Seraseq®

MSI Reference Panel Mix AF5% and AF20% 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® MSI Reference Panel AF5% and AF20% products are formulated for use with Polymerase Chain Reaction (PCR) as well as Next Generation Sequencing (NGS) assays that detect microsatellite instability in human cancer patient samples by analyzing the lengths of the regions commonly referred to as BAT-25, BAT-26, NR-21, NR-24, and MONO-27. This product is intended for use as a reference material for analysis of microsatellite instability in MSI assays under a given set of bioinformatics pipeline parameters. Product is *For Research Use Only. Not for use in diagnostic procedures.*

REAGENTS

Table 1. Seraseq MSI Products

Material No.	Product
0710-1675	Seraseq® MSI Reference Panel Mix AF5%
0710-1676	Seraseq® MSI Reference Panel Mix AF20%

WARNINGS AND PRECAUTIONS

For Research Use Only. Not for use in diagnostic procedures.
CAUTION: Handle Seraseq® MSI Reference Panel AF5% and AF20% products as though they are capable of transmitting infectious agents. These products are formulated using genomic DNA from GM24385 human cell line , which is a B-lymphocytic, male cell line from the Genome in a Bottle (GIAB) Project.

Safety Precautions

Use Centers 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 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® MSI Reference Panel AF5% and AF20% products beyond their expiration dates. Avoid contamination of the product when opening and closing the vial.

STORAGE INSTRUCTIONS

Store Seraseq® MSI Reference Panel AF5% and AF20% products frozen at -20°C or colder. Aliquoting of the product into low DNA binding tubes may be advisable to limit the number of freeze-thaw cycles.

PROCEDURE

Materials Provided

Seraseq® MSI Reference Panel AF5% and AF20% products are packaged as tumor-normal matched pair and consists of genomic DNA from a reference cell line, GM24385 blended with plasmid constructs containing specific microsatellite instability markers at defined allelic frequencies. The matched normal, consisting of purified DNA from GM24385 WT cell line, is included in the kit. The purified DNA is present in a 1 mM Tris, 0.1 mM EDTA pH 8.0 aqueous buffer, at a concentration of 20 ng/µL and a fill volume of 15 µL. Material is ready to use in PCR or NGS assays in steps that follow DNA isolation. No further purification or DNA isolation is needed.

Materials Required but not Provided

Refer to instructions supplied by manufacturers of the extraction kit to be used.

Instructions for Use

Thaw the product vial on ice. Mix by vortexing to ensure a homogenous solution and spin briefly. Seraseq® MSI Reference Panel AF5% and AF20% may be input directly into library preparation following procedures used for clinical specimens. Refer to your usual assay procedures in order to determine the amount of material to use.

EXPECTED RESULTS & INTERPRETATION OF RESULTS

Table 2. Genomic coordinates of the 5 MSI mononucleotide markers in the Seraseq® MSI Reference Panel AF5% and AF20% products.

Marker	Gene	Chromosome	Position (hg19 based)	Comment
BAT-25	KIT (intron16)	chr4	55598211	25T -> 19T
BAT-26	MSH2 (intron5)	chr2	47641559	27A -> 17A
NR-21	SLC7A8 (5'UTR)	chr14	23652346	21A -> 13A
NR-24	ZNF2 (3'UTR)	chr2	95849361	23T -> 17T
MONO-27[1]	MAP4K3 (intron3) MAP4K3 (intron13)	chr2	39573062 39536689	27A -> 21A

[1]There is ambiguity in the literature on the MONO-27 locus so two constructs are included in the product to ensure compatibility (see References 2 & 3).

Table 2 lists the genomic coordinates of the microsatellite instability (MSI) markers in the Seraseq® MSI Reference Panel AF5% and AF20% products. Detection of all 5 biomarkers at the blended AF levels may differ across different PCR and/or NGS assays and different test reagent lots. While the presence and frequency of each MSI marker in this product is confirmed during manufacture using both droplet digital PCR (ddPCR) and other PCR based methods (see **Table 3**), there may be differences in observed allele frequencies due to assay characteristics. Each laboratory must establish an assay-specific expected value for each MSI marker and lot of the Seraseq® MSI Reference Panel AF5% and AF20%. 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 changes in bioinformatics pipeline parameters. Additional support documents are available online at www.seracare.com/oncology.

SeraCare Life Sciences, Inc. | 37 Birch Street, Milford, MA 01757 USA
Phone: +1 508.244.6400 | Toll Free (US Only) 800.676.1818
info@seracare.com | www.seracare.com

13738-02 August 2022

LIMITATIONS OF THE PROCEDURE

Seraseq® MSI Reference Panel AF5% and AF20% 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. This product is offered for Research Use Only. Not for use in diagnostic procedures. Data are provided for informational purposes. SeraCare Life Sciences does not claim that others can duplicate test results exactly. Seraseq® MSI Reference Panel AF5% and AF20% is not a calibrator and should not be used for assay calibration. This material is not whole-process control and does not evaluate the method used for specimen extraction. Adverse shipping and/or storage conditions or use of outdated product may produce erroneous results.

REFERENCES

1. Siegel JD, Rhinehart E, Jackson M, Chiarello L, and the Healthcare Infection Control Practices Advisory Committee, 2007 Guideline for Isolation Precautions: Preventing Transmission of Infectious Agents in Healthcare Settings.
2. Bacher J, Halberg R, Kent-First M, Wood KV. “Methods and kits for detecting mutations” US Patent US20090068646A1 issued March 12, 2009.
3. Pino MS, Chung DC. “Application of molecular diagnostics for the detection of Lynch syndrome.” Expert review of molecular diagnostics vol. 10,5 (2010): 651-65. doi:10.1586/erm.10.45.

Table 3. Representative PCR analysis of Seraseq® MSI Reference Panel Mix AF5% and AF20%, and comparison to matched normal (WT).

MSI Marker	Bio-Rad Laboratories dPCR MSI assays			Promega’s MSI Analysis System v1.2		
	AF5%	AF20%	WT (0%)	AF5%	AF20%	WT (0%)
BAT-25	4.1	19.1	0.0	Unstable	Unstable	Stable
BAT-26	4.9	17.9	0.0	Unstable	Unstable	Stable
NR-21	4.3	18.5	0.0	Stable	Unstable	Stable
NR-24	4.2	18.2	0.0	Unstable	Unstable	Stable
MONO-27	5.1	19.9	0.2	Unstable	Unstable	Stable

SeraCare Life Sciences, Inc. | 37 Birch Street, Milford, MA 01757 USA
Phone: +1 508.244.6400 | Toll Free (US Only) 800.676.1818
info@seracare.com | www.seracare.com