



QuantiFERON®-TB Gold In-Tube assay A NEW TEST ANNOUNCEMENT

Dr Renu Vohra

Introduction

QuantiFERON®-TB Gold In-Tube assay (QFT-Gold) is an in-vitro diagnostic test for the detection of *Mycobacterium tuberculosis* infection. The test uses modern genetic recombination technology to provide fast, accurate and convenient method of diagnosing TB infection. The assay has a sensitivity of 90% and specificity of >98% for diagnosing TB.

The QuantiFERON®-TB Gold can be used in place of (and not in addition to) the Mantoux test.

Principle of the test

The QuantiFERON®-CMI is an *in-vitro* laboratory test for the measurement of cell mediated immune (CMI) responses in humans using whole blood. The test involves undiluted whole blood being stimulated with test antigen(s) & negative control antigen(s) and a mitogen. T-cell responses are then determined by the quantitative measurement of Interferon-gamma (IFN-γ) in plasma by a rapid, single-step enzyme linked immunosorbent assay (ELISA).

The TB specific antigens used in QuantiFERON®-TB Gold In-tube method are ESAT-6, CFP-10 and TB 7.7(p4). These antigens are only made by *Mycobacterium tuberculosis* complex bacteria, and therefore identify the presence of only those T-cells that are totally specific for TB infection. These antigens are absent from the tuberculosis vaccine organism BCG and from most environmental mycobacteria. Thus these proteins are precise markers of true *Mycobacterium tuberculosis* infection. As a result, unlike the tuberculin skin test (TST or Mantoux test), the QuantiFERON®-TB Gold test is completely unaffected by the BCG vaccination status of the individual being tested.

Advantages

- Unaffected by BCG vaccination
- Unaffected by nearly all non-tuberculous mycobacteria
- Requires only a single patient visit
- Simple YES / NO answer
- No possibility of adverse reactions in hypersensitive people
- Results obtained within 24 hours (48-72 hrs for the skin test)
- Reliability in immunocompromised patients for immune system competence

continued inside...

Urgent Reminder

In a recent Medicare audit of glycosylated haemoglobin (HbA1c) ordering across Australian laboratories, it came to light that for over a third of requests for this test, the laboratory could not establish that the patient was legally entitled to receive a Medicare benefit.

The Medicare Schedule allows for HbA1c to generate a rebate only:

- "in the management of established diabetes" (Item 66551) and
- "in the management of pre-existing diabetes where the patient is pregnant" (Item 66554).

There is no allowance for HbA1c to be used in the diagnostic workup of suspected diabetes.

To address this when requesting this test on a diabetic, please indicate the patient's diabetic status (e.g. by writing "DM", "IDDM", "Type 2 DM", etc) in the clinical note section.

Thank you for your attention to this.

With kind regards,

Charles Appleton

Director, Biochemistry

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Intended use

1. Assist in the diagnosis of TB infection

- a. **QuantIFERON®-TB Gold test for latent infection**
Although the QuantIFERON®-TB Gold test is highly effective in detecting active TB disease, its most common use is diagnosing latent TB infection in screening programs and public health situations. QuantIFERON®-TB Gold testing is far more accurate in detecting active TB than the skin test, with a sensitivity approaching 90%. T-cell IFN- γ responses are lower in active TB patients than in healthy, but latently, infected individuals and thus the sensitivity of QuantIFERON®-TB Gold for latent TB infection is likely to be even higher.

- b. **Better detection of active TB disease**

QuantIFERON®-TB Gold test has been shown to be much more sensitive in detecting untreated active TB. In fact the skin test misses up to three times more active cases in screening.

Please note that the QuantIFERON®-TB Gold test can only be interpreted in conjunction with other epidemiologic, historic, physical and diagnostic findings including a chest X-ray.

2. Contact investigations

In persons with recent contact with persons who have infectious TB, negative QuantIFERON®-TB Gold results should be confirmed with a repeat test performed 8 – 10 weeks after the end of exposure, as is recommended for the Mantoux test.

