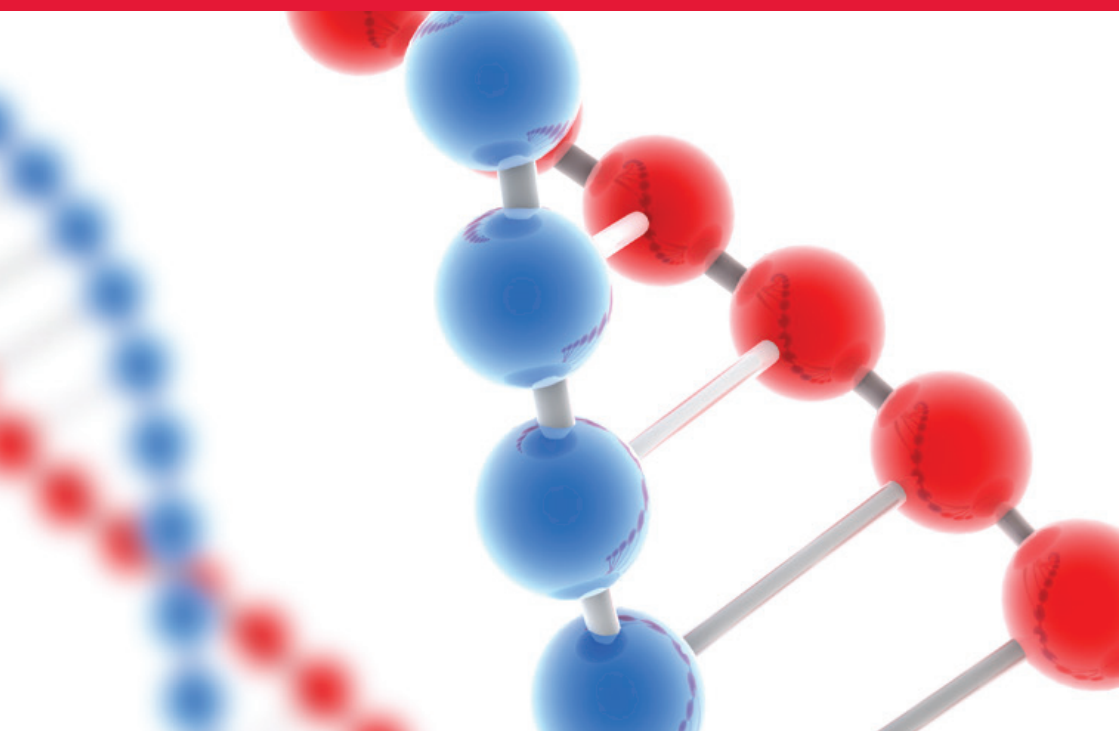
CAPITAL PATHOLOGY

"We take it personally"

PATHOLOGY HANDBOOK



CAPITAL
PATHOLOGY

"We take it personally"

Capital Pathology ABN 49 452 500 422
2 Makin Place, Deakin, ACT, 2600, Australia

All rights reserved. No part of this manual may be reproduced, stored in a retrieval system, or transmitted in any form or any means without Capital Pathology's written consent.

This Handbook has been prepared by Capital Pathology and every care has been taken in its compilation. Capital Pathology shall not be liable for any error or omission contained herein or arising from usage of material contained in this Handbook.

This Handbook remains the property of Capital Pathology.

Published–2012

Edited by Christina Vett-Joice
Printed by Buckner Printing Company

Table of Contents

Acknowledgements iii

Foreword iv

How to use this handbook v

General information

- Pathologists vi
- Overview xvi
- Services xvii
- Contact Us xviii
- Doctors Service Centre xx
- Collection Centres xxi
- Specimen Collection xxiii
- Courier Services xxiv
- Requesting Pathology Tests xxv
- Laboratories xxvii
- Results xxviii
- Reports xxix
- Billing Policy xxxi
- Education xxxiv
- Publications and website xxxv
- Other Services xxxvi
- Corporate services xxxvii

A–Z Listings

Acknowledgements

This is the fifth edition of the Capital Pathology handbook, and the production of each new edition is a huge task. Also, each edition builds on the contributions that others have made to earlier editions so that the current handbook is a testament to the hard work of many teams of people over many years.

There have been champions of the fifth edition handbook within Capital Pathology, just as there were champions of its predecessors.

Dr Paul Whiting, Director of Clinical Pathology has again been passionate about the urgency of keeping the handbook relevant and current for our referring doctors and both myself and Dr Jane Twin, Director of Cytology have also contributed extensively to our relevant sections.

Christina Vett–Joice (CV) has once again been the driving force behind the fifth edition. She has been cheerfully assisted by the scientific and administration staff of Capital Pathology, many of whom have helped produce parts of this handbook in their own precious non-work time.

Many people throughout Sonic Healthcare have been incredibly generous in their contributions to both this and earlier editions. They have shared their knowledge, experience and advice with us and we thank you. It is wonderful to have a large team of world experts at our disposal at Capital Pathology and we will continue to test the limits of your kindness and availability!

I would also like to thank the companies who have sponsored the fifth edition. Without your generosity, we would not have been able to produce a reference work of such quality and scope, and we appreciate your faith in our handbook and your support to us.

Finally I would like to sincerely thank those who have fundamentally made this handbook possible through their ongoing support and loyalty to Capital Pathology – the doctors, nurses and patients who daily trust us with the privilege of being part of their health care team. I assure you that we take our responsibilities to you very seriously. In fact “We Take it Personally”.

Dr Ian Clark

M.B., B.S., F.R.C.P.A., M.I.A.C., F.A.S.C.P., F.A.I.C.D.

Chief Executive Officer

2012

Foreword

We are constantly reminded that we are in the midst of an explosion of knowledge and information. This is nowhere typified better than in the ever-changing field of pathology. This is the fifth edition of the handbook provided by Capital Pathology to medical practitioners, nurses, practice managers, receptionists, hospital staff and others who use our practice. The handbook provides a concise guide to the range of services provided by Capital Pathology, as well as assisting to answer some of the clinical questions we are so often asked.

Capital Pathology has a long history, over 40 years, of providing pathology services to the ACT and surrounding districts. We are very aware of the privilege and responsibility associated with being an integral part of the medical community in our region, and this can be summarised in our motto “We take it personally”.

For us in the potentially rarefied atmosphere of an efficient modern pathology laboratory, this means that we constantly remind ourselves that associated with every specimen, every report, every piece of anonymous data, every phone query, there is a person who rightfully expects us to treat them “personally”.

This also means that our supporting doctors and their teams can rely on each of us at Capital Pathology, irrespective of our role, to behave as part of a medical practice where we are each known by you, and are personally committed to ensuring the best possible service for you and your practice.

I hope that this Handbook will help us improve our service to you. I also hope it will assist you and your staff to achieve the ultimate aim of us all: to improve the health of our patients and our community.

Dr Ian Clark

M.B., B.S., F.R.C.P.A., M.I.A.C., F.A.S.C.P., F.A.I.C.D.

Chief Executive Officer

2012

How to use this handbook

All listings are in alphabetical order, whether analyte, test name, disease, clinical topic, procedure, drug name, specimen type or pathogen.

Reference Ranges

For some common analytes the reference ranges are similar throughout Australia (e.g. sodium, potassium). For hormones and other tests where methodology can vary from one laboratory to another, the reference ranges can differ quite markedly. Even in the same laboratory, ranges may change from time to time as new methods are introduced. In this handbook you are provided with common reference ranges.

For other tests, please consult the reference ranges provided with all reports.
Please note reference ranges are correct at time of printing.

A reference range can be defined as a range derived from measuring an analyte in a reference population using a reference method. The reference interval then is usually taken to include the mean +/–2 SD i.e. 95th percentile. This implies that 5% of the normal population will fall outside these limits.

