

Anti-dsDNA Kit

REF IM77

50 tests

REF IM771

100 tests

For other languages
Pour d'autres langues
Für andere Sprachen
Para otras lenguas
Per le altre lingue



www.trinitybiotech.com

INTENDED USE

For the *in vitro* quantitative measurement of antibodies to double-stranded DNA (dsDNA) in human serum. Measurement range 0 to 100 IU/ml.

SUMMARY AND EXPLANATION OF THE TEST

The presence of circulating antibodies to dsDNA is highly specific to the autoimmune disease systemic lupus erythematosus (SLE), these antibodies probably being involved in the pathogenesis of the disease⁽¹⁾. Levels of anti-dsDNA antibodies are elevated in the majority of cases of clinically active SLE, falling during remission^(2,3). Measurement of anti-dsDNA antibodies may aid in the diagnosis of SLE, and in monitoring the course of drug therapy⁽³⁾. Persistently high or rapidly increasing levels are often associated with exacerbation of disease activity⁽⁴⁾. Raised levels of anti-dsDNA antibodies have also been detected in cases of discoid lupus erythematosus⁽⁵⁾. Drug induced lupus-like syndromes do not appear to involve anti-dsDNA antibodies, with the possible exceptions of hydralazine⁽⁶⁾ and some anticonvulsants⁽⁷⁾ at a low incidence.

PRINCIPLES OF THE PROCEDURE

The Anti-dsDNA Kit utilizes a technique in which the sample is incubated with an ¹²⁵I-labelled dsDNA tracer. Separation of immunoglobulin-bound tracer is effected by ammonium sulphate precipitation, followed by centrifugation and decanting of the supernatant. The amount of tracer bound is directly proportional to the level of anti-dsDNA antibody present.

WARNINGS AND PRECAUTIONS

For *In Vitro* Diagnostic Use Only.

For Professional Use Only.

Control values must fall within their "Acceptable Range", otherwise the test run is invalid. In the case of damage to the kit packaging, do not use damaged components.

Caution - Radioactive Material

This radioactive material may be received, acquired, possessed and used only by authorized persons in clinical laboratories or hospitals and only for *in vitro* clinical or laboratory tests not involving internal or external administration of the material or the radiation there from to humans or animals. Its receipt, acquisition, possession, use, transfer and disposal are subject to the regulations and a general licence of Atomic Energy Agencies or of the state/national body responsible for the exercise of such regulatory authority. These kits contain the radioisotope Iodine-125 (¹²⁵I) which has a half life of 60 days emitting ionizing X (28keV) and gamma (35.5keV) radiations.

WARNING – POTENTIALLY INFECTIOUS MATERIAL

Human blood products provided as components of this pack have been obtained from donors who were tested individually and who were found to be negative for human immunodeficiency virus (HIV-1) antibody and hepatitis B surface antigen using approved methods (enzyme immunoassays). These components have also been tested using approved methods (enzyme immunoassay) and found to be negative for Hepatitis C Virus (HCV) antibody and HIV-2 antibody.

Care should be taken when handling material of human origin. All samples should be considered potentially infectious. No test method can offer complete assurance that hepatitis B virus, HCV, HIV 1+2 and other infectious agents are absent. Handling of samples and assay components, their use, storage and disposal should be in accordance with the procedures defined by the appropriate national biohazard safety guideline or regulation.

MATERIALS PROVIDED

IM77:

- 1 bottle tracer (¹²⁵I-labelled dsDNA, <74 kBq/bottle) in buffer (10.5 ml).
- 1 bottle ammonium sulphate solution (55 ml).
- 1 set anti-dsDNA standards (freeze-dried human serum, nominal values A-F: 0, 5, 10, 25, 50, 100 IU/ml, exact values stated on standard value label). Reconstitution volumes: A 2 ml, B-F 0.2 ml.
- Instructions for use.

IM771:

- 2 bottles tracer (¹²⁵I-labelled dsDNA, <74 kBq/bottle) in buffer (10.5 ml).
- 2 bottles ammonium sulphate solution (55 ml).
- 1 set anti-dsDNA standards (freeze-dried human serum, nominal values A-F: 0, 5, 10, 25, 50, 100 IU/ml, exact values stated on standard value label). Reconstitution volumes: A 2 ml, B-F 0.2 ml.
- Instructions for use.