3. Evaluation of recent immigrants who have had BCG vaccination

4. TB screening of health care workers

Limitations

1. Currently there is no data relating to the use of the QuantIFERON®-TB Gold test in individuals under 17 years of age.
2. Performance of QuantIFERON®-TB Gold in particular its sensitivity and its rate of indeterminate results has not been evaluated in the following groups:

- a. Patients who are immunocompromised such as those with HIV infection, AIDS, and transplant recipients
- b. Patients undergoing immunosuppressive therapy
- c. Persons with other clinical conditions that may compromise the immune system: diabetes, silicosis, chronic renal failure, hematological disorders (e.g., leukemia and lymphomas), and other specific malignancies (e.g., carcinoma of head and lung)

Medicare rebate

A rebate is only available for immunocompromised or immunosuppressed patients. Medicare rebate would cover groups like: HIV positive individuals, patients on immunosuppressive therapy, elderly patients and cancer patients.

Specimen collection

1ml of blood is collected in each of the three tubes. The tubes are inverted for 8-10 times or shaken for 5 seconds ensuring that the entire surface of the tubes are coated with the blood.

Transport

Tubes should be stored and transported at room temperature ($22^{\circ}\text{C} \pm 5$). Do **not** refrigerate or freeze the samples. The tubes can be kept at room temperature for maximum of 16 hours; however it is recommended that the tubes be transferred for incubation at 37°C as soon as possible.

Because of the need to have viable lymphocytes, patients should be referred for this test early in the week to ensure optimum cell recovery.

This test is only performed once a week. For further information or clarification please contact one of our Microbiologists on (07) 3840 4444.

References:

Mazureck G H., Jereb J., et al: *Guidelines for Using the Quantiferon – TB Gold Test for Detecting Mycobacterium tuberculosis Infection*, United States. Morbidity and Mortality Report: Recommendations and Reports. Vol 54/RR-15, December 16th 2005 p49 – 54

New Central Laboratory

The New Year is upon us and it has brought with it some significant steps forward in the progress for our move to the new laboratory.

Currently we are finalising the plans for a staged relocation to ensure we continue to meet the needs of our referrers during this period. All laboratory operations will move over the Easter long weekend with duplicate machines being moved earlier to maintain our service.



These operations will be supported by our Metropolitan Stat Laboratories and nearby Branch Laboratories; ensuring all your specimens continue to be processed in a timely manner.

The change of location will bring with it both a new central laboratory address and phone numbers. Diversions will occur for a lengthy period of time, however your Medical Liaison Officers (MLOs) will be distributing materials closer to the move with all the relevant details. Please be assured that this will not cause major disruption as the old numbers you are familiar with will continue to get you in touch with our staff at QML Pathology.

As the move approaches we will continue to communicate with you via the QML Newsletter and our MLOs in order to make sure you are up-to-date and well informed.

Antibodies to Extractable Nuclear Antigens (ENA)

Dr David Heyworth-Smith

Antibodies to Extractable Nuclear Antigens (ENA) are important markers of autoimmune disorders. The antinuclear antibody (ANA) test is a good screening test for most anti-ENA antibodies. If the ANA is negative, an anti-ENA test is likely to be negative, with some exceptions.

Antibodies to ENA are antibodies to specific well characterised cellular antigens. These antibodies have known disease associations (table 1). Commonly detected antibodies are:

- Anti-SSA (Ro)
- Anti-SSB (La)
- Anti-RNP
- Anti-PM-Scl (PM-1)
- Anti-Scl70 (DNA topoisomerase)
- Anti-Sm (Smith)
- Anti-Jo

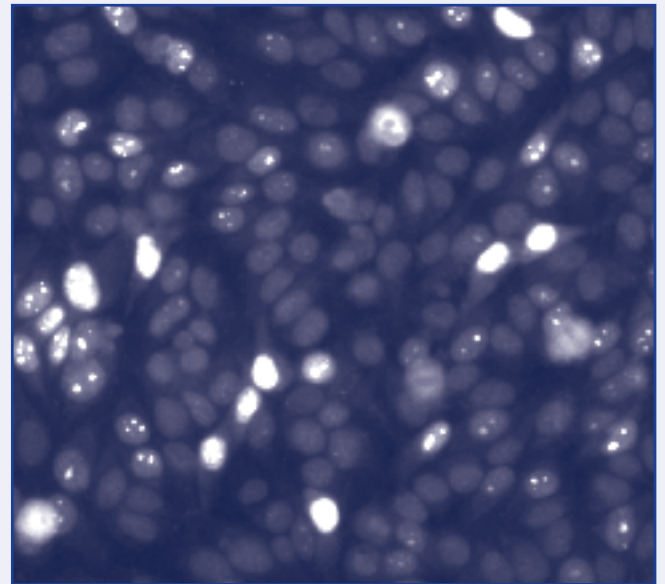


Figure 1: Speckled ANA with anti-SSA pattern

Table 1 Clinical associations of the most common anti-extractable nuclear antigen (anti-ENA) antibodies

