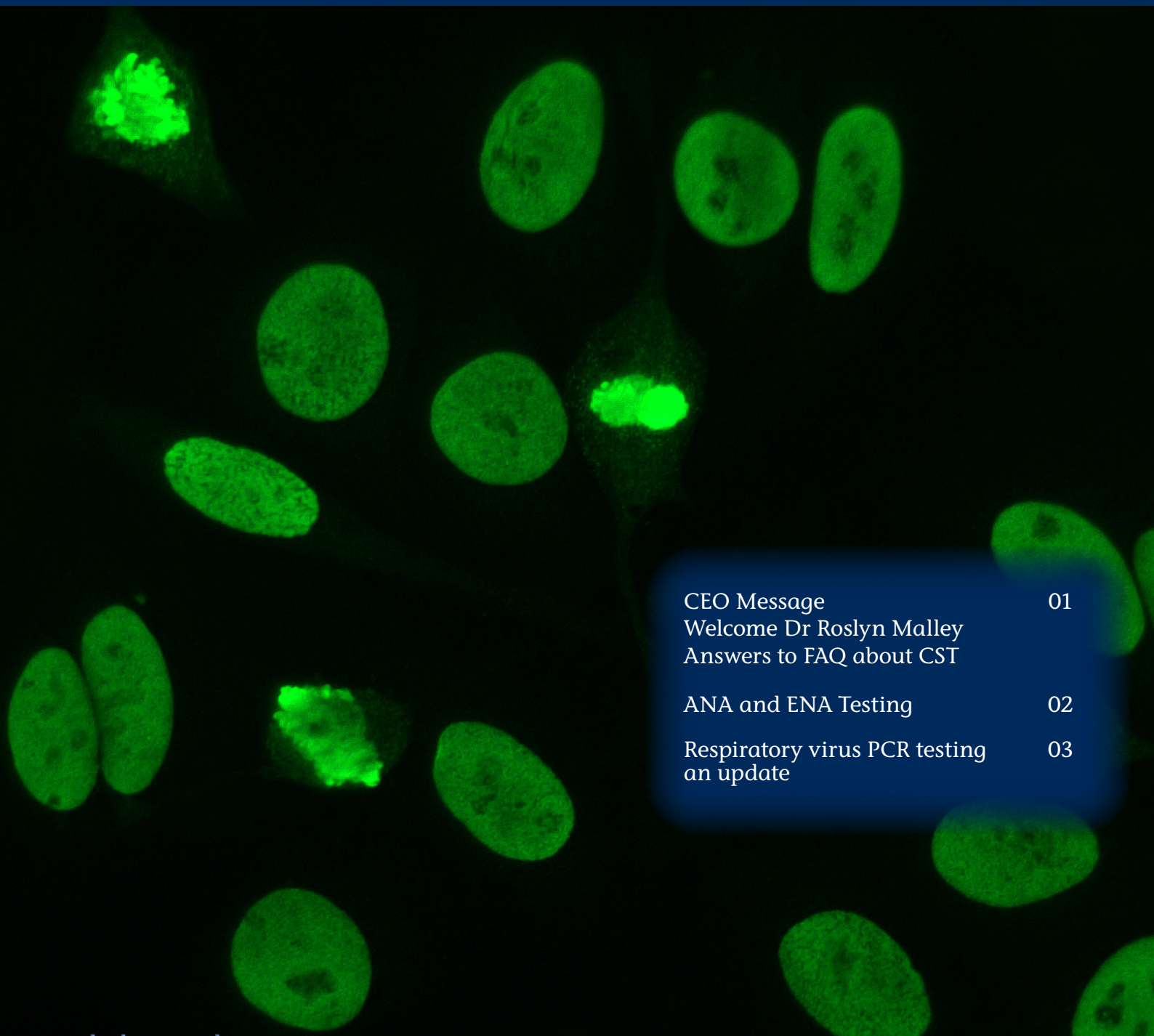
HOBART PATHOLOGY
LAUNCESTON PATHOLOGY
NORTH WEST PATHOLOGY
Quality is in our DNA

Doctors' Newsletter

ISSUE 1 | 2019



www.hobartpath.com.au
www.launcestonpath.com.au
www.northwestpath.com.au

CEO Message

Dr Shaun Donovan
BMedSci, MBBS (Hons), FRCPA, GAICD
CEO Diagnostic Services Pty Ltd (DSPL)
Hobart, Launceston and North West Pathology

I have great pleasure in introducing the first Doctor's Newsletter for 2019.