In clinical practice, a patient's results can be examined in different ways, either by comparison to a reference interval, by comparison to their own known previous values, or by consideration of desirable goals, e.g. aiming for a cholesterol of X since this has been shown to have benefit in terms of mortality / morbidity from cardiovascular disease. Some useful ranges are not reference ranges at all, but rather therapeutic ranges, e.g. warfarin control by INR.

Units

S.I. units are used throughout.

Notes on Interpretation

Comments in this handbook are intended as a guide only. Any additional queries may be directed to the appropriate pathologist.

Brief clinical details help immeasurably with interpretation,
e.g. *routine, wt–loss 1/12, pale and tired 3/12, on T4, on phenytoin, ? hepatitis.*

Pathologists

In my opinion, pathologists are undoubtedly the central focus of a successful pathology practice.

At Capital Pathology, specialist pathologists are not only responsible for management decisions, but are intimately involved in all clinical and ethical matters. Our pathologists pride themselves on their close clinical relationship with our referring doctors, and they recognise the importance of providing a comprehensive consultative and educational service. This is exemplified on a day-to-day basis by an expanding range of interpretive comments on reports. In order to optimise patient care, we understand the importance of discussion with clinicians through phone calls as well as providing advice and feedback at personal visits. Capital Pathology's specialist pathologists have a long history of commitment to continuing education for family doctors and specialists through educational seminars and regular newsletters.

Even more importantly, the continual guidance provided by our pathologists determines the quality and integrity of our whole service. Although our practice obviously must remain commercially viable, the medical influence provided by our pathologists ensures that purely commercial considerations never take precedence over ethical and professional standards.

Dr Ian Clark

M.B., B.S., F.R.C.P.A., M.I.A.C., F.A.S.C.P., F.A.I.C.D.

Chief Executive Officer

Dr Ian Clark

M.B., B.S. (Hons), F.R.C.P.A., M.I.A.C., F.A.S.C.P., F.A.I.C.D.
Chief Executive Officer
Director of Histopathology

Ian graduated with first class honours from Sydney University in 1984. He was full–time Senior Tutor in the Pathology Department of Sydney University before commencing his Registrarship in Histopathology at the Royal North Shore Hospital of Sydney, where he also developed expertise in fine needle aspiration cytology. He completed his Fellowship examinations in 1990, after which he joined the Douglass Laboratories practice in Sydney as a full–time specialist pathologist.

In 1996 Ian was appointed Deputy Director of Histopathology of the newly merged Douglass Laboratories and Hanly Moir Pathology. He joined Capital Pathology in July 1998 as Director of Histopathology, was appointed Medical Director in 2001 and became CEO in 2007. In 2005 Ian was appointed Senior Clinical Lecturer at the ANU Medical School.

Ian is a Histopathologist and Cytopathologist with keen special interests in dermatological, gastrointestinal, gynaecological and genito–urinary pathology. He also oversees the application of flow cytometry to cytological and histological specimens.

Ian is an active member of many medical associations and societies, including the Australian Society for Colposcopy and Cervical Pathology, the Australasian Society for Breast Disease and the Australian Dermatopathology Society. He holds Membership of the International Academy of Cytology and is a Foreign Fellow of the American Society of Clinical Pathologists.

He has participated in a number of research projects, and has spoken at many conferences, including the International Academy of Pathology. Ian was President of the Australian Association of Pathology Practices from 2007 to 2009 and is the Honorary Secretary of the Royal College of Pathologists of Australia. He is also the Pathology craft representative on the ACT AMA.

The fundamental importance of medical leadership underpins Ian's role as Chief Executive Officer, and he is committed to maintaining the high quality and ethical standards for which the practice is renown.

If you have any enquiries for Ian, please do not hesitate to contact him on 02 6285 9800.

Dr Jane Twin

M.B., B.S., F.R.C.P.A., F.I.A.C., Dip. Cytopath.
Director of Cytopathology

Jane is a graduate of the University of Tasmania and undertook her Pathology training at Royal Hobart Hospital. Following attainment of the Fellowship of the Royal College of Pathologists of Australasia in 1983, Jane worked as a researcher and lecturer at the University of Tasmania with an appointment to the Royal Hobart Hospital as a provider of Cytology Services. After training with Dr Svante Orell in Adelaide, Jane moved to Hobart Pathology in 1989 to set up a Fine Needle Aspiration Service. This service grew rapidly and was augmented by provision of FNA and Histopathology to BreastScreen Tasmania. Other involvement in BreastScreen Tasmania included her appointment as Tasmania's representative on the National Pathology Q–group and a long term role as Pathology adviser and auditor. Jane was also involved in the setting up of the Tasmanian Cervical Cytology Register and continued on the Technical Working Party until leaving Tasmania in December 2001.

Other achievements include attainment of further qualifications in Cytopathology by examination, including Fellowship of the International Academy of Cytology (1992) and the Royal College of Pathologists of Australasia Diploma of Cytopathology (1997). Jane has been involved with the Australian Society of Cytology at a National Level since 1991 as a Councillor and on the Board of Examiners, and has a strong interest and experience in Gynaecological Pathology. Jane is a member of the advisory committee for the Cytology Quality Assurance Program and is also a member of the RCPA cytology discipline advisory committee. She is also a member of the ACT Papsmear Register management committee. At Capital Pathology, Jane continues her interests in cytology and the histological aspects of gynaecological and breast pathology while contributing generally to the histology department.

If you have any enquiries for Jane, please do not hesitate to contact her on 02 6285 9867.

Dr Paul Whiting

M.B., B.S., F.R.C.P.A., M.A.S.M.
Director of Clinical Pathology

Paul studied for his medical degree at the University of Melbourne, graduating in 1987. After residency and a year in General Practice, Paul commenced his pathology training at Geelong Hospital. He moved to Canberra in 1996 where he completed his general pathology training. He was admitted as a Fellow of the Royal College of Pathologists of Australasia (RCPA) before joining Capital Pathology as the Director of Clinical Pathology in 1998. In this role, he supervises the haematology, biochemistry, immunology and microbiology laboratories, and is responsible for ensuring that the highest quality standards are always met. Paul encourages consultation on the wide variety of problems in clinical medicine, and is always available to discuss pathology results.

Paul is involved with many professional associations and is on several hospital advisory committees. He is a member of the Haematology Society of Australia, the Australian Association of Clinical Biochemists, the Australian Society for Microbiology and is also a member of the American Society of Clinical Pathologists. Paul is active with many quality assurance interests, and acts as a referee in the RCPA external quality assurance programs. In Canberra he maintains a continuing interest in medical education and regularly presents to local specialist and general practitioner audiences.

If you have any enquiries for Paul, please do not hesitate to contact him on 02 6285 9895.

Dr John Docker

M.B., B.S., D.C.P. (London),.D.R.C. Path (UK), F.R.C.P.A.

Specialist Pathologist

John graduated from Sydney University in 1969 and did his early training in general pathology at the Charing Cross Hospital in London, where he was subsequently appointed Senior Registrar and Lecturer at the Charing Cross Medical School. John returned to Sydney with his young family in 1975 and after a year at St. Vincent's Hospital and the Prince Henry / Prince of Wales Hospitals, he qualified for his Fellowship and returned to his boyhood town of Goulburn in 1978. Since then he has practised in Goulburn as a General Pathologist with a strong emphasis on histopathology. John joined Capital Pathology on a part-time basis in June 1999.

If you have any enquiries for John, please do not hesitate to contact him on 02 6285 9867.

Dr Juli Ferguson

M.D., Ph.D., F.C.A.P.

Specialist Histopathologist

Juli graduated with a BS cum laude from the College of St. Mary, Omaha, Nebraska, U.S. in 1971. She entered graduate school completing her MS (1974) and Ph.D. (1977) in the field of Biochemistry at the University of Nebraska. She then entered medical school, completing her M.D. program in 1981 with acceptance into the Alpha Omega Alpha medical honour society. After her Registrarship at the University of Iowa Hospitals and Clinics, she joined Physicians Associates in Grand Island, Nebraska in 1985. She became a fellow of the College of American Pathologists in 1989.

