC v91	AKT1 p.E17K (COSV62571334*, substitution, c.49G>A, Exon 2) BRAF p.V600E (COSV56056643*, substitution, c.1799T>A, Exon 15) ERBB2 p.E770_A771insAYVM (new: p.Y772_A775dup) (COSV54062409*, insertion, c.2313_2324dup, Exon 19) KRAS p.G12D (COSV55497369*, substitution, c.35G>A, Exon 1) KRAS p.Q61K (COSV55502066*, substitution, c.181C>A, Exon 2) KRAS p.A146T (COSV55501778*, substitution, c.436G>A, Exon 3) PIK3CA p.C420R (COSV55874020* substitution, c.1258T>C, Exon 7) PIK3CA p.E542K (COSV55873227*, substitution, c.1624G>A, Exon 9) PIK3CA p.E545A (COSV55873209*, substitution, c.1634A>C, Exon 9) PIK3CA p.E545D (COSV55874040*, substitution, c.1635G>T, Exon 9) PIK3CA p.E545G (COSV55873220*, substitution, c.1634A>G, Exon 9) PIK3CA p.E545K (COSV55873239* substitution, c.1633G>A, Exon 9) PIK3CA p.Q546E (COSV55882350* substitution, c.1636C>G, Exon 9) PIK3CA p.Q546R (COSV55876869* substitution, c.1637A>G, Exon 9) PIK3CA p.H1047L (COSV55873401* substitution, c.3140A>T, Exon 20) PIK3CA p.H1047R (COSV55873195*, substitution, c.3140A>G, Exon 20) PIK3CA p.H1047Y (COSV55876499* substitution, c.3139C>T, Exon 20) p.G719S (COSV51767289*, substitution, c.2155G>A, Exon 18) p.E746_A750delELREA (COSV51765066*, deletion, c.2236_2250del15, Exon 19) p.S752_1759delSPKANKEI (COSV51774879*, deletion, c.2254_2277del24, Exon 19) p.S768I (COSV51768106* substitution, c.2303G>T, Exon 20) p.V769_D770insASV (new: p.A767_V769dup) (COSV51850427* Insertion, c.2303_2304insTGTGGCCAG, Exon 20) p.T790M (COSV51765492*, substitution, c.2369C>T, Exon 20) p.L858R (COSV51765161*, substitution, c.2573T>G, Exon 21) p.L861Q (COSV51766344*, substitution, c.2582T>A, Exon 21)
ALLELE FREQUENCY	0%

QUALITY	DNA quantity metrologically traceable to internationally certified reference material (ERM_AD442K). 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.			
STORAGE CONDITIONS	+ 2-8 °C			
MANUFACTURING SITE	SensID GmbH Schillingallee 68, 18057 Rostock, Germany			
TEST METHOD AND ACCEPTANCE CRITERIA	Quality control	Test method		Acceptance criteria
	Fragmentation	Fragment length analysis Agilent High Sensitivity DNA Kit (Agilent Technologies)		Peak size 167 bp ± 10% (151 bp – 181 bp)
	Quantification	Total DNA measurement (ssDNA): Spectrophotometry** dsDNA measurement: Qubit dsDNA BR Assay Kit (Invitrogen)		Total DNA: not applicable dsDNA: 17.5 – 22.5 ng/µl
	**Protocol NK603 – Community Reference Laboratory for GM Food and Feed; Measured before filling in product tube			
	Allele frequency	Allele frequency analysis ddPCR (BioRad QX200™)		AF 0.0% (0.00–0.03%, except for PIK3CA E545A: ≤0.7%)
RESULTS OF ANALYSIS	Quality control	Result		PASS / FAIL
	Fragmentation	173 bp		PASS
	Quantification	35 ng/µl (total DNA)		PASS
		20,5 ng/µl (dsDNA)		
	Allele frequency	Mutation	AF in %	PASS / FAIL
		AKT1 p.E17K	0,00	PASS
		BRAF p.V600E	0,00	PASS
		ERBB2	0,00	PASS
		KRAS p.G12D	0,00	PASS
		KRAS p.Q61K	0,00	PASS
		KRAS p.A146T	0,00	PASS
		PIK3CA p.C420R	0,00	PASS
		PIK3CA p.E542K	0,00	PASS
		PIK3CA p.E545A***	0,70	PASS
		PIK3CA p.E545D	0,00	PASS
		PIK3CA p.E545G	0,02	PASS
		PIK3CA p.E545K	0,00	PASS
PIK3CA p.Q546E		0,00	PASS	
PIK3CA p.Q546R		0,00	PASS	
PIK3CA p.H1047L	0,00	PASS		
PIK3CA p.H1047R	0,02	PASS		
PIK3CA p.H1047Y	0,00	PASS		
p.G719S	0,00	PASS		
p.E746_A750delELREA	0,00	PASS		
p.S752_I759delSPKANKEI	0,00	PASS		
p.S768I	0,00	PASS		
p.V769_D770insASV	0,00	PASS		
p.T790M	0,00	PASS		
p.L858R	0,00	PASS		
p.L861Q	0,02	PASS		

\*\*\*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.

COMMENTS / REMARKS	Additional information: Measurement of copy number		
	Mutation	CN wt/ $\mu$ l	CN mut/ $\mu$ l
MEASUREMENT OF COPY NUMBER	AKT1 p.E17K	2079	0
	BRAF p.V600E	1750	0
	ERBB2 p.E770_A771insAYVM	2918	0
	KRAS p.G12D	2687	0
	KRAS p.Q61K	3167	0
	KRAS p.A146T	3738	0
	PIK3CA p.C420R	2058	0
	PIK3CA p.E542K	4557	0
	PIK3CA p.E545A	4884	36
	PIK3CA p.E545D	3169	0
	PIK3CA p.E545G	4392	1
	PIK3CA p.E545K	2448	0
	PIK3CA p.Q546E	5297	0
	PIK3CA p.Q546R	5494	0
	PIK3CA p.H1047L	3990	0
	PIK3CA p.H1047R	3925	1
	PIK3CA p.H1047Y	3991	0
	p.G719S	4236	0
	p.E746_A750delELREA	3974	0
	p.S752_I759delSPKANKEI	2572	0
	p.S768I	3173	0
	p.V769_D770insASV	3438	0
	p.T790M	3669	0
	p.L858R	3698	0
	p.L861Q	4917	1
	wt: wildtype; mut: mutation		
<p><i>The table above indicates the values of the QC assays performed by SensID GmbH with a DNA input of ~20 ng. The value for the respective mutation results from the mean value of QC samples according to ISO 2859-1:2014-08 (CN values are rounded). CN concentration values per microliter (<math>\mu</math>l) 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.</i></p>			

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

Björn Nowack, Managing Director

Date of batch release: 12.02.2021

Signature batch release: Björn Nowack

This document has been created electronically and is valid without signature.