



Stanbio RaPET® s-LE Procedure No. 1160

Latex Agglutination Slide Test for the Qualitative and Semi-Quantitative Determination of Anti-DNP Associated with Systemic Lupus Erythematosis in Human Serum

Summary and Principle¹⁻⁷

In s-LE, autoantibodies directed against native deoxyribonucleic acid and other nuclear constituents are produced. It is classed as the prototype of severe autoimmune diseases, involving a variety of tissues and associated with a wide range of antibodies in the circulation. Characteristics of the disease are antibodies against native DNA, nucleoprotein, denatured DNA and other extractable nuclear antigens. S-LE also affects a wide range of tissue. Organs affected are, in decreasing incidence, joints, skin, kidney, central nervous system, heart and lungs. One other important feature is the high frequency of the disease in women, approximately 3 to 4 times more frequent than in men. The high incidence of s-LE between monozygous twins (70-80%) and of close relatives (5-10%) indicates that s-LE may be a hereditary disease.

Anti-DNP antibodies are demonstrated by a number of laboratory procedures which include the LE cell test, immunofluorescence, and agglutination of coated latex particles. In the STANBIO RaPET® s-LE, latex particles are bound with native deoxyribonucleic protein (DNP) by means of an intermediary albumin matrix. These coated latex particles combine with anti-DNP antibodies in serum to give a visible agglutination.

Reagents

RaPET sLE Latex Reagent (white cap), Ref. No. 1161

Suspension of polystyrene latex particles coated with DNP extract in a saline buffer.

sLE Positive Control (red cap), Ref. No. 1162

Stabilized s-LE positive human serum.

Negative Control (green cap), Ref. No. 1192

Stabilized human serum, negative for s-LE.

Precautions: *For In Vitro Diagnostic Use.*

sLE control sera have been tested and found nonreactive for Hepatitis B Surface Antigen (HBsAg) and HTLV-111; however, all human serum products and patient specimens should be considered potentially hazardous and handled in the same manner as an infectious agent. Reagents contain sodium azide. This agent is known to react with copper and lead in sink drains to form explosive azides. Disposed materials should be flushed with large quantities of water to prevent azide accumulation.

Reagent Storage and Stability: Reagents are stable at 2-8°C until the expiration date shown on the label. DO NOT FREEZE!

Materials Provided

Plastic slide

Disposable pipette/mixers

Materials Required But Not Provided

Timer

Test tubes (Titration only)

Serological Pipettes (Titration only)

Physiological saline (0.9% NaCl) (Titration only)

Specimen Collection and Preparation¹¹

It is recommended that serum only be used. Do not heat inactivate test sera or controls. Avoid repeated freeze-thawing of specimens. Do not use visibly hemolyzed specimens, as these have been known to produce false-positive results.

Sample Stability: Serum is stable for 48 hours when stored at 2-8°C.

Interfering Substances: Use only a clean, dry slide washed in mild detergent and rinsed with distilled water.

Patients with Rheumatoid Arthritis, Sjogren Syndrome, Mixed Connective Tissue Disease, Progressive Systemic Sclerosis and Discoid L.E. may show reactivity when using this test. Many widely used drugs may, in fact, induce a systemic lupus erythematosis syndrome. Hydralazine, isoniazid, procainomide and a number of anti-convulsant drugs fall into this category.

Latex reagent **must** be shaken vigorously for 30 seconds before use in order to avoid aggregation of latex particles.

Procedure

- Bring all reagents to room temperature and mix gently prior to use.
- Place in separate divisions (cells) of the same slide:

Serum	1 drop
Positive Control (red cap)	1 drop
Negative Control (green cap)	1 drop
- Add one drop of sLE latex reagent to each cell.
- Mix with flat end of pipette/mixer and spread fluid evenly over each cell.
- Tilt the slide back and forth slowly for 3 minutes while observing for agglutination.

Semi-quantitative Test

Prepare dilutions of the specimens as shown below:

Dilution	
1:2	(1 part serum + 1 part saline)
1:4	(1 part serum + 3 parts saline)
1:8	(1 part serum + 7 parts saline)
1:16	(1 part serum + 15 parts saline)
1:32	(1 part serum + 31 parts saline)
1:64	(1 part serum + 63 parts saline)

Proceed as in screening test.

The serum s-LE antibody titer is the highest dilution of serum showing agglutination of the latex reagent within 3 minutes after mixing.

Quality Control: Positive and Negative controls should be run with each series and results compared with patient specimens.

Results

Any degree of agglutination visible within 3 minutes is to be interpreted as positive. Test is considered s-LE negative when no difference in agglutination is observed between specimen and negative control.

Performance Characteristics

The s-LE latex test was compared with a standard LE cell preparation test as well as a fluorescent ANA test. The three tests showed excellent agreement on serum from clinically active sLE patients: s-LE latex 82% positive, LE cell prep 86% positive, ANA test 82% positive. Serum from clinically inactive s-LE patients: positive reactions were s-LE latex 19%, ANA test 71%. Patients with connective tissue disease showed no positive reactions with the s-LE latex test, but 17% and 50% positive reactions with the LE cell prep and ANA test, respectively.

Additional published studies have confirmed the sensitivity and specificity of the s-LE latex test.

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