Seraseq™

Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material Package Insert

PLEASE NOTE:
THESE REAGENTS MUST NOT BE SUBSTITUTED FOR THE MANDATORY POSITIVE AND NEGATIVE CONTROL REAGENTS PROVIDED WITH MANUFACTURED TEST KITS.

NAME AND INTENDED USE
The Seraseq™ Circulating Tumor DNA-I (AF1.2) Reference Material is formulated for use with targeted Next Generation Sequencing (NGS) assays, real-time quantitative PCR (qPCR) or digital PCR (dPCR) assays that screen for circulating tumor DNA (ctDNA) in a cell-free plasma sample. The Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material is intended as a quality material for translational oncology researchers, assay developers, and clinical pathology laboratories, and monitors sample purification, library preparation, and sequencing (if using an NGS-based assay), and detection performance. *For Research Use Only. Not for use in diagnostic procedures.*

SUMMARY
A well-designed quality control program provides added confidence in the reliability of results obtained for unknown specimens. The use of independent reference products may provide valuable information concerning assay sensitivity and bioinformatics pipeline analysis.

PRINCIPLES OF THE PROCEDURE
Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material is ready to use in NGS, real-time qPCR or dPCR assays starting with DNA extraction, similar to an actual test sample. The Reference Material contains processed human genomic DNA formulated in a commutable matrix (simulated plasma) that is compatible with varying NGS target enrichment and sequencing or digital PCR-based detection methods following DNA purification.

REAGENTS
Item No. 0710-0014*. 1 vial, 5 mL per vial, 12 ng/mL concentration (60ng extractable nucleic acid per vial).

* Patent Pending

WARNINGS AND PRECAUTIONS
For Research Use Only. Not for use in diagnostic procedures.
CAUTION: Handle Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material and all materials derived from human blood products as though they are capable of transmitting infectious agents. Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material is manufactured using processed human genomic DNA and biosynthetic mutant sequence. Purified genomic DNA mixture is formulated in a commutable matrix (simulated plasma) containing human protein isolates.

Safety Precautions
Use Center for Disease Control and Prevention (CDC) recommended universal precautions for handling reference materials and human specimens[1]. Do not pipette by mouth; do not smoke, eat, or drink in areas where specimens are being handled. Clean any spillage by immediately wiping up with 0.5% sodium hypochlorite solution. Dispose of all specimens and materials used in testing as though they contain infectious agents.

Handling Precautions
Do not use Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material beyond the expiration date. Avoid contamination of the product when opening and closing the vials.

STORAGE INSTRUCTIONS
Store Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material refrigerated at 2 – 8 ºC. Do not freeze. Samples are designed to be single use.

INDICATIONS OF REAGENT INSTABILITY OR DETERIORATION
Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material is a mixture of human genomic and biosynthetic DNA formulated in a commutable matrix (simulated plasma). It should appear as a clear to pale yellow liquid. Alterations in this appearance may indicate instability or deterioration of the product and vials should be discarded.

PROCEDURE
Materials Provided
Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material is a mixture of human genomic DNA in a commutable matrix (simulated plasma). Five (5) mL is provided per tube.

Materials Required but not Provided
Refer to instructions supplied by manufacturers of the test kits to be used.

Instructions for Use
Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material may be inserted into workflows in a manner consistent with plasma fractions prior to extractions. Mix by vortexing to ensure a homogeneous solution. Do not centrifuge. Following extraction, Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material must go through the entire library preparation and sequencing or real-time qPCR or alternatively digital PCR steps in parallel with the test specimens. Refer to your usual assay procedures in order to determine the amount of material to use. Each vial is intended for a single-use.

Quality Control
Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material does not have assigned values for variant allele frequencies. However, the product is formulated using digital PCR quantitation to target each variant listed in Table 1 to be present at 1.2%. There are many reasons why assays may observe deviation from this target, which may or may not be of significance. It is therefore recommended that each laboratory qualify the use of each lot of Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material with each assay system prior to its routine use.