Anti- ENA	Major clinical associations	Comments
SS-A/Ro	Sjogren's syndrome SLE Neonatal lupus syndromes C2/C4 deficiency	Anti-SSA/Ro antibodies are the most commonly detected anti-ENA antibody. There is some controversy as to whether anti-SSA antibodies may be detected in otherwise normal people.
SS-B/La	Sjogren's syndrome SLE Neonatal lupus syndromes	Anti-SSB/La antibodies are generally only found along with anti-SSA/Ro antibodies. Thus an isolated anti-SSB/La result should be interpreted with caution.
Sm	SLE	Anti-Sm antibodies are considered to be highly specific for SLE, but only occur in about 1/3 of cases. They are more likely to be detected in subjects of Afro-Caribbean or Asian ethnic backgrounds, in whom they are associated with an increased risk of developing more severe disease (including renal lupus)
U1-RNP	MCTD SLE	While anti-U1-RNP antibodies are classically associated with MCTD, they can also be found in individuals with SLE (often in association with anti-Sm antibodies).
Scl-70	Scleroderma	In patients with scleroderma, anti-Scl-70 antibodies are associated with an increased risk of developing internal organ involvement (including lung and renal disease) and extensive cutaneous disease. However, anti-Scl antibodies are occasionally detected in the absence of scleroderma.
Jo-1	Polymyositis/ Dermatomyositis	Anti-Jo-1 antibodies are associated with an increased risk of developing interstitial lung disease. As Jo-1 is only found in the cytoplasm of cells, the ANA may be reported as negative

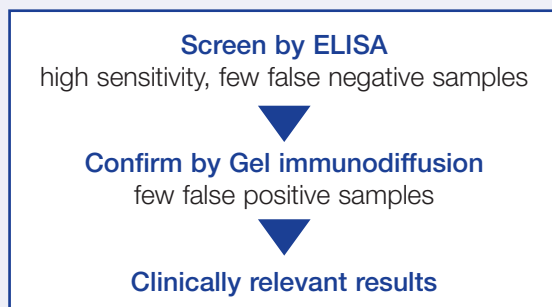
Most anti-ENA are specific for nuclear antigens, with a notable exception of anti-Jo. Anti-Jo antibodies are specific for a cytoplasmic antigen, namely histidyl t-RNA synthetase. Anti-Jo antibodies are indicative of a form of polymyositis or dermatomyositis with interstitial lung disease named “synthetase syndrome”

Thus testing for anti-ENA can provide further information in patients with suspected autoimmunity, beyond that provided by the ANA. Similarly, anti-double stranded DNA antibodies which are associated with systemic lupus erythematosus (SLE) may provide confirmation of the significance of a positive ANA.

There are several testing methodologies for the detection of anti-ENA and anti-ds-DNA antibodies. Different methods produce results of differing clinical significance. The gold standard for the detection of anti-ENA is gel immunodiffusion and the gold standard for the detection of anti-ds DNA is radioimmunoassay (RIA). The descriptive clinical syndromes utilised for the categorisation and diagnosis of autoimmune disorders were defined on the basis of measurement of anti-ENA by gel immunodiffusion, and anti-ds DNA by RIA. Newer methodologies for the detection of autoantibodies include enzyme linked immunosorbent assay (ELISA), immunoblotting, and multiparameter bead arrays (Luminex). In general ELISA suffers from enhanced sensitivity for most antibodies, but diminished specificity compared to the gold standard assays. The role of immunoblotting and Luminex for the detection of anti-ENA remains unclear at this time.

