



03/21

SCL-70 IgG ELISA Kit

(Catalog # E4980-100; 96 assay; Store at 4°C)

I. Introduction:

DNA Topoisomerase I is an important nuclear enzyme that unwinds the double-stranded DNA, relaxes, and reanneals the strand. Initially, the enzyme is synthesized as a 100 kDa protein. However, this precursor undergoes proteolytic cleavage to yield a 70 kDa form, which is also known as SCL-70. Antibodies to SCL-70 antigen are detected in 20% of the patients suffering from diffused scleroderma, and in 10% of the patients suffering from systemic scleroderma (CREST Syndrome). The presence of SCL-70 antibodies has also been reported in patients suffering from systemic lupus erythematosus (SLE). BioVision's SCL-70 IgG (Human) ELISA kit quantitatively measures SCL-70 IgG antibodies in human serum and plasma samples. Samples, calibrator, and controls are added to the wells pre-coated with SCL-70 antigen. IgG antibodies to SCL-70, if present in the samples, will bind to the SCL-70 antigen. The wells are then washed with Wash Buffer, followed by incubation with the enzyme conjugate. After incubation, any unattached conjugates are washed off by Wash Buffer. The enzymatic reaction is detected by the addition of TMB-substrate. Finally, the reaction is terminated with an acidic stop solution. The color developed is directly proportional to the amount of IgG antibodies to SCL-70 present in the sample.

II. Features and Benefits:

Easy, convenient and time-saving method to measure the amount of SCL-70 IgG in serum and plasma samples No significant cross-reactivity or interference with other autoantibodies was observed

III. Sample Type:

Serum, Plasma

IV. Kit Contents:

Components	E4980-100	Part Number
Micro ELISA plate coated with SCL-70 antigen	8 x 12 Strips	E4980-100-1
Sample Diluent	22 ml	E4980-100-2
Calibrator	1 ml	E4980-100-3
Positive Control	1 ml	E4980-100-4
Negative Control	1 ml	E4980-100-5
Enzyme Conjugate	12 ml	E4980-100-6
TMB Substrate	12 ml	E4980-100-7
Stop Solution	12 ml	E4980-100-8
Wash Buffer (20X)	25 ml	E4980-100-9

V. User Supplied Reagents and Equipment:

- Microplate reader capable of measuring absorbance at 450 nm
- · Distilled or deionized water

VI. Storage Conditions and Reagent Preparation:

- The entire kit may be stored at 4°C.
- Wash buffer (1X): Prepare Wash buffer (1X) by adding 25 ml of Wash Buffer (20X) to 475 ml of distilled or deionized water. Store
 at room temperature (20-25 °C).
- Prepare reagents within 30 minutes before the experiment. Before using the kit, spin tubes and bring down all components to the bottom of tubes.
- Samples may be refrigerated at 2–8 °C for up to seven days or frozen for up to six months. Avoid repetitive freezing and thawing.
 Lipemic or hemolyzed samples may cause erroneous results.

VII. Assay Protocol:

Note: Bring all reagents and samples to room temperature 30 minutes prior to the assay. It is recommended that all standards and samples be run at least in duplicate. A standard curve must be run with each assay.

- 1. Place the desired number of coated strips into the holder.
- 2. Negative Control, Positive Control, and Calibrator are ready to use. Prepare 1:21 dilution of test samples, by adding 10 μl of the sample to 200 μl of Sample Diluent and mix well.
- 3. Dispense 100 µl of diluted serum, Calibrator and Controls into the appropriate wells. Add 100 µl Sample Diluent in well for black control. Tap the plate to remove air bubbles from the liquid and mix well. Incubate for 20 minutes at room temperature.
- 4. Aspirate liquid from all wells. Wash wells three times with 300 µl of Wash Buffer (1X). Blot on absorbance paper or paper towel.
- 5. Dispense 100 µl of Enzyme Conjugate to each well and incubate for 20 minutes at room temperature.
- Remove Enzyme Conjugate from all wells. Wash wells three times with 300 μl of Wash Buffer (1X). Blot on absorbance paper or paper towel.
- 7. Dispense 100 µl of TMB substrate and incubate for 10 minutes at room temperature.
- 8. Add 100 µl of stop solution.
- Read O.D. at 450 nm using ELISA reader within 15 min. A dual wavelength is recommended with reference filter of 600-650 nm.

VIII. Calculation:



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Check **Calibrator Factor (CF)** value on the calibrator bottle. This value might vary from lot to lot.

Calculate the cut-off value: **Calibrator OD x Calibrator Factor (CF)**.

Calculate the **Ab (Antibody) Index** of each determination by dividing the O.D. value of each sample by cut-off value.

IX. Related Products:

SSB IgG (Human) ELISA Kit (E4978) MOG (Human) ELISA Kit (E4963) Complement C3 (Human) ELISA Kit (E4976) SSA IgG (Human) ELISA Kit (E4979) GAD1 (Human) ELISA Kit (E4962)

FOR RESEARCH USE ONLY! Not to be used on humans.