Firstly, I would like to welcome our new general pathologist, Dr Roslyn Malley. Ros will greatly enhance the supervision of our laboratories across the state and will be a valuable addition to our team of local pathologists.

In this newsletter, there is a timely article detailing answers to frequently asked questions about the new cervical screening program, Dr Alistair McGregor has provided a respiratory virus update and Dr Ros Malley an article on ANAs and ENAs.

Finally, I would like to take this opportunity to thank all our referrer's for your continued support.

Welcome

Dr Roslyn Malley
BMedSci(Hons), MBBS(Hons), PhD, FRCPA
General Pathologist

Hobart, Launceston and North West Pathology would like to welcome Dr Roslyn Malley to our team of pathologists.

Ros grew up in the North West Coast of Tasmania, before completing her Medical Degree (MBBS) in Hobart at the University of Tasmania.

She has travelled extensively, completed a PhD in Immunology, become a mother to two children, Samuel and Euan, and completed her Fellowship in General Pathology.

Ros continues to lecture medical students at the University of Tasmania and participate in research activities.

Answers to some frequently asked questions about Cervical Screening Test (CST)

1. The follow up test for a HPV positive and/or LSIL LBC result is a repeat HPV test in 12 months. A repeat co-test (HPV and LBC) is not clinically indicated and therefore is not rebated by Medicare.
2. The test of cure is only for previous histologically confirmed HSIL lesions. LSIL confirmed lesions and HSIL/possible HSIL LBC results are not eligible for a test of cure. Patients with histologically confirmed AIS or cancer results are eligible for a co-test indefinitely.
3. Previously confirmed HSIL on histology requires a test of cure. This means the patient is required to have 2 negative co-tests 12 months apart. Prior to 2008 these patients were required to have yearly pap smears indefinitely until the test of cure was introduced.
4. Discharge, dyspareunia, cervical polyp, ectropian, contact bleed, spotting on OCP are examples of patient symptoms that are NOT eligible for a co-test or an early repeat HPV.
5. There is NO Medicare rebate for routine repeat CST within the 57 month screening interval. Dates of previous CST can be found by calling the National Cancer Screening Register, 1800 627 701. The laboratory may also be able to assist with this if the previous smear was at a Diagnostic Services laboratory (Hobart, Launceston or North-West Pathology). If the smear was done by another laboratory the results may not be in our system.
6. There is NO Medicare rebate for routine CST on women under 24 years and 9 months. These women can have a co-test if symptomatic or are eligible for ONE CST between the ages of 20 and 24 if they were sexually active before the age of 14 (ie prior to eligibility for the HPV vaccine).
7. Women are eligible to join the new CST program at any time regardless of the timing of their pap smear in the previous screening program.
8. Women can request a CST at any time they wish within the 57 month interval, if they would like to privately fund the test or can add an LBC to their routine HPV.

The costs of these are as follows:
HPV only $65* (please note: if HPV is detected the reflex LBC is required)
LBC only $65*
HPV and LBC $100*

*Prices correct at time of printing

Anti-Nuclear Antibody (ANA) and Extractable Nuclear Antigen (ENA) testing

Autoantibodies that can bind to self-proteins, are produced by plasma cells and result from the failure of immune self-tolerance. They can be induced by unknown causes, exposure to infectious agents, medications or after tissue injury. Autoantibodies often are not pathogenic in themselves, with exceptions, for example autoantibodies in autoimmune haemolytic anaemia, Grave's disease and Myasthenia gravis. Autoantibodies can be identified in low-levels in disease-free individuals, however in others they are 'markers' of autoimmune diseases. The autoantibodies associated with rheumatological diseases are listed in Table 1. A circulating form of autoantibody in the serum is the anti-nuclear antibody (ANA) which in the right clinical context can be a marker of connective tissue disease.

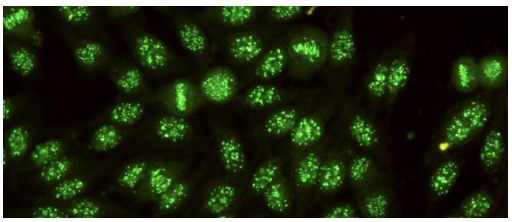
In the ANA assay patient serum is incubated with cultured human epithelial cells (Hep2 or Hep2000) attached to glass microscope slides. The serum is then washed off and any autoantibodies attached to the cells are detected by a fluorescently labelled antibody to human IgG. This pattern of staining is visualized with a fluorescence microscope. The fluorescent pattern of staining is nuclear or cytoplasmic with specific subtypes. For nuclear patterns of staining the titre is reported. The titre relates to the dilution of the patient serum that the antibody concentration is detected at and is a measure of antibody concentration. To be considered "detected" the titre must be 1:80. Titres 1:160, 1:320, 1:640, 1:1280 and 1:2560 or >2560 are reported. Low levels of ANA (titres <1:320) may be pathological but may also occur in disease free individuals and do so more commonly with advancing age.

