Anti-Nuclear Antibody Test System

TEST LIMITATIONS
1. No diagnosis should be based upon a single ANA test result, since various host factors must be taken into consideration.
2. Among these host factors are age and sex. There is an increasing significance in positive ANA results in both males and females as age increases. Normal females between 20-60 have a 7% incidence of ANA: normal males, a 4% incidence. Both sexes over 80 years of age have a 50% incidence of ANA.
3. Various medications including antibiotics, tranquilizers, aspirin and birth control pills can induce a lupus like condition resulting in high ANA titres. Drug-induced Lupus generally goes into a sustained clinical remission following removal of the triggering medication.
4. Various autoimmune processes induce positive ANA tests.
5. Further evidence for diagnosis of SLE is provided by low complement levels, particularly C1, C3 and C4.
6. ANA tests may not agree with LE Prep tests or with latex tests.
7. Presence of antibodies to double stranded native DNA is diagnostic for SLE.
8. Management of therapy should be based not only on positive serologic tests for SLE, but should include the presence of active clinical disease.
9. Elderly patients with SLE have a better prognosis and their clinical symptoms differ substantially from those seen in younger patients.
10. Although the predominant class of antinuclear antibodies (ANA) is immunoglobulin G, the presence of immunoglobulin E may be of pathogenic importance in SLE.

<table>
<thead>
<tr>
<th>TABLE 1: INCIDENCE OF ANA IN VARIOUS DISORDERS</th>
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<tbody>
<tr>
<td><strong>Disease</strong></td>
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<tr>
<td>Systemic lupus erythematosus</td>
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<tr>
<td>Lupoid hepatitis</td>
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<tr>
<td>Progressive systemic sclerosis (scleroderma)</td>
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<tr>
<td>Rheumatoid arthritis</td>
</tr>
<tr>
<td>Juvenile arthritis</td>
</tr>
<tr>
<td>Feltty’s syndrome</td>
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<tr>
<td>Sjögren’s syndrome</td>
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<tr>
<td>Chronic discoid lupus</td>
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</tbody>
</table>


INSTRUCTIONS FOR USE

**Antinuclear (ANA) Antibody Test System**

K4805 - 48 Tests  K9605 - 96 Tests
K5005 - 50 Tests  K0005 - 100 Tests

Also for: Rat Liver slides S4010 - 4 well  S8101 - 8 well
          ANA Homogeneous Positive control S0101 - 5 well  S0110 - 10 well
          ANA Speckled Positive control C002N
          ANA Nucleolar Positive control C003N
          Reticulin Positive control C014A / C014G
          Centromere Positive control C018B
          ANA MSA Positive control C019

INTENDED USE
The Bio-Diagnostics Antinuclear Antibody Test System is an immunofluorescent antibody (IFA) test to detect the presence of antinuclear antibodies in human serum.

SUMMARY AND EXPLANATION
Antinuclear Antibody (ANA) tests are commonly performed on sera from patients with various connective tissue diseases, particularly in systemic lupus erythematosus (SLE), for diagnostic evidence, prognostic significance, and management of therapy. The highest titres of ANA are found in active SLE and the presence of these antibodies is the second most common manifestations of SLE. Immunofluorescence is the test of choice for screening for the presence of ANA since it detects 95 - 100% of active SLE cases. The presence of ANA has been well documented in different disease states as well as in healthy relatives of SLE patients. The incidence of positive ANA varies with each disease (see Table 1). Rat or mouse liver is utilized for ANA detection in this test system.

PRINCIPLE OF THE TEST
ANA antibodies are not organ or species specific. The primary test reaction involves circulating antinuclear antibodies present in the patient’s serum, which attach to their homologous nuclear antigens. This occurs during the incubation period whilst the serum covers the antigen surface. A rinsing period is followed by a secondary reaction. The reagent used in the secondary reaction is a fluorescein labelled anti-human globulin conjugate. The antigen surface is then thoroughly rinsed free of unbound conjugate and viewed under an appropriate fluorescent microscope to visually identify various morphological patterns of nuclear fluorescence.

The clinical significance of the various nuclear immunofluorescence patterns is useful in evaluating patients for the presence of one of the connective tissue diseases. The homogeneous pattern is the most common pattern and is associated with SLE. True speckled nuclear fluorescence is seen in Scleroderma, Raynaud’s disease, Rheumatoid Arthritis, and Sjögren’s syndrome. Nucleolar fluorescence is seen mainly in Scleroderma and Sjögren’s syndrome. Various drugs have been reported to induce or activate SLE and patients on these drugs often demonstrate varying levels of ANA in their serum.
Anti-Nuclear Antibody Test System

WARNINGS AND PRECAUTIONS

1. All human components have been tested by radioimmunoassay for (HBsAg) and HTLVIII/LAV by an FDA approved method and found to be negative (not repeatedly reactive). However, this does not assure the absence of (HBsAg) or HTLVIII/LAV. All human components should be handled with appropriate care.
2. The controls included in the kit contain 0.1% sodium azide as a preservative. Although this is at a low concentration, these reagents should be considered toxic. They should not be ingested or allowed to come into contact with either the skin or the mucous membranes. Sodium azide may also cause the formation of potentially explosive lead or copper azides in sinks.
3. Do not use components beyond their expiration date.
4. Follow the procedural instructions exactly as they appear in this insert to ensure valid results.
5. For in vitro diagnostic use only.
6. Handle slides by the edges since direct pressure on the antigen wells may damage the antigen.
7. Once the procedure has started do not allow the antigen in the wells to dry out. This may result in false negative test results, or unnecessary artefacts.

KIT CONTENTS

SLIDE

Rat liver substrate antigen slides (S4101, S8101, S5101 or S0101)

CONJ IgG

FITC Conjugate with Evans Blue Counterstain: J501/J501-5. This reagent contains antibodies that will react with the human IgG (H+L) Immunoglobulin class.

CONTROL + ANA homogenous Positive control no: C001N / C001N-0.5

CONTROL + ANA speckled Positive control no: C002N / C002N-0.5

CONTROL + ANA nucleolar Positive control no: C003N / C003N-0.5

CONTROL - Universal Negative control no: C000

IF/DF/A PBS Buffer Pack no: R002

MM Mounting Medium no: R005

Note: All kit components are available separately. Please see the Bio-Diagnostic Ltd catalogue for more details.

Additional Materials Required But Not Provided

Test tubes and rack microtitre system
Disposable pipettes
Staining Dishes and Slide Forceps
Moisture Chamber
Volumetric Flask (500 ml)
Distilled Water
Fluorescence Microscope
Paper Towels - lint free

REAGENT PREPARATION


KEY FOR OTHER SYMBOLS

Used in this instruction leaflet and on accompanying product packaging:

- Contains sufficient for <5 tests
- Ready for Use
- In vitro diagnostic medical device

STORAGE AND STABILITY

The IFA Test System components (except PBS) must be stored at a temperature of +2°C to +8°C. Do not freeze the test kit. The stability of the kit is as indicated by the expiry date on the packaging under the above storage conditions. This applies to unopened and opened reagents.

Phosphate Buffered Saline is stable at room temperature storage. The reconstituted Buffer does not contain preservatives and should be stored at 2-8°C. Care should be taken to avoid contamination.

SPECIMEN COLLECTION

Serological specimens should be collected under aseptic conditions. Haemolysis is avoided through prompt separation of the serum from the clot. Serum should be stored at 2-8°C if it is to be analysed within a few days. Serum may be held for 3 to 6 months by storage at -20°C or lower. Lipaemic and strongly haemolytic serum should be avoided.