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Diagnostic value of the Avitex-SLE latex test for screening and Diagnosis of Systemic Lupus Erythematosus (SLE) compared to the LE cell preparation test

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Objective : *To study the diagnostic value of the Avitex-SLE latex test for screening and Diagnosis of Systemic Lupus Erythematosus (SLE), compared to the LE cell preparation test*

Setting : *King Chulalongkorn Memorial Hospital*

Design : *Prospective study*

Subjects : *70 patients with suspected Systemic Lupus Erythematosus (SLE) assessed from November to December 1999*

Methods : *Patients' un-coagulated blood samples were analyzed by both the Avitex-SLE latex test and the LE cell preparation test, and the sensitivity, specificity, positive predictive value, negative predictive value and accuracy of both tests were calculated.*

Results : *Eight patients were shown to have Systemic Lupus Erythematosus (SLE) by the standard criteria of diagnosis. The sensitivity and specificity, positive predictive value and negative predictive value of the Avitex-SLE latex test were all 100 % and 100 %, 100 % and 100 %, while the sensitivity and*

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specificity, positive predictive value and negative predictive value of the LE cell preparation test were 62.50 %,100 %, 100 % and 95.38 %, respectively. The accuracy of the Avitex-SLE and LE cell tests were 100 % and 95.71 %, respectively.

Conclusion : *According to our results the Avitex-SLE latex test is more sensitive for the diagnosis of SLE than the LE cell preparation test. We conclude that this test is useful as a SLE rapid screening test in patients with suspected SLE.*

Key words : *Systemic Lupus Erythematosus(SLE),Latex test, LE cell, Test.*

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วัตถุประสงค์ : *เพื่อศึกษาการตรวจคัดกรองและ Systemic Lupus Erythematosus ด้วยชุดตรวจแอนติบอดี้ Avitex-SLE latex test เปรียบเทียบกับวิธี LE cell preparation test*

สถานที่ที่ทำการศึกษา : *โรงพยาบาลจุฬาลงกรณ์*

รูปแบบการศึกษา : *การศึกษาแบบไปข้างหน้า*

ประชากรที่ศึกษา : *ผู้ป่วยที่สงสัยว่าจะเป็นโรค Systemic Lupus Erythematosus ในระหว่างเดือนพฤศจิกายนถึงเดือนธันวาคม 2542*

วิธีการศึกษาและวัดผล : *ทำการตรวจเลือดผู้ป่วยด้วยชุดตรวจแอนติบอดี้ Avitex-SLE latex test และวิธี LE cell preparation และคำนวณเปรียบเทียบค่า sensitivity, specificity, positive predictive value, negative predictive value และ accuracy ของทั้ง 2 วิธี*

ผลการศึกษา : *จากการศึกษาในผู้ป่วยทั้งหมด 70 ราย พบว่าเป็นผู้ป่วยโรค Systemic Lupus Erythematosus ตามเกณฑ์การวินิจฉัยมาตรฐาน 8 ราย จากการศึกษาพบว่าชุดตรวจแอนติบอดี้ Avitex-SLE latex test มีค่า sensitivity, specificity, positive predictive value และ negative predictive value เท่ากับ ร้อยละ 100, ร้อยละ 100, ร้อยละ 100 และ ร้อยละ 100 ตามลำดับ และวิธี LE cell preparation test มีค่า sensitivity, specificity, positive predictive value และ negative predictive value เท่ากับร้อยละ 62.50 %, ร้อยละ 100, ร้อยละ 100 และ ร้อยละ 95.38 % ตามลำดับ ค่า accuracy ของ Avitex-SLE test และการตรวจหา LE cell เท่ากับร้อยละ 100 และร้อยละ 95.71 % ตามลำดับ*

วิจารณ์และสรุปผล : *ชุดตรวจแอนติบอดี้ Avitex-SLE latex test มีความไวในการตรวจคัดกรองและวินิจฉัยโรค Systemic Lupus Erythematosus มากกว่าการตรวจหา LE cell ดังนั้นจากผลการศึกษานี้คณะผู้วิจัยสรุปว่า ชุดตรวจ Avitex-SLE latex test สามารถใช้ในการคัดกรองโรค Systemic Lupus Erythematosus*

Systemic Lupus Erythematosus (SLE) is a multisystem chronic inflammatory disease of unknown etiology which has a strong predilection for young women in the reproductive years, affects many organs, and is characterized by the production of multiple autoantibodies, typically antinuclear and anti-DNA antibodies. In some patients, autoantibodies are also produced against platelets, lymphocytes, and other cellular antigens.[1] Most of these autoantibodies are not specific for SLE and might be produced non-specifically as a result of polyclonal B cell activation.