MATERIALS REQUIRED BUT NOT PROVIDED

Precision pipettes, repeating dispenser (optional), control sera, 5 ml assay tubes and racks, plastic or metal film (to cover tubes), vortex mixer, water bath, decanting racks, gamma scintillation counter.

Reagent Preparation and Storage

Do not use reagents beyond the expiration date stated on the package label. Store unopened reagents at 2-8°C. Do not freeze.

Standards: reconstitute standard A with 2 ml and standards B-F with 0.2 ml distilled water. Store at -20°C (with no more than 2 freeze-thaw cycles).

All other reagents are supplied ready for use, store at 2-8°C after opening.

REAGENTS REQUIRED BUT NOT SUPPLIED

Catalogue No.	Item Description	Quantity
QAS-114	Amerlex Radioassay Controls 31 and 32	
	Control 31 (Low) (freeze-dried human serum)	6 x 0.2 ml
	Control 32 (High) (freeze-dried human serum)	6 x 0.2 ml

SAMPLE COLLECTION, PREPARATION AND STORAGE

Serum samples only may be used. Store samples at 2-8°C for up to 5 days or at -20°C for longer periods. Avoid repeated freezing and thawing.

QUALITY CONTROL AND PROCEDURAL NOTES

1. Use only reagents from the same pack for each assay. The tracer, standards and samples should be brought to 18-28°C. The ammonium sulphate solution should be kept at 2-8°C. All samples and reagents should be thoroughly mixed (avoiding excessive foaming) before use. The total dispensing time of the tracer and ammonium sulphate solution should not exceed 12 minutes. All reagents should be dispensed without interruption.
2. Run a separate standard curve for each assay. Standards, controls and samples should be assayed in duplicate. Good laboratory practice requires that controls be run to verify the performance of the assay.
3. Samples likely to contain >100 IU/ml anti-dsDNA should be diluted in zero standard prior to assay. Determination of total counts is optional. Centrifugation should be carried out within 20 minutes of ammonium sulphate precipitation. Do not reinvert drained tubes once they have been turned upright.

TEST PROTOCOL

PROCEDURE

1. Assemble and label assay tubes.
2. Pipette 25 µl standard, control or sample into appropriate tubes.
3. Dispense 200 µl tracer into all tubes. Set aside total counts tubes (optional).
4. Vortex, cover and incubate at 37°C for 60±10 minutes.
5. Dispense 1 ml cold ammonium sulphate solution into all tubes.
6. Vortex, centrifuge at 2-28°C and 1500 g for 15 minutes.
7. Decant and drain for 10-15 minutes with blotting.
8. Count tubes so as to accumulate at least 1000 counts in the zero standard tubes.

RESULTS

Plot standard curve, either manually or using an IRMA curve fit programme. Results may be calculated using linear plotting.

Linear-linear Plotting

Plot standard counts against concentration on linear graph paper. Draw the best curve through the mean of duplicate points, rejecting grossly aberrant counts. Read the concentration of the unknowns from the standard curve.

Table 1: Sample Calculation
(expressed in IU/ml)

Tube Number	Sample	Standard Conc.	Counts/min X1	Counts/min X2	Mean anti-dsDNA Conc
1-2	Totals		48343	48159	
3-4	Std A	0	865	876	
5-6	Std B	5	2057	1903	
7-8	Std C	10	3283	3355	
9-10	Std D	25	8680	8529	
11-12	Std E	50	17362	17094	
13-14	Std F	100	32090	31884	
15-16	U1†		1718	1646	4.0
17-18	U2		3693	3642	10.9
19-20	U3		25813	25644	76.7

† Unknown.

LIMITATIONS OF THE PROCEDURE

1. The results obtained from this or any other diagnostic kit should be used and interpreted only in the context of an overall clinical picture.
2. Lipemic, haemolyzed and icteric samples may be used up to levels of 67.8 mmol/l triolein, 2.5 g/l haemoglobin and 0.85 mmol/l bilirubin. Do not use turbid samples.