Juli has been with Capital Pathology since immigration to Australia in 1989. Juli's special interests are urologic pathology and dermatopathology. She is a member of the American Society of Clinical Pathologists, College of American Dermatopathology Society and International Academy of Science.

If you have any enquiries for Juli, please do not hesitate to contact her on 02 6285 9867.

Dr Peter Harper

M.B., Ch.B.(N.Z.), F.R.C.P.A.

Specialist Histopathologist

Dr Peter Harper graduated from the University of New Zealand (Otago) and his residency was undertaken in large and small teaching Hospitals and a year in remote General Practice before commencing his Pathology career.

At the beginning of 1968 he migrated to Australia pursuing Pathology under Dr Baird in Biochemistry and Haematology at the Royal Melbourne Hospital. At various times he has been tutor in Pathology at Melbourne and Monash Universities.

He then entered Private Pathology Practice selling his own Practice in 1987 and joining Gribbles Pathology as Head of Histopathology. He undertook a stint in country NSW and Victorian public and Private Practice and was a Coronial Pathologist in NSW and Victoria.

He again entered private practice in 1992 with Macquarie Pathology and Mayne Health and in 2005 joined SDS prior to joining Capital Pathology in Canberra in 2008.

Peter has a keen interest in skin pathology, and welcomes discussion with referring doctors. He can be contacted on 02 6285 9867.

Dr Tracey Lu

M.B., F.R.C.P.A., B.M.L.S

Specialist Histopathologist and Cytopathologist

Tracey has dual degrees in both Medicine and Medical Laboratory Science. She obtained her primary medical degree from Fujian Medical University China in 1986 and worked there when she graduated as a full-time assistant lecturer, researcher and clinical anatomical pathologist before immigrating to New Zealand.

After settling in NZ, Tracey gained a Bachelor degree in Medical Laboratory Science from Auckland University of Technology and worked as a Scientist at Labplus, Auckland Hospital whilst she passed the New Zealand Medical Registration Exam (NZREX). She then worked as a House Surgeon at a New Zealand teaching hospital before commencing her Pathology Registraship in 2004.

Tracey trained in Anatomical Pathology in Auckland and Tauranga NZ for a total of five years. She worked in rotations between three public hospitals and one private laboratory and gained experience in a wide variety of subspecialties, including cytopathology, breast, gynaecology, urology, GI, cardiothoracic, neuropathology, head and neck, bone and soft tissue, paediatric, forensic, and lymphoma pathology.

Tracey has joined Capital Pathology as a specialist Histopathologist and Cytopathologist after she was awarded her Fellowship from The Royal College of Pathologists of Australasia.

If you have any enquiries for Tracey, please do not hesitate to contact her on 02 6285 9867.

Dr Sumi Ranjit

MBBS, DCH, MRCP (Paediatrics), FRCPA (Haematology), FRCPA (Anat Path)

Specialist Haematologist and Anatomical Pathologist

Sumi brings with her a rich and diverse medical background stretching over a period of more than 20 years.
After obtaining her primary medical qualification and training in Paediatrics from Madras University in India, she moved to the UK to undertake training in Paediatric Haematology at the Royal Manchester Children's Hospital and St James's University Hospital in Leeds. Her Haematology skills were further honed while working as Registrar and Research Fellow at Prince of Wales Hospital in Sydney (1999–2002).

After obtaining her specialist qualifications in Haematology (FRCPA) in 2002, Sumi worked as a specialist Haematologist in Canberra and overseas. However, in order to fulfil a long felt desire to gain more in depth knowledge, Sumi undertook further training in Anatomical Pathology at The Canberra Hospital and at Capital Pathology, passing her final examinations and obtaining her Fellowship in Anatomical Pathology in 2010.

Sumi co-edited a Handbook of Common Clinical Emergencies while still a medical student in India. More recently, she has made several presentations at national and international meetings, including the American Society of Haematology and the Australian Society of Cytopathology. She has also held the post of Associate Lecturer at the Australian National University Medical School where she was involved in teaching medical students.

Sumi and her family enjoy living in Canberra and she is passionate about the environment. Her other interests include reading, travelling, cooking and natural history.

If you have any enquiries for Sumi, please do not hesitate to contact her on 02 6285 9867.

Dr Melissa Robbie

M.B.B.S., F.R.C.P.A.

Specialist Histopathologist and Cytopathologist

After graduating from medical school at the University of Melbourne in 1983, Melissa began her post-graduate training at Queen Victoria Hospital (later Monash Medical Centre), where she entered the Pathology training program. This was followed by further registrar positions at Monash Medical Centre and Dandenong District Hospital. She received her fellowship from the Australian Royal College of Pathologists in 1991 and had her first consultant appointment in the General Pathology Service at Ballarat Base Hospital. Melissa has a keen interest in teaching and subsequently spent three years teaching 2nd and 4th year medical students at Monash Medical University Medical School, as well as consulting in Anatomical Pathology at Dandenong Hospital and Eastern Suburbs Pathology Services.

From 1995 until 2000, Melissa was Senior Pathologist / Deputy Director of Pathology at Mercy Hospital for Women & Werribee Mercy Hospital. From 2000 to early 2005, she worked at St Vincent's Hospital, Melbourne. In addition, Melissa continues to hold an honorary appointment at Peter MacCallum Cancer Institute, where for 9 years she was central reviewer for Gynaecologic Pathology and now has continuing research collaborations investigating the mechanisms of breast and ovarian cancer.

Melissa has special interests in Gynaecological & Obstetric Pathology, Cytopathology, and Gastro-intestinal pathology.

She is a member of the Australian Cancer Network's committee on ovarian cancer, which recently produced the first national guidelines on ovarian cancer management for doctors and patients. Melissa also contributed a chapter to an international collaborative work on *Controversies in Gynecologic Cancer*, edited by David Gershenson et al (Churchill Livingstone 2004) and is now a member of the Editorial board for the journal Gynecologic Oncology.

If you have any enquiries for Melissa, please do not hesitate to contact her on 02 6285 9867.

Overview

Capital Pathology is a fully NATA accredited Category GX medical testing laboratory that provides quality pathology services to general practitioners, medical specialists, nursing homes, private hospitals and the community in the ACT, South Coast, Snowy Mountains and Goulburn regions.

We are proud to have provided services to the ACT and adjoining regions for over 40 years.

Our purpose built, state of the art main laboratory is situated in Deakin, right in the heart of Canberra. We also have regional laboratories conveniently located in both Bega and Goulburn. Our practice currently employs in excess of 300 local staff. Our practice encompasses all disciplines of pathology. Our specialist pathologists, scientists and technical staff are available for consultation and general enquiries.

Capital Pathology is a division of Sonic Healthcare Limited, an Australian owned, publicly listed company.

Our Mission

We help doctors help patients by providing specialist pathology services.

Our Values

Commit to Service Excellence

To willingly serve all those with whom we deal with unsurpassed excellence.

Treat Each Other with Respect and Honesty

To grow a workplace where trust, team spirit and equity are an integral part of everything we do.

Demonstrate Responsibility and Accountability

To set an example, to take ownership of each situation to the best of our ability and to seek help when needed.

Be Enthusiastic about Continuous Improvement

To never be complacent, to recognise limitations and opportunities for ourselves and processes and to learn through these.

Maintain Confidentiality

To keep all information pertaining to patients as well as professional and commercial issues, in strict confidence.

Services

Mission Statement

'We help doctors help patients by providing specialist pathology services'

We provide a comprehensive range of pathology testing for doctors and their patients serving:

- General Practitioners
- Specialists
- Private Hospitals
- Nursing Homes, Aged Care, Supported Care
- Day Surgeries.