The diagnosis of SLE is based upon the appropriate clinical features and is confirmed by serological, pathological, and other laboratory findings. The fluorescent antinuclear antibody (FANA/ANA) test is the most widely used diagnostic screening test for SLE.

Commonly used tests in the diagnosis of SLE are [2-6]

1. The anti-nuclear antibody test (ANA): a test to determine if autoantibodies to cell nuclei are present in the blood.

2. The anti-DNA antibody test: to determine if the patient has antibodies to the genetic material of the cell.

3. The anti-Sm antibody test: to determine if there are antibodies to Sm, which is a protein found in the cell nucleus.

4. Tests to detect the presence of immune complexes in the blood.

5. Tests to examine the total level of serum complement – a group of proteins which can occur in immune reactions – and tests to assess the specific level of C3 and C4, two proteins of this group.

6. LE cell preparation (LE cell prep) or LE cell test: An examination of the blood looking for a certain kind of cell which has ingested the swollen antibody-coated nucleus of another cell. This test is used less frequently than the ANA test, because the ANA is more sensitive for SLE than the LE cell preparation

The Lupus erythematosus (LE) cell factor is an antibody which reacts towards deoxyribonucleoprotein (DNP). This test was the first laboratory test devised to screen for SLE, but is less sensitive than another test available now,the ANA test.[7-8] The LE cell preparation test is positive in about 40 - 50 % of patients with SLE, and also positive in 80 % of patients with active untreated SLE. Unfortunately, the test can also be positive in up to 20 % of patients with rheumatoid arthritis, in some patients with other rheumatic conditions like Sjogren's syndrome or scleroderma, in patients with liver disease, and in persons taking certain drugs (such as procainamide, hydralazine,[9] and others)

The Avitex-SLE latex test is a rapid latex agglutination test kit for the presumptive detection of the Systemic Lupus Erythematosus (SLE) in human serum by the detection and quantitation of serum antinucleoprotein (DNP) factors. The Avitex-SLE latex particles are coated with DNP, and when the latex suspension is mixed with serum containing antinuclear antibodies, a clear agglutination will be seen within 1 minute. The antibodies detected are those directed against deoxyribonucleoprotein (DNP), which are most commonly associated with SLE. These antibodies are supposed to be the cause of the formation of the LE cell in vitro. If this test is an effective screen for SLE, it will benefit patients since the diagnosis can be achieved easily and rapidly. The objective of this study was to determine the diagnostic accuracy of the

Avitex-SLE latex test in SLE and non-SLE patients compared to the LE cell preparation test.

Materials and Methods

Patients

Seventy patients in King Chulalongkorn Memorial Hospital, from November to December 1999 who were suspected of having SLE, based on clinical findings were included in this study. All patients were examined and investigated following the criteria of diagnosis of SLE, and uncoagulated blood samples were collected to perform the study.

LE cell preparation test

The basis of this in-vitro test is that when serum containing antibodies with anti DNA histone specificity reacts with damaged (antibody permeable) cell nuclei, the nuclei swell and form basophilic round bodies (LE bodies) which are phagocytized by PMNs in the reaction mixture, thus forming typical inclusion body-containing LE cells.

The procedure for performing an LE test in brief is as follows: 10 ml. of non-anticoagulated blood are drawn, the sample is allowed to clot and the serum separated from the clot. The clot is forced through a fine screen to disrupt cell membranes and release nuclear material. The serum is added to the preparation and incubated. If the serum contains antibodies against nuclear proteins the antibodies will attach to nuclear proteins released by the cell damage. Neutrophils that were not ruptured will phagocytize the antigen - antibody complexes. The resulting cell is the LE cell. The cytologist must see several LE cells to call the test positive.

Avitex-SLE latex test

The Avitex-SLE latex particles are coated with DNP. When the latex suspension is mixed with serum containing antinuclear antibodies, a clear agglutination is seen within 1 minute.

The procedure for performing Avitex-SLE latex test and the interpretation in brief are as follows:

1. Allow test reagents and sera to reach room temperature.

2. Transfer one drop of patient serum to a test circle on the slide.

3. Shake the latex reagent vigorously, then, using the dropper provided, add one drop of reagent to the test circle.

4. Mix the drops using a disposable stirrer ensuring coverage of the test circle with the mixture.

5. Gently and evenly, rock and rotate the test slide for 1 minute whilst examining the test slide for agglutination.

6. Examine the test slide under a strong light source after 1 minute. A positive result is indicated by the obvious agglutination pattern of the latex in a clear solution. A negative result is indicated by no change in the latex suspension on the test slide.

Approximately 1 % of healthy individuals can give positive reactions in SLE latex tests. This figure is considerably less than the ANA procedure performed on the same population (6 %). Patients with scleroderma, rheumatoid arthritis, dermatomyositis and a variety of connective tissue disorders may show reactivity in the SLE latex test.