QML Pathology utilises a form of gel immunodiffusion for detection of anti-ENA and RIA for detection of anti-ds DNA antibodies. However QML Pathology is changing strategies for the detection of anti-ENA, and will utilise a sensitive ELISA for the initial screening of samples for anti-ENA, followed by specific confirmation of positive screen samples by gel immunodiffusion (Figure 2).

Figure 2



With all changes in methodology, there are potential pitfalls to avoid. ELISA does not detect antibodies to uncharacterised or unknown nuclear antigens. Gel immunodiffusion systems do however, and previously QML Pathology has reported these as “unknown precipitin lines” (UPLs). The clinical significance of an UPL is uncertain, because it represents an uncharacterised antibody. The new screening ELISA method will not detect an isolated UPL, it will only be detected if accompanied by a characterised positive ENA in the sera. The clinical significance of UPL only

positive anti-ENA is probably low for most patients. However if you think detection of a UPL is relevant to your patient, please request “ENA by CIEP” (CIEP or counter immunoelectrophoresis represents a type of gel immunodiffusion) or “ENA by gel diffusion”.

CASE EXAMPLE

A 56 year old female presents with complaints of dry eyes and a dry mouth, thinning of her hair, painful finger joints, and fatigue. Examination demonstrates mild puffiness and tenderness of the proximal interphalangeal joints only.

The FBE is normal, ELFTs shows increased globulins, and the ESR is 65. Suspecting an autoimmune condition, further investigations are performed.

- ANA positive, titre 1:640, speckled pattern
- ENA positive, anti-SSA and anti-SSB
- Anti-dsDNA negative
- Rheumatoid factor 1:64 (Rose Waaler)
- Urinalysis – trace protein
- Serum protein electrophoresis - polyclonal hypergammaglobulinaemia.

Q What is the most likely diagnosis?

A The history of dry eyes and mouth is suggestive of xerophthalmia and xerostomia (collectively “sicca symptoms” which may include vaginal dryness). This could be confirmed by a Schirmer’s tear production test. Mild alopecia is a frequent complaint in the connective tissue disorders. There is probably mild proximal interphalangeal joint arthritis with synovial inflammation. Fatigue is non-specific but may be an accompaniment of most connective tissue disorders.

The increased ESR is a consequence of the hypergammaglobulinaemia and possibly elevation of other “acute phase” serum proteins such as fibrinogen. The positive speckled ANA alone is a non-specific finding. However further information has been provided about the specificity of the ANA by anti-ENA testing. In this case demonstrating the presence of anti-SSA/SSB (Ro/La) which suggests a diagnosis of Sjogren’s syndrome. Polyclonal hypergammaglobulinaemia and elevation of rheumatoid factor are frequent in active primary Sjogren’s syndrome. Secondary Sjogren’s syndrome occurs in the context of another connective tissue disorder such as rheumatoid arthritis or SLE. The picture is most suggestive of primary Sjogren’s syndrome but evolution of another disorder could not be excluded.

For further information or clarification please contact one of our Immunologists on (07) 3840 4444.

Announcement of Stage 3 of the Rh (D) Antenatal Prophylaxis Program

Antenatal and postnatal prophylaxis for all Rh (D) negative women using solely Australian Rh (D) Immunoglobulin (Anti-D)

Australia is now self-sufficient in its supply of Rh (D) Immunoglobulin (Anti-D). Professor Richard Smallwood, Chair of the National Blood Authority Advisory Board, has announced that routine Rh (D) antenatal and postnatal prophylaxis can now be given to all Rh (D) negative women without pre-formed anti-D using solely Australian Rh (D) immunoglobulin.

By March 31 2006, the Australian Red Cross Blood Service (ARCBS) will cease to routinely issue *WinRho SDFTM*. From that date, imported product will only be issued when access to an intravenous preparation is warranted. This change represents Stage 3, the final implementation phase of the routine Rh (D) antenatal prophylaxis program.