Our laboratory offers:

- Main laboratory open 24 hours, 7 days a week
- Specialised Pathologists on-site
- Doctors Service Centre dedicated to providing results and assisting with enquiries
- A comprehensive range of routine and emergency pathology testing
- Fine needle aspiration service available on-site
- Regional laboratories in Bega and Goulburn
- Rapid turn around of results
- Innovative report formats
- Personalised options in reporting
- Statistical analysis of various reports available
- Conveniently located collection centres in the local and surrounding districts open extended hours and offering appointments
- Home visits available if required
- Extensive courier network
- Electronic access and downloading of results
- Publications and ongoing education for referring practitioners and their staff.

We also offer a broad range of services for corporate and industrial customers including:

- Occupational and environmental testing
- Insurance and corporate testing
- Clinical trials
- Food and water testing.

Our staff are always available to support practitioners in their clinical practice.

Our Pathologists encourage doctors to contact them for information or advice at any time.

Our Client Services Department is available to assist all our customers with enquiries, issues and information.

Contact Us

2 Makin Place, Deakin, ACT, 2600

The laboratory provides services 24 hours a day, 7 days a week.

Doctors Service Centre (DSC)
Results and General Enquiries..... 02 6285 9803

Front Reception Switchboard..... 02 6285 9800

Accounts 02 6285 9888
Chief Executive Officer 02 6285 9836
Client Services..... 02 6285 9805/9802/9801
Collection..... 02 6285 9881
Couriers 02 6285 9877
Cytology..... 02 6285 9868
Histology..... 02 6285 9867

Home Visits:

Northside 02 6251 5121
Southside 02 6281 7277
Central 02 6285 9885

IT Services..... 02 6285 9860

Main Laboratory 02 6285 9811

Microbiology 02 6285 9846

Pathologists

CEO/Medical Director/Director of Histopathology .....Dr Ian Clark 02 6285 9800

Director of Clinical Pathology .....Dr Paul Whiting 02 6285 9895

Director of Cytopathology.....Dr Jane Twin 02 6285 9867

Stores 02 6285 9813

Fax

Administration..... 02 6281 1941

Doctors Service Centre 02 6285 2946

Website.....www.capitalpath.com.au

Email .....info@capitalpath.com.au

Doctors Service Centre (DSC) 02 6285 9803

Results Enquiries

If you wish to phone us for results or any general enquiry:

Doctors Service Centre (DSC) 02 6285 9803
Toll free–outside ACT 1800 807 556

Please supply surname, first name and date of birth of the patient, and the name of the referring doctor when requesting results.

Alternatively you may wish to speak with a pathologist for interpretation of a result. If so, please refer to the contact phone numbers on the ‘Contacts Page’.

Confidentiality

To maintain patient confidentiality, the staff in the Doctors Service Centre and other laboratory departments make every effort to ensure that only authorised recipients are given results.

Capital Pathology apologises for any inconvenience this may cause.

Collection Centres

Appointments are not required for most tests, however for your patients' convenience appointments are available at most of our Collection Centres.

Please phone specific Collection Centre for further details.

Tests that do require an appointment include:

- Glucose Tolerance Test (GTT)
- Helicobacter Breath Test
- Holter Monitors
- 24 Hour Blood Pressure.

ACT

AINSLIE 02 6249 1940
100 Wakefield Gardens, Ainslie

BARTON 02 6273 4505
5/3 Sydney Avenue, Barton

BELCONNEN 02 6253 0146
Unit 12 North Point Plaza
8 Chandler Street, Belconnen

CALVARY 02 6251 5121
Suite 4, Calvary Clinic,
Haydon Drive, Bruce (rear of Calvary Hospital)

CHARNWOOD 02 6258 9197
123 Tillyard Drive, Charnwood

CIVIC 02 6248 0899
34 Marcus Clarke Street, Civic

DEAKIN 02 6285 3765
Peter Yorke Building Plaza, Calvary John James Hospital
173 Strickland Crescent, Deakin

DICKSON 02 6248 7804
Dickson Professional Centre,
Cnr, Antill and Cowper Streets, Dickson

GARRAN 02 6281 7277
Brindabella Specialist Centre,
Dann Close, Garran

GUNGAHLIN 02 6242 8328
Unit 132, Hinder Street,Gungahlin

HAWKER 02 6255 2241
Unit 7, Birubi Chambers,
Cnr, Beetaloo Street and Hawker Place, Hawker

LANYON 02 6294 7409
Unit 5/3 Sidney Nolan Street, Condor

ACT (continued)

MANUKA 02 6239 5074
Unit 1/21 Murray Crescent, Manuka

PHILLIP/WODEN 02 6293 3860
Suite 11 Corinna Chambers
36–38 Corinna Street

TUGGERANONG 02 6293 3860
Unit 5, Cnr, Anketell and Reed Streets, Tuggeranong

WANNIASSA 02 6296 1644
Unit 2, Erindale Chambers,
Gratton Court, Erindale Shopping Centre, Wanniassa

NSW

BEGA 02 6492 2049
101 Carp Street, Bega

BERRIDALE 02 6452 3150
6 Myack Street, Berridale

COOMA 02 6452 3150
190 Sharp Street, Cooma

CROOKWELL 02 4843 2555
17 Kialla Road, Crookwell

EDEN..... 02 6496 3508
Curalo Medical Centre
60 Princes Highway, Eden

GOULBURN 02 4821 4011
127 Bourke Street, Goulburn

JERRABOMBERRA 02 6299 8642
Unit 12, Jerrabomberra Shopping Village Centre
2 Limestone Drive, Jerrabomberra

JINDABYNE 02 6457 2209
Nugget's Crossing Shopping Centre, Jindabyne

MERIMBULA 02 6495 3045
Unit 2/93 Main Street, Merimbula

MERIMBULA (Sapphire Clinic)..... 02 6495 4879
44 Merimbula Drive, Merimbula

PAMBULA 02 6494 3024
17 Quondola Street, Pambula

QUEANBEYAN 02 6297 3600
1st Floor
23 Antill Street, Queanbeyan

Specimen Collection

Specimen labelling

A correctly labelled specimen is essential. Persons collecting specimens must positively identify the patient, and ensure that there are at least two unique identifiers recorded on the specimen.

1. Patient first and surname.
2. Date of birth (and/ or medical record number for hospital patients).

A correctly labelled specimen should arrive in the laboratory with a completed pathology request form that includes the date and time of collection.

There are exacting requirements for blood bank specimens and paperwork.

See Blood Banking.

There are also specific requirements for all antenatal and prenatal specimens.

1. All pre- transfusion and antenatal and perinatal specimens must be labelled **with surname, given name(s), date of birth, date and time of collection**
2. The **person collecting the blood** is to **sign** or **initial** the **tubes**
3. The **person collecting the blood** is to **sign** a **declaration** on the **request form** similar to the following:

   "I certify that I collected the accompanying sample from the above patient whose identity was confirmed by enquiry and/or examination of their name band and that I labelled the sample immediately following collection."

Home Visits / Hospital Visits

Capital Pathology provides extensive collection services to hospitals and nursing homes. Staff are also available to collect specimens and perform ECG tracings or Holter Monitors from patients at home who are unable to visit a Collection Centre. Please phone the appropriate Collection Centre to arrange a house call for Northside, Southside or Central locations.

Materials for Specimen Collection

Materials necessary for the collection of pathology specimens can be supplied at no charge, subject to Medicare guidelines. A form labelled "Request for Pathology Supplies", which lists the supplies available, can be obtained from your Capital Pathology courier. To order supplies, please fill in this form and send in to the laboratory with your courier or via fax.

Courier Services

Routine Courier Services

An extensive courier service is provided throughout our practice areas in ACT and NSW, seven days a week. Each courier delivers reports and pathology supplies and picks up specimens. A courier will call upon surgeries regularly, or if preferred, only when requested. Please contact Client Services Department on 02 6285 9802 to arrange timings for regular courier pick ups.

Urgent Courier Services

Urgent specimen pick–up is available in emergencies by phoning the Courier Department on 02 6285 9877.

Specimen Boxes

For convenience, a secure specimen box can be installed at surgeries for specimens awaiting out of hours pick–up.

Please phone the Client Services Department on 02 6285 9802 to arrange this service.