Statistical analysis

The sensitivity, specificity, positive predictive

value, negative predictive value and accuracy of the Avitex-SLE latex test and the LE cell preparation test were calculated for 1) all patients and 2) SLE patients alone.

Results

There were men and women, mean age was 34.50 years (range 16 - 68). According to this study, 8 patients met the diagnostic criteria of SLE (11.43 %). The criteria for gold standard diagnosis of positive cases [10,11] for SLE are shown in table 1. The results of the study are tabulated in table 2 and 3. The sensitivity (TP/TP+FN), specificity (TN/TN+FP), positive predictive value (PPV) (TP/TP+FP) and negative predictive value (NPV) (TN/TN+FN) of the Avitex-SLE latex test were all 100 %, while those of the LE cell test were 62.50 %, 100 %, 100 % and 95.38 %, respectively. The accuracy [(TP+ TN) /(TP + TN + FP+ FN) x 100] of the Avitex-SLE test and LE cell preparation tests were 100 % and 95.71 %, respectively.

Discussion and Conclusion

Systemic lupus erythematosus (SLE) is an autoimmune disease where autoantibodies are frequently targeted against intracellular antigens of the cell nucleus, histones, and extractable nuclear antigens (ENAs). An ideal test for SLE would be specific, sensitive, have a high positive predictive value (PPV) and high negative predictive value (NPV). Furthermore, assay results may reflect disease activity, correlated with organ involvement, or predict relapse, thus allowing pre-emptive treatment. At present, no test or test panel can perform all these tasks. The LE

Table 1. The American Rheumatology Association (ARA) criteria for diagnosis of SLE.

ARA criteria 1982 (updated 1997)	Detail
Photosensitivity	Photosensitive skin rash
Malar rash	Flat or raised fixed erythema
Discoid rash	Raised with plugging/scarring/scaling
Oral ulcers	Usually painless
Arthritis	Non-erosive, 2+ peripheral joints
Serositis	Pleural or cardiac
Renal disorder	Proteinuria or cellular casts
Neurological disorder	Convulsions or psychosis without other cause
Hematological disorder	Hemolysis, cytopenia
"Immunological disorder" (modified 1997)	Anti-ds DNA, anti-Sm, antiphospholipid antibodies (ACAs, LA, or FP VDRL)
Antinuclear antibody (ANA)	"Abnormal titre" ANA at any time point by IIF or equivalent assay

ACA, anticardiolipin antibody; ANA, antinuclear antigen; dsDNA, double stranded DNA; FP VDRL, false positive venereal disease reference laboratory test; IIF, immunofluorescence; LA, lupus anticoagulant.

Table 2. Diagnostic activity of Avitex-SLE latex test.

	SLE patients	Non-SLE patients	Total
Positive Avitex-SLE	8	0	8
Negative Avitex-SLE	0	62	62
Total	8	62	70

Sensitivity	= 100 %	Specificity	= 100 %
Positive predictive value	= 100 %	Negative predictive value	= 100 %
Accuracy	= 100 %		

Table 3. Diagnostic activity of LE cell preparation test.

	SLE patients	Non-SLE patients	Total
Positive LE cell	5	0	5
Negative LE cell	3	62	65
Total	8	62	70

Sensitivity	= 62.50 %	Specificity	= 100 %
Positive predictive value	= 100 %	Negative predictive value	= 95.38 %
Accuracy	= 95.71 %		

cell preparation test is a diagnostic test for (SLE) that is based on an in vitro immunologic reaction between the patient's autoantibodies to nuclear antigens and damaged nuclei in the testing medium. It is subject to numerous experimental variables and dependent on subjective interpretation. However, this test is no longer done in most laboratories, having been superseded by the more sensitive fluorescent ANA test or its equivalent. Avitex-SLE latex test is a test kit using rapid latex agglutination technique to detect serum antinucleoprotein factors the antibodies directed against deoxyribonucleoprotein (DNP), which are most frequently associated with SLE. This test is easy-to-use and takes only one minute, by contrast, the LE cell preparation test consumes more time and repuires more specialized skill to perform. In a previous study by Edmund L.[12], the SLE Latex test kit was found to be quite specific when the test is used for excluding the diagnosis of SLE disease. In this study, the results showed that the Avitex-SLE latex test is more sensitive in screening for SLE than the LE cell preparation test.

According to this study, we concluded that the Avitex-SLE latex is a useful rapid screening technique in patients with suspected SLE.

Discussion

However, the number of patients in this study was still small and might effect the results of sensitivity, specificity and accuracy of this study. Additional studies will be useful to strengthen the diagnostic value of this test.

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