CRITICAL DIAGNOSTICS

3030 Bunker Hill St. Suite 117A
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Presage® ST2 Control Kit

REF# BC-1066E

Advancing Medicine, Saving Lives®

Presage® ST2 Control Kit Instructions for Use

Intended Use

The Presage ST2 Controls Kit is designed for use with the Presage™ ST2 Assay Kit which is a monoclonal antibody sandwich ELISA in a 96 well microtiter plate format. The controls are provided in a sealed, lyophilized vial format. Following reconstitution in the specified volume of deionized water the controls will be analyzed using the same protocol as a clinical test sample.

Introduction

Using standard PCR techniques a human cDNA clone for ST2, GeneBank accession number NM 003856, was used as the source sequence to create an expression vector including the entire human soluble ST2 (sST2) sequence with a histidine purification tag incorporated in the amino terminus region of the protein. Integrity of the expression clone was confirmed by DNA sequencing. Recombinant protein is produced by transient transfection and expression in human embryonic kidney cells (HEK293). Recombinant ST2 protein is purified by passing cell lysate over a metal chelate column specifically binding the histidine purification tag incorporated into the expressed protein. Purification of recombinant ST2 is confirmed by Coomasie stained PAGE gel to verify the expected molecular weight of 58 kDa. Quantification is performed by Bradford protein assay and concentrations are normalized using a proprietary ST2 reference stock.

The base matrix for the Presage sST2 Assay kit controls is delipidized human serum with sodium azide (0.09%) added as a preservative. There are no known effects on the assay from this matrix. Further, the control is produced at a concentration such that for use it will be diluted 50-fold prior to analysis, exactly the same procedure used for clinical test samples, which may be EDTA plasma, heparin plasma or serum.

Control Concentrations

Controls are produced at two (2) concentrations; one in the normal reference range and one in the concentration range of individuals with cardiac disease, such as acute heart failure (AHF). The median ST2 concentration determined in the reference interval analysis was 18.8 ng/mL and the 97.5th percentile was 45.6 ng/mL. The low concentration control is prepared to have a final concentration in the range bounded by the median value, 18.8 ng/mL, and an upper bound ≤35 ng/mL, which is approximately the 85th percentile of normal. Patients with AHF have a median sST2 concentration of 65.3 ng/mL with a 75th percentile at 105 ng/mL. The high concentration control is prepared to have a final concentration in the range bounded by these values with a working objective concentration of ≤85 ng/mL. The concentration range of 65.3 to 105 ng/mL represents both an analytically effective and a clinically significant concentration. Each lot of controls is provided with specific assigned target values and QC ranges that are printed on the corresponding Certificate of Analysis sheet.

Table 1: ST2 Concentrations at Specific Thresholds Self-Reported Healthy Cohort

Parameter	sST2 (ng/mL)	95% CI
median	18.8	18.1 to 19.9
75th percentile	25.3	23.8 to 27.0
80th percentile	27.8	25.5 to 29.5
90th percentile	34.3	32.2 to 35.7
95th percentile	37.9	35.6 to 41.6
97.5th percentile	45.6	39.5 to 48.8

Reagents and Material Provided:

1. ST2 Low Control (1 vial)
2. ST2 High Control (1 vial)
3. Instruction for Use

Materials Required But Not Provided:

1. Precision Pipettes: 5 µl, 100 µl, and 1.0 ml
2. Disposable pipette tips

Warnings and Precautions

These controls are intended solely for in vitro diagnostic use with the Presage® ST2 Assay Kit.

These controls contain human serum derived material and should be handled as though capable of transmitting infectious disease. Each serum, plasma or whole blood unit used in the manufacture of this material was tested and found to be negative for anitibodies to HIV, HCV and nonreactive for HBsAg. Because no test method can completely assure that infectious agents are absent, this material should be handled as though capable of transmitting infectious disease, and disposed as biohazardous waste according to local regulations.

This product contains less than 0.1% sodium azide that may react with lead or copper plumbing to form potentially explosive metal azides. On disposal, flush with a large volume of water to prevent azide build-up.

Storage Conditions:

Sealed vials are stable for 1 year when stored at refrigerated temperatures (2-8°C). Vials stored after reconstitution are stable for a maximum of seven(7) days when properly stored at refrigerated temperatures (2-8°C).

Reagent Preparation and Use:

1. Remove vial from package, reconstitute with the volume of deionized water specified in the control lot Certificate of Analysis, and gently swirl occasionally for 30 minutes. Do not shake. Do not mix mechanically.

2. Refer to the Presage® ST2 Assay Kit instructions for specimen testing instructions, and run the control material like any other specimen.

3. After use, replace the stopper and store at 2-8C to obtain the maximum 7 day reconstituted stability.

4. It is recommended that controls be included (in duplicate) with each assay performed.

Expected Results

When evaluating an individual assay, the standard curve should be similar to the example provided in the Instruction for Use that accompany the Presage ST2 Assay Kit, and results from both controls should be within the assigned QC ranges. If the either of these conditions is not satisfied the user should consider repeating the assay and evaluating potential reasons for unexpected performance.

Additional Information

For additional information or questions call Critical Diagnostics at 1-877-700-1250 ext. 3, or 1-858-270-2400.

Precision

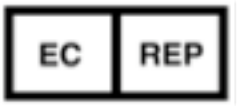
A set of three (3) randomly selected vials of each level were reconstituted and tested per the standard protocol procedure using three (3) assay kits. The results are summarized in table 2. Each concentration and each lot meets the specification of ≤10% CV.

Table 2: Results from Control Lot 1 (R23-33A and R23-33B)

	R23-33A			R23-33B		
	Ave ST2	SD	CV	Ave ST2	SD	CV
plate 1	24.8	1.04	4.2%	74.0	3.09	4.2%
plate 2	24.1	0.89	3.7%	72.3	1.91	2.6%
plate 3	25.1	0.63	2.5%	74.0	3.37	4.6%
cumulative	24.7	0.94	3.8%	73.4	2.90	3.9%

Interferences

The matrix reagent, delipidized human serum, is tested for endogenous ST2 concentration prior to formulation of the specific controls. Formulation of the individual control concentrations is then performed by adding a calculated quantity of recombinant ST2 to the matrix reagent to achieve the desired final concentration. There are no known interferences or other effects on the assay from this matrix.



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