Requesting Pathology Tests

Request Forms

Requests for pathology tests may be written on the surgeries' own stationery or on Capital Pathology request forms. The reverse page of pre printed Capital Pathology request forms details information on our current Collection Centres with opening times and services offered at each centre. Information on the reverse of form is correct at time of printing.

For request form replacements, please fill in the re–order form included in the request pad or phone Front Reception on 02 6285 9800.

The following are available:

1. Request forms personalised with the doctor's name and address.
2. Laser request forms for use in printers.

When completing request forms, please supply the patient's full name, sex, date of birth, address and phone number. Relevant clinical history is very helpful to the laboratory, and this information will be included in the completed report.

Requesting Tests

When ordering tests, please make requests as specific as possible. Certain approved abbreviations may be used to order tests or groups of tests. Acceptable abbreviations are published in the Medicare Benefits Schedule book.

Tests do not need to be handwritten, however all requests should be signed by the requesting practitioner.

Verbal Requests

Requests for pathology tests may be phoned to either the Doctors Service Centre on 02 6285 9803 or your nearest Laboratory. However, to ensure that your patients receive Medicare Benefits, the laboratory is required to receive signed confirmation of these tests within 14 days of the verbal request.

The laboratory will send a printed request form, which includes the patient details, for signing and confirmation of the tests requested to the requesting doctor.

Requests for a Series / Rule 3 Exemption

Under certain circumstances, Medicare regulations allow a single signed request form to cover repeat testing over a six-month period. The table below details the maximum number of repeat tests allowed under these circumstances. Once the maximum number of tests has been reached, or the six-month period has expired, a new request form for repeat testing will be required. Please state on the request form “Rule 3 exemption” with the relevant therapy/condition and the tests requested.

Condition/Treatment	Tests requested	Maximum tests per request (6 month period)
Anticoagulant therapy	INR	Unlimited
Methotrexate or Leflunomide therapy	Full Blood Count (FBC) Basic chemistry items including; Liver Function Tests (LFT) Urea, Electrolytes and Creatinine (U&E) CRP	6
Chemotherapy or immunosuppressant therapy	Full Blood Count (FBC)	6
Cis-Platinum or cyclosporin therapy	Urea, Electrolytes and Creatinine (U&E)	6
Lithium therapy	Lithium	6
Gold, penicillamine , clozaril, ticlopidine hydrochloride or sulphasalazine therapy	Full Blood Count (FBC)	6
Patients with chronic renal failure being treated in a dialysis programme conducted by a recognised hospital	Urea, Electrolytes and Creatinine (U&E)	6
Vitamin D therapy	Calcium, Albumin	6
Patients with cancer receiving biphosphonate infusions	Calcium, Phosphate, Magnesium, Urea, Electrolytes and Creatinine	6

Laboratories

Our laboratories have been accredited to ISO 15189 Standard under the accreditation scheme of the National Association of Testing Authorities and the Royal College of Pathologists of Australasia (NATA/RCPA). To maintain this recognition, our laboratories are re-evaluated periodically by NATA to ensure their continued compliance with requirements, and to check that their standard of operation is being maintained.

ACT

2 Makin Place
Deakin 2600

t: 02 6285 9800
f: 02 6281 1941

South Coast

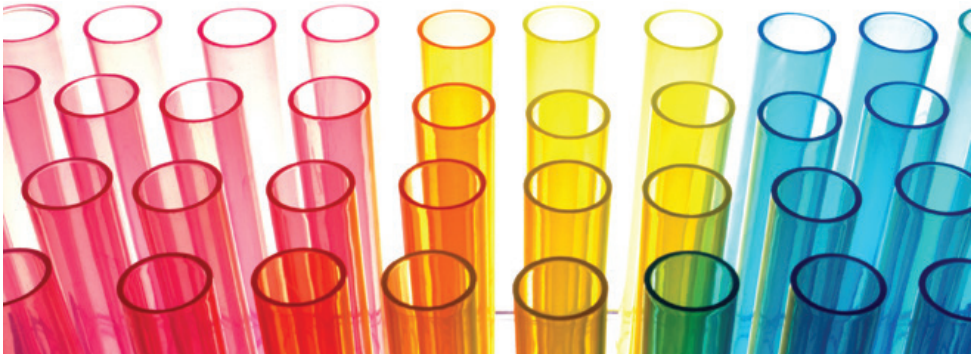
Unit 7/89 Auckland Plaza
Auckland Street
Bega 2550

t: 02 6492 0013
f: 02 6494 7130

Goulburn

127 Bourke Street
Goulburn 2580

t: 02 4821 4011
f: 02 4821 4224



Results 02 6285 9803

Doctors Service Centre.....02 6285 9803

Routine Results

Results are both printed and downloaded continually throughout the day and delivered routinely to practices throughout ACT and NSW. Grossly abnormal results are phoned or faxed to the requesting doctor or surgery.

Urgent Results

Requests marked “Urgent” will receive priority processing and results will be phoned or faxed as soon as available. Please assist us by providing a date and time result is required.

Results by Fax

As a supplement to hard copies, results can routinely be sent directly to a fax machine. To arrange for this service, please contact the Doctors Service Centre on 02 6285 9803.

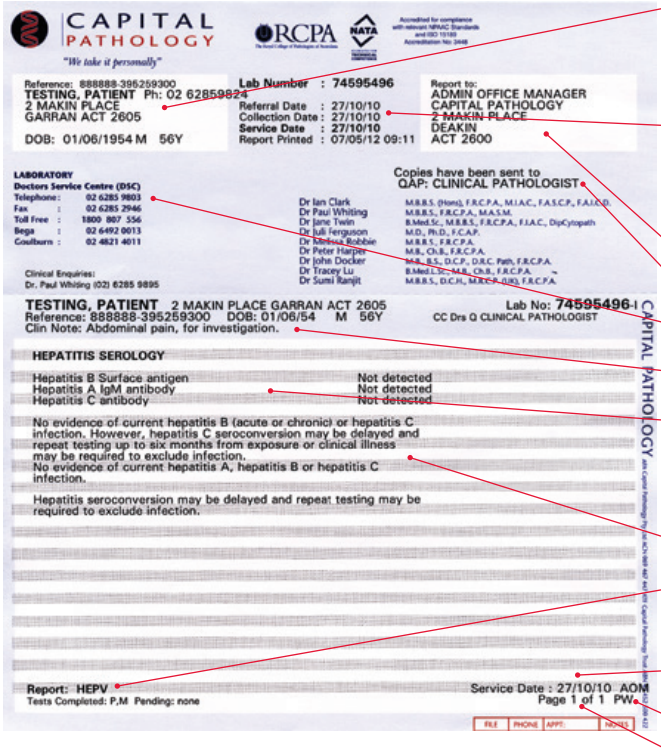
Results by Electronic Download

Results may be downloaded and imported into all major practice management software systems.

Results may also be viewed via the internet using Webster, our secure result viewing application.

For further information on results transfer systems and Webster, please phone IT Services on 02 6285 9860.

Reports: paper and ‘paperless’



Patients Details
Your reference number (if required).
Patients telephone number (if supplied).
DOB, sex and age.

Episode Information
Lab number in bold.
Referral date included.
Collection date included.
Service date in bold (date received in laboratory).

Doctor's Details

Referring doctor or copy to doctor information

Laboratory/Pathologist contact information

Patient Details
Clinical history (if supplied).

Result
Standard format for cumulative results.
Reference ranges given.
Abnormal results bolded and flagged.

Appropriate results information

Laboratory Information
Test code for result shown on this page.

Date of service
Plus recipient initial.

Pathologist Initials

Indicates number of pages and page sequence in report

Preferences of report style are very much an individual choice, and will vary from surgery to surgery. Capital Pathology provides several options for customising reports to suit the requirements of different doctors and other clients.

These report options are available using either A4 or A5 paper size. Please note that a combination of A4 and A5 is not available.

For customising of your reports, please contact the Client Services Department on 02 6285 9802.