The fluorescence pattern of ANA staining has clinical associations as illustrated by Figure 1. To further clarify the ANA antigens, performing assays to detect antibodies to the extractable nuclear antigens (ENA) and double stranded DNA (ds-DNA) can be helpful. There is screening assay for ENA and if positive, further characterization is performed (See Table 1). The Hep2000 epithelial cells have been engineered to hyper-express SSA a type of ENA which is the most common autoantibody detected by the ENA test. A dense fine speckled ANA pattern is negatively associated with connective tissue disease. In this case ENA and ds-DNA testing is recommended.

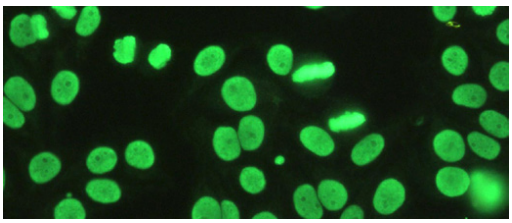
Table 1 Rheumatological disorder and associated Antibodies (Ab)

Test name	Disease	Clinical Features	Significance of Laboratory Result
Rheumatoid Factor (*RF*)	Rheumatoid Arthritis	Inflammatory polyarthritis	The presence of higher titres has prognostic significance.
Cyclic citrullinated polypeptide Ab (*CCP Ab*)	Rheumatoid Arthritis	Inflammatory polyarthritis	May precede RF. Predicts more aggressive erosive disease. Level reflects inflammatory activity.
Anti-nuclear antibody (*ANA*)	Connective tissue disorders	SLE, scleroderma, other connective tissue disorders	Positive in the majority of connective tissue disease, with low levels being in the initial phase of the illness. ANA titres may vary with the disease activity in some patients.
Double stranded DNA (*ds-DNA Ab*)	SLE	SLE	Present in patients with renal or cerebral lupus and when present are a useful marker of disease activity.
Chromatin or nucleosome Ab (*Chromatin Ab*)	SLE Drug induced lupus		Identifies antibodies to histone as well as ds DNA. Inferior disease activity marker to dsDNA.
Antibodies to Extractable Nuclear Antigens (*ENA*) See disease associations for subtypes	***Connective tissue disorder subtypes*** - **Ribosomal-P:** Seen in 15% SLE patients, highly specific, more common in patients with neuropsychiatric manifestations. - **Sm:** Highly specific, only positive in 10% SLE patients. - **RNP:** 30% patients with SLE and the only positive Ab in patients with mixed connective tissue disease. - **Scl-70**: 70% patients with diffuse scleroderma and 15% with CREST syndrome or limited scleroderma. - **Jo-1:** 30% of patients with polymyositis-dermatomyositis and may indicate a greater likelihood of associated pulmonary fibrosis. - **SSA and SSB:** Both, primary Sjogren syndrome. Women with SLE with both of these Ab are more likely to develop sicca symptoms, cutaneous involvement and their babies may be more at risk of developing congenital heart block as well as transient cutaneous lupus. SSA alone: May either have SLE or Sjogren syndrome. SSB alone: Usually have milder disease.		
Anticardiolipin Antibody (*ACL Ab*) Anti-phospholipid Antibodies (*Lupus anticoagulant*)	Anti-phospholipid syndrome	Thrombosis, miscarriages	Ab to cardiolipin of (IgG and less commonly IgM) isotypes have been found in increasing frequency in women with excess fetal loss as well as venous and arterial thrombosis. Higher levels, particularly IgG isotype with beta2-glycoprotein I antibodies are more frequently correlated with disease. Many, but not necessarily the majority of persons have connective tissue disorders.

Figure 1: Examples of ANA patterns



Centromere patterns suggest a limited form of scleroderma known as CREST syndrome (Calcinosis, Raynauds, oEsopageal involvement, Sclerodactyly and Telangiectasia).



Homogenous pattern is common in patients with SLE, drug induced lupus and autoimmune chronic active hepatitis.