INTERPRETATION OF RESULTS
Detection of variants and the variant allele frequency may vary with different NGS targeted sequencing-based cancer panels, different real-time qPCR or different digital PCR assays, and different test reagent lots. Since the reference material does not have an assigned value, the laboratory must establish an acceptable range for each lot of Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material. When results for the product are outside of the established acceptance range, it may indicate unsatisfactory test performance. Possible sources of error include: deterioration of test kit reagents, operator error, faulty performance of equipment, contamination of reagents, or change in bioinformatics pipeline parameters.

LIMITATIONS OF THE PROCEDURE
Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material MUST NOT BE SUBSTITUTED FOR THE CONTROL REAGENTS PROVIDED WITH MANUFACTURED TEST KITS.
TEST PROCEDURES provided by manufacturers must be followed closely. Deviations from procedures recommended by test kit manufacturers may produce unreliable results. Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material is not a calibrator and should not be used for assay calibration. These materials are also not patient-like, whole process reference materials and are not suitable to optimize the methods used for specimen extraction.

Adverse shipping and storage conditions or use of outdated product may produce erroneous results.

seracare™
508.244.6400
800.676.1881
info@seracare.com
SeraCare.com

March 2016 13050-01

EXPECTED RESULTS

Specific detection of cancer variants and variant allele frequencies will vary among different assays, different procedures, different lot numbers, and different laboratories. Each laboratory should establish its own range of acceptable values. For example, the acceptable range for each variant might include all values within two standard deviations of the mean of 20 data points obtained in 20 runs[2]. Table 1 lists mutations that are present in the product. Note that the GM24385 human cell line contains a heterozygous HRAS mutation (COSM249860) and heterozygous KIT mutation (COSM28026) that may be detected (depending on the assay utilized) at approximately 50%.

SPECIFIC PERFORMANCE CHARACTERISTICS

Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material has been designed for use with targeted NGS, real-time qPCR or dPCR assays for the purpose of assessing assay characteristics. The product is manufactured from mixture of human genomic and biosynthetic DNA formulated in a commutable matrix (simulated plasma). Although the product is formulated with a 1.2% minor allele frequency as determined by digital PCR, Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material does not have assigned values. Procedures for implementing a quality assurance program and monitoring test performance on a routine basis must be established by each individual laboratory.

REFERENCES

1. Siegel JD, Rhinehart E, Jackson M, Chiarello L, and the Healthcare Infection Control Practices Advisory Committee, 2007 Guideline for Isolation Precautions: Preventing Trasnmission of Infectious Agents in Healthcare Settings.

2. Statistical Quality Control for Quantitative Measurements: Principles and Definitions; Approved Guideline-Second Edition. NCCLS document C24-A2, 1999.

TABLE 1: Seraseq Circulating Tumor DNA-I (AF1.2) Reference Material Mutations

	Gene	COSMIC ID of Mutation	Position (hg19)	CDS	Mutation Type	Amino Acid Change	Target VAF
1	BRAF	COSM476	140453136	c.1799T>A	SNV	p.V600E	1.2%
2	EGFR	COSM6240	55249071	c.2369C>T	SNV	p.T790M	1.2%
3	EGFR	COSM12378	55249013	c.2310_2311insGGT	Small Insertion	p.D770_N771insG	1.2%
4	EGFR	COSM6225	55242465	c.2236_2250del15	Large Deletion	p.E746_A750delELREA	1.2%
5	KIT	COSM1314	55599321	c.2447A>T	SNV	p.D816V	1.2%
6	KRAS	COSM521	25398284	c.35G>A	SNV	p.G12D	1.2%
7	NRAS	COSM584	115256529	c.182A>G	SNV	p.Q61R	1.2%
8	PIK3CA	COSM775	178952085	c.3140A>G	SNV	p.H1047R	1.2%
9	PIK3CA	COSM12464	178952150	c.3204_3205insA	Insertion	p.N1068fs*4	1.2%