“Interims Only”

This allows for reports to be issued as results are available.

“Finals Only”

“Finals Only” reports are produced when all results on a single patient episode number have been completed (excluding Histology, Cytology and tests sent to other laboratories). Only one report is produced when the final test on the patient’s episode is completed. The advantage of this system is the reduction in the number of reports produced requiring filing. However, the report is not generated until the “longest to complete test” is finalised. This type of report is suitable for surgeries that have an electronic download of their pathology reports, so the possibly increased turnaround time of the final printed report will not be significant.

Accumulated Interims

Reports are produced as each test is completed, and include all other reported tests from that particular episode on the report. This means that when the final test is completed, all tests will be included on the report (excluding Histology, Cytology and tests sent to other laboratories). As progressive reports are available as tests are completed, turnaround time of reports is not compromised. However, additional reports and therefore extra paper is produced.

Interims / Finals

Interim reports are produced as each test is completed. A “Final” report is printed once all tests have been completed and includes all tests on the episode. This will result in individual tests being available with no delay in turnaround time and a consolidated report, with all patient results (excluding Histology, Cytology and tests sent to other laboratories), also being produced.

Cumulatives

Cumulative reports are available for certain tests. These list past and current results on the one report and are designed to facilitate monitoring by the practitioner. The most recent results appear on the right hand side of the report.

Duplex (Double–Sided)

Single–sided reports are the standard report format produced. As surgeries use a wide range of record filing systems, a choice of either single or double–sided reports (duplex) is available.

Paperless

Individual doctors, or complete surgeries may decide they no longer wish to receive paper reports and only use electronic copies. If you wish to arrange this, please contact our Client Services Department on 02 6285 9802.

Copies of Reports

If extra copies of reports are required, please indicate the name and address of the recipient in the “copy to” space on the request form. This space may also be used to indicate if the surgery wishes to receive additional copies. If multiple copies of reports are always required, this can be arranged by phoning the Client Services Department on 02 6285 9802.

Billing Policy

Capital Pathology is a private pathology practice without direct government subsidy. Medicare cutbacks over the past decades have meant that average rebates for testing are now 10% less in actual terms and 60% less in real terms than they were in 1985. Over the past decade there have been no Medicare rebate increases, but two sizable cuts occurred in 2008 and 2009. We are, however, aware of the ever increasing fee burden for our patients, so have put in place mechanisms to minimise patient costs.

For out-patient testing, our practice fees are generally those recommended by the AMA, which we currently cap, so that for each episode of testing 'gap' out of pocket costs are limited to $50. This means your patients usually pay no more than $50 for an episode of Medicare rebateable testing after claiming their rebate from Medicare.

Patient Accounts

We have a range of brochures outlining our billing policy for both inpatients and outpatients that you may find of benefit. Please contact us if you wish to obtain copies for your practice.

Payment for accounts may be made by either paying the account directly to Capital Pathology first and then claiming the rebate from Medicare, or by claiming from Medicare first and the patient then forwarding the rebate cheque to us together with any additional payment. Details of payment options are included with the account.

For any further enquiries please contact our Accounts Department on 02 6285 9888 or Client Services Department on 02 6285 9802.

Out-patient Services

We routinely bulk bill Medicare eligible patients who hold the following cards:

- Pensioner cards
- Healthcare cards
- Veteran Affairs.

We also bulk bill:

- Children under 16
- Nursing home patients.

And as directed by the referring doctor.

In-patient Services

In-patient pathology tests are performed for patients who are:

- A private patient in a private hospital or approved day hospital facility
- A private patient in a recognised (public) hospital.

Fees for patients who have eligible cover with a health fund that has a no-gap agreement with Capital Pathology.

Patients who have eligible private health insurance with a Health Fund that has a no-gap agreement with Capital Pathology may have their account billed directly to Medicare Australia and the private health fund. They will not incur any additional out of pocket expenses for tests that are eligible for Medicare rebates. For information on whether specific health funds are covered by such an agreement, please contact our Accounts Department.

Fees for uninsured patients and those covered by a health fund that does NOT have an agreement with Capital Pathology.

Patients who are uninsured or who do not have eligible cover with a health fund will receive an account for their pathology tests. Our fees for in-patient testing are generally at, or below, those recommended by the AMA. When the account is paid, the patient may present the receipt to Medicare Australia and their private health fund to claim their rebate.

Medicare Rebate Eligibility

Please note that some pathology testing is not eligible for a Medicare rebate. Patients who present to our Collection Centres are informed of the approximate cost involved prior to testing taking place.

Pathology tests for the following circumstances are also not eligible for a Medicare rebate:

- employment
- immigration
- insurance
- superannuation
- Work Cover
- visa applications.

In some cases, tests may be sent to a reference laboratory who may send out an individual account. Details are available from individual Collection Centres or the Specimen Reception Department on 02 6285 9873.

Corporate accounts may be initiated where testing is independent of Health Insurance Commission guidelines. Please contact the Collection Manager on 02 6285 9881 or access Corporate Services at our website on www.capitalpath.com.au

The Coning System

In 1995 Federal Government budget changes saw the introduction of a scheme in which Medicare payments cut out after ***three pathology items*** on any single episode ordered by General Practitioners. This change should not have restricted the ordering of pathology tests in any way (i.e. there is no need for doctors to change their usual pattern or diagnostic approach). With the greater complexity of modern medicine and the ageing population, the number of patient episodes with more than three requested items has been steadily increasing. However, we absorb the cost of tests beyond the three items so the patient suffers no extra out-of-pocket expenses.

Pap smears are not included in the coning scheme.

Patient Episode Initiation Fees (P.E.I.)

This fee was introduced by the Health Insurance Commission in 1992 in conjunction with reduced Medicare benefits for certain pathology tests.

An initiation fee is designed to:

1. Represent costs other than those directly involved in the actual test procedure and includes transporting specimens to the laboratory, collecting samples, forwarding the completed report to the referring doctor and administration costs.
2. Describe to the Health Insurance Commission how the pathology service originated. e.g. Collection Centre, doctor collection or hospital collection.

A pathology provider can charge only one initiation fee per referral unless a Rule 3 Exemption applies. Please see section entitled ‘Requests for a Series / Rule 3 Exemption’ under “Requesting Pathology Services”.

The fee is claimable from Medicare along with the fee/s for the actual pathology test/s.

Education

Capital Pathology has a commitment to provide ongoing education to medical practitioners, practice and nursing staff.

Specialists

Our specialist Pathologists run regular educational sessions throughout the year with various groups of specialists to review specific cases of interest.

General Practitioners

We offer regular educational workshops throughout the year on various interesting and topical subjects.

Practice and Nursing Staff

Workshops are held regularly by Capital Pathology Senior Scientists throughout the year.

For more information on any of these educational sessions please contact the Client Services Department on 02 6285 9802 or access the website on www.capitalpath.com.au

Publications and website www.capitalpath.com.au

Capital Pathology aims to provide up to date information on the range of services provided by our practice, plus publish articles relating to new developments and changes in pathology and health in general.

Patient Information Sheets

Patient Information Sheets are available regarding a number of common test procedures and 'What is pathology?' These are available on our website, from individual Collection Centres or by contacting our Stores Department on 02 6285 9813.

Billing brochures

Information regarding billing for both inpatients and outpatients are available on request. Please contact Client Services Department on 02 6285 9805 if you would like copies.

Doctor's Newsletters and Capital Letter

These publications are printed regularly throughout the year and are available on our website, or by contacting the Client Services Department on 02 6285 9802.

Doctor's Handbook

Copies are available in hard copy, CD or on the website. Please contact the Client Services Department on 02 6285 9802 if you would like copies in either format.

Guides

Laminated guides are available on a range of subjects including:

Vacutainer Guide
Swab Guide
Paediatric Tube Guide
Calenders
Collection Centre Listings
Reference Ranges
Test Abbreviations

Please contact the Client Services Department on 02 6285 9802 if you would like copies of any of these.