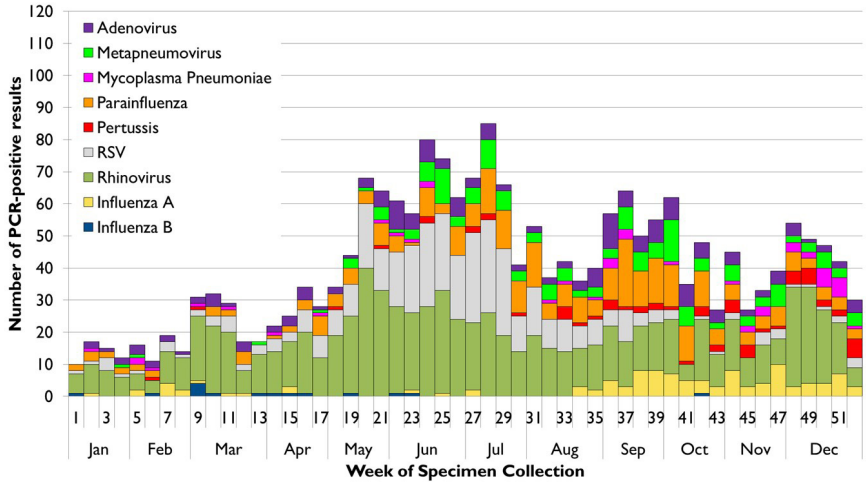
In conjunction with the clinical picture, ANA, ENA and ds-DNA Ab testing can be a valuable tool for the diagnosis and monitoring of autoimmune disorders. The presence of an autoantibody in more than low levels can assist in the prediction and diagnosis of disease and the absence of autoantibodies, in some cases, can exclude some autoimmune diseases. Autoantibody titres may fall when the disease has been present for a long time and may be unreliable in immunodeficient patients. Occasionally clinical features of the disorder may proceed the development of autoantibodies. This sometimes occurs in connective tissue disorders. If the clinical features are suspicious for connective tissue disease repeat testing after 4-6 weeks should be considered.

Autoantibodies by immunologists Dr Karl Baumgart and Dr Andrew Broadfoot in conjuction with Dr Roslyn Malley.

Respiratory virus PCR testing – an update

As winter approaches it is likely that increasing numbers of patients will be presenting with viral respiratory tract infections over the next few months.

Although Influenza is often a major concern, it is important to remember that there are a range of other viral agents that circulate in the community throughout the year, as is shown in Tasmanian data for 2018.



Clinical presentations vary from the trivial, where laboratory investigations are not required and management is symptomatic, to potentially severe Influenza Like Illness (ILI).

An ILI is defined as an illness characterised by: The sudden onset of at least one of: Cough (new or worsening), sore throat, shortness of breath and at least one of: Fever, malaise, headache, myalgia

At Hobart, Launceston and North West Pathology we currently offer two types of respiratory virus PCR testing:

1. Rapid Influenza and RSV PCR

See "Respiratory viruses PCR – Hospital or high risk patients" in the Test Collection Manual)

- Provides a rapid (same day) result for Influenza A/B and RSV only
- An expensive test, performed on a "stat" basis
- Requires a nasopharyngeal specimen collected using a propriety flocked swab and liquid transport media
- Is currently available for patients in high risk settings where proven infection with Influenza (or RSV) would result in changes in patient management, or be of infection control or public health significance i.e.

  - Hospital inpatients and those attending emergency departments
  - Patients in nursing homes or other long term care facilities where there is the potential for an Influenza outbreak. *(Note: as per recent Public Health advice approximately five specimens per institution should be sufficient to establish the presence of an outbreak in this setting)*

In community patients at high risk of Influenza, where rapid testing may be indicated (e.g. Children <2, pregnant women, patients immunosuppressed due to chronic disease or immunosuppressive therapy) please contact your local laboratory to discuss specimen collection arrangements.

2. Respiratory virus (multiplex) PCR

(See "Respiratory viruses PCR" in the Test Collection Manual)

- Covers wider range of potential pathogens (Influenza A/B, RSV, Human Metapneumovirus, Rhinovirus, Parainfluenza virus)
- A less expensive test, performed in batches at an external reference laboratory, therefore a longer turnaround time
- Most appropriate for general use in non-high risk settings
- Requires nasopharyngeal swab collected with a flocked swab without transport media and /or dry throat swab.

Dr McGregor or Dr Malley can be contacted at Hobart Pathology to provide any clarification.

Dr Alistair McGregor
MBChB, FRACP, FRCPA
Clinical Microbiologist

Acknowledgements: Tasmanian respiratory virus data was kindly provided by the Department of Microbiology at the Royal Hobart Hospital and the Communicable Diseases Prevention Unit.

Dr Daniel Owens
BMedSci, MBBS (Hons), FRCPA, FRACP, GAICD
State Medical Director