For Stage 3, Rh (D) Immunoglobulin products should be used as indicated below:

- First trimester sensitising events¹ (<12 weeks): Rh (D) immunoglobulin 250 IU
- First trimester sensitising events¹ (multiple pregnancies <12 weeks): Rh (D) immunoglobulin 625 IU
- Second and third trimester sensitising events¹: Rh (D) immunoglobulin 625 IU
- All Rh (D) negative women without preformed Anti-D: Rh (D) immunoglobulin 625 IU at 28 and 34 weeks gestation
- Postnatal prophylaxis: Rh (D) immunoglobulin 625 IU

To facilitate smooth implementation of the revised policy, the Joint Rh (D) Consultative Committee (JCC) has reviewed the Guidelines for the use of Rh (D) Immunoglobulin with representatives from all key stakeholders, including:

- Royal Australian and New Zealand College of Obstetrics and Gynaecology (RANZCOG)
- Royal Australian College of General Practitioners (RACGP)
- Australian College of Midwives Inc (ACMI)
- Abortion Providers' Federation of Australia (APFA)
- Australian and New Zealand Society of Blood Transfusion (ANZSBT)
- Australian Institute of Medical Scientists (AIMS)
- Australian Red Cross Blood Service (ARCBS)
- Royal College of Pathologists of Australasia (RCPA)
- CSL Limited (CSL)
- National Blood Authority (NBA)

Information is currently being updated and will be placed on all relevant web sites. A range of revised support materials will also be available. Further information will be forwarded to you within the next month.

For further information please contact your local ARCBS centre, CSL Limited or your professional college or society.

Note 1: Sensitising events include ectopic pregnancy, miscarriage, termination of pregnancy and ultrasound guided procedures such as chorionic villus sampling, amniocentesis, cordocentesis and fetoscopy, as well as abdominal trauma considered sufficient to cause fetomaternal haemorrhage, external cephalic version, antepartum haemorrhage and normal delivery.

COLLECTION CENTRE NEWS

For the convenience of our doctors and patients, we have listed the latest changes to QML Pathology's network of clinics:

NEW CLINICS

Oakey (07) 4691 2481

Shop 1, 81 Campbell St
Mon – Fri 8.30am - 12.30pm
1.00pm - 4.30pm

Ashmore (07) 5564 9572

Ashmore Plaza Shopping Centre,
130 Cotlew St
Mon – Fri 7.30am - 12.30pm
1.00pm - 4.00pm

Labrador Park (07) 5537 4162

Shop 18, Labrador Park S/C,
100 Brisbane Rd
Mon – Fri 7.30am - 1.00pm
2.00pm - 4.00pm

Please contact your local branch for further information.

Holland Park (07) 3397 6976

Suite 1, Holland Park Prof. Suites,
1000 Logan Rd
Mon – Fri 7.30am - 12.30pm
1.00pm - 3.30pm

Redland Bay (07) 3206 8671

Shop 30, Bld 7, Redland Bay S/C,
133 Broadwater Tce
Mon – Fri 7.00am - 12noon
1.00pm - 3.30pm

Belrowes (07) 4724 3213

Shop 8, Belrowes Plaza,
49 Bundock St, Belgian Gardens
Mon – Fri 7.30am - 12noon
1.00pm - 4.00pm
Saturday 7.30am - 11.00am

CLINIC CHANGES

Scarborough (07) 3880 4119

Suite 1, 107B Landsborough Ave
Mon – Fri 8.00am - 12.30pm
1.30pm - 4.30pm

Oxenford (07) 5573 7039

Oxenford Medical & Professional
Centre, 5 Michigan Dr
Mon – Fri 7.30am - 4.30pm
Sat 8.00am - 11.00am

Hermit Park (07) 4721 0756

18 Bayswater Rd
Mon – Fri 8.00am - 1.00pm
2.00pm - 4.30pm
Sat 8.00am - 12noon

Kippa Ring (07) 3883 1558

Cnr Boardman Rd & Anzac Ave
Mon - Fri 7.30am - 6.00pm
Sat 8.00am - 12noon



Doctors' Notice Board

Dr Richard Freihaut, Orthopaedic Surgeon specialising in Foot and Ankle surgery, and Lower Limb Joint Replacement has commenced practice in Lismore.

- Suite 5, St Vincents Specialist Centre,
Dalley Street, Lismore NSW 2480
Ph: (02) 6621 6397
Fax: (02) 6621 6703

Dr Perumalla has been a full-time Staff Specialist in Urology at Nambour General and Caloundra Hospitals since October 1999. He is now moving into full-time private practice consulting at both Nambour Selangor Private Hospital & Caloundra Private Hospital.