EXPECTED VALUES

It is recommended that each laboratory establish its own reference interval. As a guide, the upper limit of the normal range (mean ± 2 standard deviations of log transformed values) derived from a study of 272 normal subjects was 7 IU/ml. Of 612 patients with active SLE, 96% had anti-dsDNA levels of >7 IU/ml. Of 436 patients with inactive SLE and 331 patients with other autoimmune disorders, 82% and 94% respectively had anti-dsDNA levels <7 IU/ml.

PERFORMANCE CHARACTERISTICS

- Calibration**
The kit has been developed in IU/ml against the 1st International Reference Preparation (Wo/80). A correlation between the Anti-dsDNA Kit and the Anti-DNA Kit, has been obtained by measuring a panel of 1067 patient samples from a variety of clinical categories: Anti-dsDNA Kit = 1.11 x Anti-DNA Kit - 11.6 (IU/ml), with a correlation coefficient of 0.754.
- Recovery**
Recovery is the increase in value seen when a known concentration of analyte is added to a sample, i.e. the measured increase expressed as a percentage of the expected increase. The mean recovery for 2 normal samples spiked with human anti-dsDNA at 3 levels was 102% , with a range of 96% to 109%.
- Reproducibility**
Four freeze-dried control sera were assayed in 10 replicates to determine within-assay reproducibility. To determine between-assay reproducibility the controls were measured in duplicate in 10 assays. The data are typical of the reproducibility found between operators and reagent batches over the shelf life of the kit.

Table 2: Reproducibility (expressed in IU/ml)

Within-assay			Between-assay	
Mean	CV(%)	Mean	CV(%)	CV(%)
6.4	8.1	7.2	11.5	
19.1	4.0	18.9	5.0	
51.0	3.2	50.7	2.5	
79.9	2.9	77.1	3.0	

- Sensitivity**
Sensitivity is defined as the concentration 2 standard deviations from the zero standard. The sensitivity of the Anti-dsDNA Kit is typically better than 2.5 IU/ml.
- Specificity**
Levels of complement C1q up to 300 mg/l do not interfere with the Anti-dsDNA Kit.

REFERENCES

- Christian CL. *New Eng J Med* **280**: 878-884 (1969).
- Davis P et al. *BMJ* **3**: 88-89 (1973).
- Hughes GRV et al. *Ann Rheum Dis* **30**: 259-264 (1971).
- Swaak AJG et al. *Ann Rheum Dis* **41**: 388-395 (1982).
- Mandel MJ et al. *Arch Dermatol* **106**: 668-670 (1972).
- Hahn BH et al. *Ann Intern Med* **76**: 65-74 (1972).
- Alarcon-Segovia D & Fishbein E. *J Rheumatol* **2**: 167 (1975).

















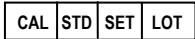
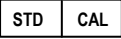




ORDERING INFORMATION

Catalogue No.	Maximum assays
IM77	50
IM771	100

Optional Reagents

Catalogue No.	Item Description	Quantity
QAS-114	Amerlex Radioassay Controls 31 and 32	
	Control 31 (Low) (freeze-dried human serum)	6 x 0.2 ml
	Control 32 (High) (freeze-dried human serum)	6 x 0.2 ml

GUIDE TO SYMBOLS

 Catalogue Number	 Use-by date
 Batch code	 Temperature limit
 <i>In vitro</i> diagnostic medical device	 Consult instructions for use
 Contents	 www.trinitybiotech.com eIFU indicator
 Standard	 Manufacturer
 Calibrator A	 Calibrator B
 Calibrator C	 Calibrator D
 Calibrator E	 Calibrator F
 Calibration Standard Set Lot	 Standard Calibrator
 Tracer (¹²⁵ I-labelled dsDNA, <74 kBq/bottle)	 Ammonium Sulphate Solution
 Biological risks	 Radiation Hazard



Trinity Biotech plc,
IDA Business Park
Southern Cross Road
Bray, Co. Wicklow
Ireland
Tel. + 353 1 2769800
Fax. +353 1 2769888
www.trinitybiotech.com

CANADA ENQUIRIES:
Trinity Biotech USA
2823 Girts Road
Jamestown, NY 14701
Tel: 1800 325 3424
Fax: 716-488-1990

