

PIK3CA-11 mutations 12.5% AF cfDNA (SID-000099) – Instructions for use

For Research Use Only

SensID Bringing Precision to Molecular Diagnostics

Every diagnostic test as well as R&D needs references and controls. SensID GmbH manufactures High Quality Reference Materials / Controls for Molecular Diagnostics. Our mission is to provide certified standards ready for your needs in the highest quality to ease your processes. For more information visit www.sens-id.com

Content

Vial	Description	Catalog No.
1	PIK3CA-C420R + PIK3CA-E545A	SID-000099
2	PIK3CA-Q546R + PIK3CA-E542K	
3	PIK3CA-H1047L + PIK3CA-E545D	
4	PIK3CA-H1047R + PIK3CA-E545G	
5	PIK3CA-H1047Y + PIK3CA-E545K	
6	PIK3CA-Q564E	
7	Wildtype (Ashkenazim son cfDNA Cat. No.: SID-000003)	

Symbols

	Catalog number
	Lot number
	Use by
	Legal manufacturer
	Not for reuse
	Temperature limitations
	GHS07: Harmful

Storage

The product should be stored at 2°C to 8°C upon arrival. DO NOT FREEZE. The product is solved in TE buffer (Tris-EDTA (10 mM Tris, 1mM EDTA), pH 8,0, and stable until the expiration date when stored under these conditions.

Intended Use

The product contains a precisely defined allele frequency as indicated on the corresponding certificate. It was designed for use in **liquid biopsy** with the **intended application**:

1. For spike-in experiments
2. As comparative probe for validation of processes for the verification of 11 PIK3CA mutations
3. Control in workflow verification / validation
4. Validation and development of targeted sequencing protocols (amplicon sequencing) and PCR protocols
5. Analyze the performance of your amplicon-based NGS pipeline (including capture-based assays) by comparing to freely available datasets

Protocol: PIK3CA-11 mutations 12.5% AF cfDNA

Important point before starting:

- It is recommended to centrifuge SID-000099 briefly to avoid liquid holding back in the lid of the vial!
- To avoid contaminations in the vial work in clean environment (e.g. laminar flow hood)
- Mix by pipetting up and down 10 times to obtain a homogeneous suspension. Do not vortex!
- No further purification or DNA isolation steps needed
- DNA purified from a reference cell line, GM24385
- The purified DNA is present in cfDNA (human) at a fragment size of 167 bp $\pm$ 10%
- While the presence and frequency of each mutation and/or amplification in this product is evaluated during manufacture using ddPCR assay, there may be differences in observed allele frequencies due to specific assay characteristics

Technical Assistance

Our Technical Service Assistance is staffed by experienced scientists with extensive practical and theoretical expertise with our products. If you have any questions or experience any difficulties regarding the particular product or SensID GmbH products in general, please do not hesitate to contact us.

SensID GmbH customers are a major source of information regarding advanced or specialized uses of our products. This information is helpful to other scientists as well as to the researchers at SensID GmbH. We therefore encourage you to contact us if you have any suggestions about product performance or new applications and techniques. For technical assistance and more information, please see our Website www.sens-id.com or call one of the SensID GmbH Technical Service Assistance.

Product Use limitations

Attention should be paid to expiration dates and storage conditions printed on the box and labels of all components. Do not use expired or incorrectly stored components. Check primary packaging before first opening. Do not use products from damaged primary packaging.

Quality Control

In accordance with SensID's Quality Management System, each lot of PIK3CA-11 mutations 12.5% AF cfDNA is tested against predetermined specifications to ensure consistent product quality.

The product should appear as a clear liquid. Alterations in this appearance may indicate instability or deterioration of the product and vials should be discarded.

Warnings and precautions

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, please consult the appropriate safety data sheets (SDSs). These are available online in convenient and compact PDF format at www.sens-id.com, where you can find, view, and print the SDS for each SensID GmbH products, kit component and other products.

Avoid contamination of the product when opening and closing the vial.

CAUTION: Handle as though it is capable of transmitting infectious agents. This product is formulated using the cell line GM24385, which is a B-lymphocytic, male cell line from the Personal Genome Project offered by the NIGMS Human Genetic Cell Repository (<https://catalog.coriell.org/1/NIGMS>).

Equipment and Reagents to Be Supplied by User

- Pipets (adjustable)¹
- Sterile pipet tips with filters

¹ Ensure that instruments have been checked and calibrated according to the manufacturer's recommendations.