Kid's Courage Pack's

We understand that having a specimen taken can be a stressful procedure, particularly for our younger patients. As such we have developed a 'Kid's Courage Pack' which contains a number of activities to help children during this time. These are available at all our Collection Centres.

Website

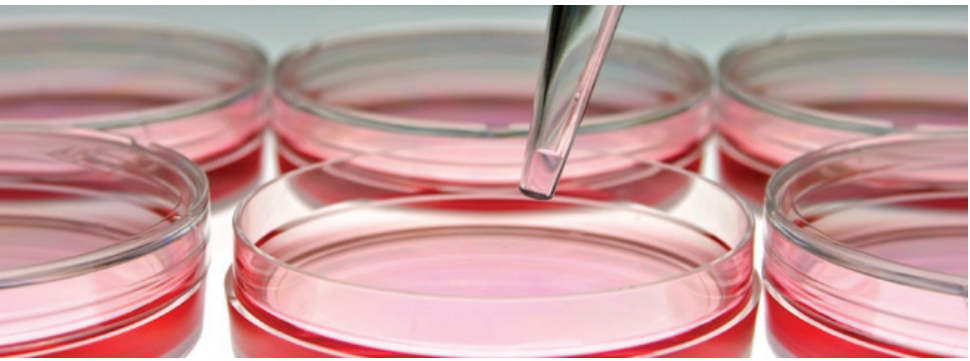
Our website has information updated regularly and is accessible on www.capitalpath.com.au

Other Services

Other services provided by Capital Pathology include:

- Biological Indicator monitoring
- Blood Bank and transfusion service
- Bone Marrow collections
- Drug Testing
- Corporate Medical Screening and Vaccinations
- ECG Recording and Interpretation by specialist Cardiologists
- Fine Needle Aspiration
- Frozen Section diagnosis
- Genetic Testing
- Helicobacter Pylori breath testing
- Holter Monitor service and reporting by specialist Cardiologists
- Infection Control Advice for hospitals, nursing homes and surgeries
- Mantoux Tests (Tuberculin Sensitivity Test)
- Pap Smear Reminder service, including follow-up letters for abnormal pap smears and statistical evaluation of pap smears
- Spirometry
- Water testing
- 24 Hour Blood Pressure Monitors.

For further information on any of these services, please contact the Client Services Department on 02 6285 9802.



Corporate Services

Capital Pathology is available to assist businesses assess known or suspected workplace risk factors that may contribute to ill-health and thus potential loss of productivity.

Our dedicated Corporate Services Team are able to perform a variety of tests and procedures both on and off site as part of the assessment of workers and workplaces.

The following tests are available:

- Corporate health testing
- Insurance pathology
- Insurance paramedicals
- HIV antibody testing
- Hepatitis screening
- Vaccination programmes including hepatitis A & B, tetanus and influenza
- Audiometry- hearing tests
- Spirometry- lung function
- Cardiologist reported ECG services including 24hr ambulatory
- Occupational biological monitoring
- Occupational allergy testing
- Heavy metals testing
- Trace metals toxicology
- Drug and alcohol testing (AS4308)
- Chemical exposure monitoring
- Pesticide exposure monitoring
- Water testing
- Therapuetic & drug trial monitoring
- Genetic studies
- Zoonoses – Q fever.

Please contact Collection Manager on 02 6285 9881
or email corporate.services@capitalpath.com.au for further information

A

Acetylcholine Receptor Antibody

Specimen: Serum – Gel

Reference Range: Not detected

This is a useful indicator in the diagnosis of Myasthenia gravis, with receptor antibodies detected in more than 90% of patients with generalised myasthenia.

Acid–Base Status

See *Bicarbonate*

Acid–Fast Bacilli (AFB)

See *Mantoux Test*
Tuberculosis

ACTH (Adrenocorticotrophic Hormone)

Specimen: Plasma – EDTA
Spin, separate and freeze plasma immediately.

Reference Range: Supplied with report

Note: special collection requirements apply. It is preferable to send patient to collection centre.

Actinomyces

An anaerobic gram–positive filamentous bacillus found as a normal commensal in the mouth and gut. The most commonly encountered species is *A.israelii*. Actinomyces species are susceptible to penicillin and doxycycline. Most isolates are resistant to metronidazole.

The most common form of infection is cervicofacial infection characteristically occurring in association with poor oral hygiene and tooth abscesses or decay. Thoracic and gastrointestinal infections also occur. All these deep–seated infections may declare themselves by developing draining sinuses.

Actinomyces species also grow in association with IUCDs (intrauterine contraceptive devices). They may be detected in a cervical smear or by culture of a removed IUCD in an asymptomatic woman. Although pelvic actinomycosis has been reported in association with IUCD use, the individual risk of infection with a colonised IUCD is small.

Activated Protein C Resistance (APCr)

Specimen: Plasma – Sodium Citrate x 2
If being performed as part of a thrombophilia screen collect sodium citrate x 6
Sodium Citrate tubes must be filled to capacity.

Reference Range: Supplied with report

Activated protein C resistance occurs in patients with the Factor V mutation known as Factor V Leiden (FV_{Q506}). This mutation is present in 5% of a normal Caucasian population. It is the commonest of the recognised thrombophilia markers. It is detected in 16–20% of consecutive patients presenting with thromboembolism but the frequency is higher in selected patient groups. Although it increases the thrombotic risk 5–10 fold, it is generally regarded as a relatively weak risk factor.

APCr is measured using an APTT ratio with and without activated protein C. The normal ratio is 2.4–4.0. Heterozygous positive patients with the Factor V mutation have ratios of 1.6 or less. Homozygosity, which is much less common, is associated with a higher thrombotic risk and a lower APCr.

See: *Factor V Leiden*

Acute Leukaemias

The diagnosis is usually obvious because of blast cells in the blood film, raised total white count, anaemia, thrombocytopenia and neutropenia, but one or more of these features can be absent at the time of diagnosis.

Acute Lymphoblastic Leukaemias (ALL)
The more common childhood leukaemia. Prognosis is relatively good in children with 80–90% remission rates. Remission rates are lower in adults.

Acute Myeloid Leukaemias (AML)
The more common leukaemia in adults. AML may arise *de novo* or through transformation of chronic myeloid leukaemia or myelodysplastic syndromes. Transformed AML responds poorly to treatment.

In *de novo* AML, first remission is achieved in 70–80% of patients with 30–40% long term survivors when treated with chemotherapy alone. With allogeneic bone marrow transplantation for patients aged less than 55 years there is a 50–60% five year survival.

The M3 subtype, acute promyelocytic leukaemia (APML), is treated with ATRA (a retinoic acid derivative) initially, to promote cell differentiation, followed by chemotherapy. This gives 85–90% remission rate with 80% 12–month disease free survival.

Acute Phase Proteins (reactants)

Specimen: Serum – Gel and EDTA

Reference Range: Supplied with report

Following inflammation or tissue destruction there is an increase in the so–called acute phase proteins with the development of a typical pattern on serum protein electrophoresis.

Analyte	Rise	Response times
CRP	Up to 1000x	6–10h
Alpha 1 antitrypsin	2–4x	24–48h
Haptoglobin	2–4x	24–48h
Fibrinogen	2–4x	24–48h
Ceruloplasmin	1.5–2x	48–72h
C3	1.5–2x	48–72h
C4	1.5–2x	48–72h

Adenovirus Antibodies

Specimen: Serum – Gel

Reference Range: Supplied with report

Adrenal Antibodies

Specimen: Serum – Gel

Reference Range: Not detected

Adrenal antibodies are present in 80% of autoimmune Addison's disease.

Adrenaline

See *Catecholamines*

Adrenal Function

Suspect?	Test
Addison's disease	Cortisol, ACTH, Adrenal cortex Abs, Synacthen stimulation test – due to risk of anaphylaxis, the Synacthen test is only to be performed under medical supervision with access to full resuscitation facilities
Congenital adrenal hyperplasia	17–OH Progesterone
Cushing's Disease	AM/PM cortisol, DHEAS, Prolonged dexamethasone suppression test, Short dexamethasone suppression test, Testosterone, U–Free cortisol
Phaechromocytoma	24-hour U–Catecholamines

Aeromonas

Aeromonads are widely distributed in stagnant or flowing fresh water and in soil.