His main interests are in urolithiasis, urological oncology, management of lower urinary tract symptoms and kidney transplantation.

- Nambour Selangor Private Hospital
62 Netherton St, Nambour
- Caloundra Private Hospital
Beerburum St, Caloundra

Telephone for appointment on (07) 5441 6477.

Dr Frank Carmody would like to inform you of his available medical suites for use in Sunnybank.

- Newly refurbished - for sessional or permanent lease
- Opposite Sunnybank Private Hospital
- Support services available nearby include xray/pathology/specialist centre
- Undercover parking available

For enquiries, phone Jenny Stuart on (07) 3371 4933.

Dr John O'Neill, ENT Surgeon, is pleased to advise that he is commencing private practice at -

- John Flynn Medical Centre, Suite 2B,
Sessional Suite, Inland Dve, Tugun

For appointments please telephone (07) 5598 9156.

His interests include paediatric and adult ENT conditions, especially rhinology and head and neck surgery.

Dr Paul Vasey, Medical Oncologist, would like to announce the commencement of his private practice at both the Wesley and Mater clinics of the Haematology Oncology Clinics of Australasia. Dr Vasey specialises in ovarian carcinoma and gynaecological and urological cancers. For appointments and enquiries please see the details outlined below:

- Wesley Medical Centre
1st Floor, 40 Chasley Street, Auchenflower
Ph: (07) 3737 4500
- Mater Medical Centre
Level 5, 293 Vulture Street, South Brisbane
Phone: (07) 3335 1900

Mobile: 0400 018 646

Email: pvasey@hoca.com.au

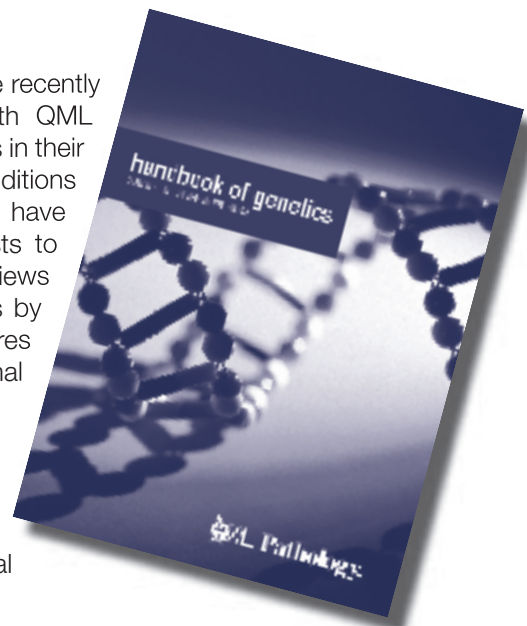
Anti-D Injection Reminder

To ensure you receive your orders as soon as possible, if you require anti-D injection to be delivered, please call us directly with patient and surgery details on 3840 4424 to arrange delivery on the next courier.

Introducing our Handbook of Genetics

Sue White and Dr Stephen Withers from our Genetics Department have recently completed and published a Genetics Handbook in association with QML Pathology. This handbook has been written to assist health professionals in their care of patients. There has been an increasing interest in genetic conditions from the perspective of both clinicians and patients. As a result we have identified the need for Paediatricians and Obstetricians/Gynaecologists to have at one's fingertips the highest quality visual aids and clear overviews of conditions when explaining concepts to patients. This handbook is by no means a definitive text. It does however provide many useful pictures and diagrams when explaining a range of conditions from chromosomal disorders through to basic Mendelian Inheritance.

Over the coming months the handbook will be delivered to Paediatricians and Obstetricians/Gynaecologists across Queensland and northern New South Wales. If you have not yet received a copy and would like to get this in the immediate future please contact your Local Medical Liaison Officer.



QML Pathology.



Symbion Pathology Pty Ltd ABN 84 007 190 043 t/a QML Pathology

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This newsletter has been prepared and published by QML Pathology for the information of referring doctors. Although every effort has been made to ensure that the newsletter is free from error or omission, readers are advised that the newsletter is not a substitute for detailed professional advice.

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