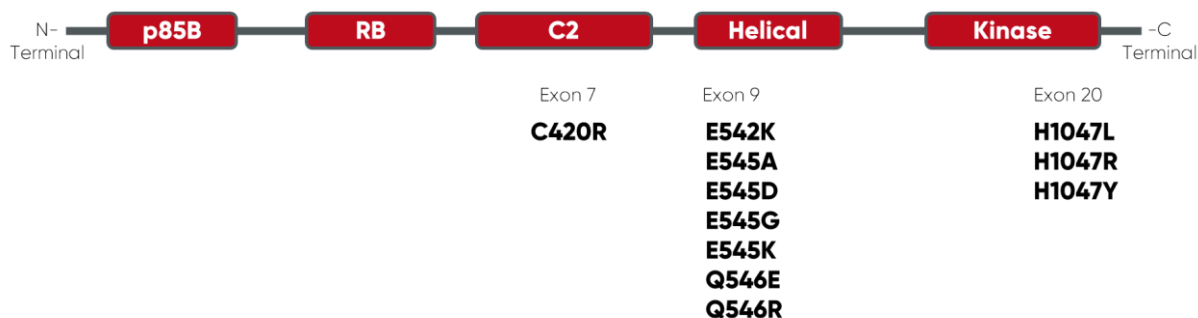


Table 1 General information about PIK3CA. Taken from <https://www.ncbi.nlm.nih.gov/gene/5290>.

Official Symbol	PIK3CA
Official Full Name	phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit
Organism	<i>Homo sapiens</i>
Also known as	MCM; CWS5; MCAP; PI3K; CLAPO; CLOVE; MCMTC; PI3K- α ; p110- α
Summary	Phosphatidylinositol 3-kinase is composed of an 85 kDa regulatory subunit and a 110 kDa catalytic subunit. The protein encoded by this gene represents the catalytic subunit, which uses ATP to phosphorylate PtdIns, PtdIns4P and PtdIns(4,5)P ₂ . This gene has been found to be oncogenic and has been implicated in cervical cancers. A pseudogene of this gene has been defined on chromosome 22. [provided by RefSeq, Apr 2016]

Table 2 Mutations present in the SensID PIK3CA-11 mutations 12.5% AF cfDNA reference material. HGVS = Human Genome Variation Society; * = GRCh38 · COSMIC v91

Gene	Legacy Identifier	Genomic Mutation ID	Type of mutation	HGVS Nomenclature	Localisation in Genome (GRCh38)	Amino acid change
PIK3CA	COSM757*	COSV55874020*	Substitution Missense	c.1258T>C	3:179210192..179210192 Exon 7	C420R
	COSM760*	COSV55873227*	Substitution Missense	c.1624G>A	3:179218294..179218294 Exon 9	E542K
	COSM12458*	COSV55873209*	Substitution Missense	c.1634A>C	3:179218304..179218304 Exon 9	E545A
	COSM765*	COSV55874040*	Substitution Missense	c.1635G>T	3:179218305..179218305 Exon 9	E545D
	COSM764*	COSV55873220*	Substitution Missense	c.1634A>G	3:179218304..179218304 Exon 9	E545G
	COSM763*	COSV55873239*	Substitution Missense	c.1633G>A	3:179218303..179218303 Exon 9	E545K
	COSM6147*	COSV55882350*	Substitution Missense	c.1636C>G	3:179218306..179218306 Exon 9	Q546E
	COSM12459*	COSV55876869*	Substitution Missense	c.1637A>G	3:179218307..179218307 Exon 9	Q546R
	COSM776*	COSV55873401*	Substitution Missense	c.3140A>T	3:179234297..179234297 Exon 20	H1047L
	COSM775*	COSV55873195*	Substitution Missense	c.3140A>G	3:179234297..179234297 Exon 20	H1047R
	COSM774*	COSV55876499*	Substitution Missense	c.3139C>T	3:179234296..179234296 Exon 20	H1047Y

Figure 1 Schematic overview of PIK3CA mutations in different Exons of PIK3CA gene included in the product. It is reported that PIK3CA mutations which have been localized mostly on exon 9 and 20 hot spots are detected 25–40 % in Breast Cancer².² <https://www.ncbi.nlm.nih.gov/pubmed/26921096>