Human infections include:

- Cellulitis or wounds contaminated with soil or water
- Acute diarrhoeal disease–worldwide, affecting any age
- Septicaemia
- Other infections, including the urinary tract.

Aeromonads are mostly resistant to the penicillins but are often susceptible to trimethoprim/ sulphamethoxazole and quinolones, e.g. ciprofloxacin. Nalidixic acid or nitrofurantoin may be used for urinary tract infection.

AFP (Alpha–fetoprotein)

Specimen: Serum – Gel

Reference Range: Age related reference ranges supplied with report

AFP has two uses:

- Tumour marker–large elevations (> 100) can be found in hepatomas and testicular tumours and are used for monitoring treatment and in diagnosis.
  Smaller non–diagnostic elevations are found in many forms of liver disease and malignancy.
- Screening for neural tube defects and Down's syndrome

See *Pre-Natal Testing*

Alaninine Transaminase or Amino Transferase (ALT)

Specimen: Serum – Gel

Reference Range: Adult 5–40 U/L
Age related reference ranges supplied with report.

See *Liver Function Test / Interpretation*

Albumin

Specimen: Serum – Gel

Reference Range: Adult 35–50 g/L

Decreases:

- Acute or chronic inflammatory illness
- Urinary protein loss
- Advanced liver disease
- Gastrointestinal loss
- Severe malnutrition.

Increases are usually due to dehydration.

Albumin Excretion Rate (AER)

Specimen: Urine – Timed overnight collection
24 hour collection (nil preservative).
Urine results expressed as Albumin Excretion Rate (AER).

Reference Range: < 20 ug/min normal
20–200 ug/min indicates microalbuminuria
> 200 ug/min indicates clinical proteinuria

Alcohol

Specimen: **Legal Whole blood** – EDTA or Flouride Oxalate or Heparinised blood with tamper proof seal.
Special collection requirements apply, please contact Doctors Service Centre for further information.

Non-Legal Whole blood – Flouride Oxalate

Reference Range: Supplied with report

Aldosterone

Specimen: Plasma – EDTA
Urine – 24 hour (nil preservative).

Reference Range: Supplied with report

Alkaline Phosphatase (ALP)

Specimen: Serum – Gel

Reference ranges vary with age and sex, with an increase during the teenage growing years and also with a gradual increase with advancing age. See individual test reports for specific reference ranges.

Reference range in adults 25 to 45 years

Males 35–110 U/L

Females 20–105 U/L

The bone isoenzyme comes from active osteoblasts, hence high levels during childhood, pubertal growth spurt, healing fractures.

In the liver, ALP is found in biliary canaliculi, increased production being stimulated by biliary obstruction and to a lesser extent other liver pathology.

Causes of hyperphosphatasaemia can be group under three main headings:

Liver disease
ALP of liver origin is usually identified by an accompanying rise in GGT which is also an "obstructive" enzyme. AST and ALT are usually also elevated to varying extents in liver disease.

For further discussion see Liver Function Test / Interpretation

Bone and other non–liver disease
- Paget's diseases of bone
- Malignant disease, secondary or primary
- Hyperparathyroidism
- Rickets/osteomalacia
- Chronic renal failure
- Healing fractures
- Thyrotoxicosis
- Rheumatoid arthritis
- Intestinal disease, e.g. Crohn's, ulcerative colitis.

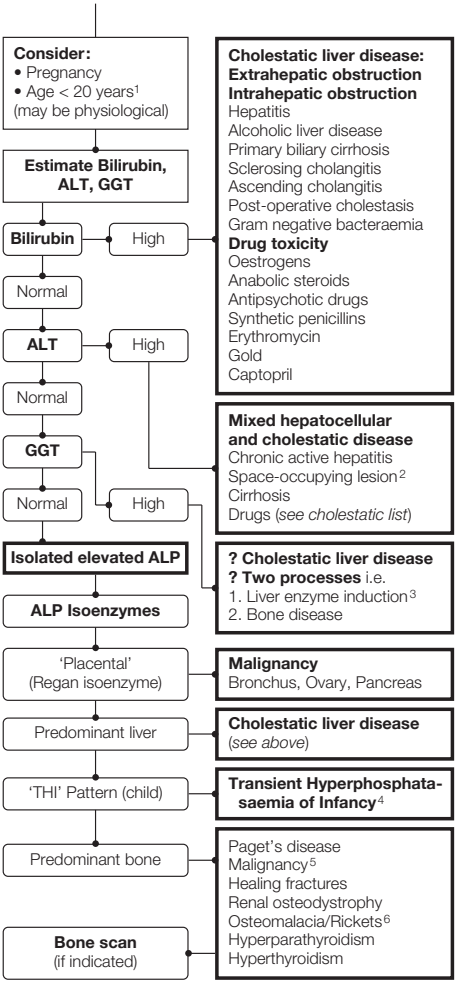
Benign isolated elevations of ALP
- Growth in childhood, particularly the pubertal growth spurt when levels can be up to 650 U/L.
- Third trimester of pregnancy, up to 400 U/L.
- Isolated elevations of ALP are common in the elderly and usually attributed to foci of subclinical Paget's disease provided there is no evidence of malignancy. Another possibility is osteomalacia due to vitamin D deficiency.
- In transient hyperphosphatasaemia of infancy and early childhood, ALP can go up to 3000 U/L for up to 3 months. Other liver enzymes remain unaffected. Aetiology remains obscure but viral infection is a possibility.
- Familial benign hyperphosphatasaemia shows as a raised ALP throughout life.

EVALUATION OF AN ISOLATED SERUM ALKALINE PHOSPHATASE (ALP) ELEVATION

Possible causes	Further investigations
Consider	
Pregnancy (increased placental isoenzyme)	
Age < 20 years (may be physiological)	
Due to the skewed distribution of ALP, levels of up to 400 IU/L are not uncommon in growth periods. Levels higher than this have also been reported.	
ALP Isoenzymes	
Predominant bone	Bone scan (if indicated)
Paget's disease	
Healing fractures	
Renal osteodystrophy	
Hyperparathyroidism	
Hyperthyroidism	
Malignancy (prostate, breast, kidney, myeloma, lymphoma, etc.)	
Osteomalacia/Rickets (due to Vitamin D deficiency or resistance)	
Placental (Regan isoenzyme)	
Malignancy (bronchus, ovary, pancreas)	
Predominant liver	
Cholestatic liver disease	
THI pattern (child)	
Transient Hyperphosphatasaemia of Infancy.	
Typical age < 5 years; very high ALP (> 700).	
May follow a viral illness.	
Benign and asymptomatic.	
High ALP persists for 8–12 weeks.	

SERUM ALKALINE PHOSPHATASE (ALP) ELEVATION

Suggested scheme for evaluation of a high serum ALP



1. Due to the skewed distribution of ALP, levels up to 400 IU/L are not uncommon during growth periods—higher levels have also been reported.
2. Malignancy (primary, secondary), abcess, cyst.
3. Alcohol, drugs (phenytoin, warfarin, benzodiazepines, tricyclics), obesity, diabetes mellitus, hypertriglyceridaemia.
4. Typically age < 5 years; very high ALP (> 700); may follow a viral illness. Benign and asymptomatic; high ALP persists for 8–12 weeks.
5. Prostate, breast, kidney, myeloma, lymphoma, etc.
6. Due to Vitamin D deficiency or resistance.

Alkaline Phosphatase Isoenzymes

Specimen: Serum – Gel

Assessment of other laboratory tests and the clinical picture will usually explain elevations of ALP without resorting to isoenzyme fractionation–which usually provides a clear–cut answer only when the answer is already obvious.

A useful generalisation is that, where both ALP and GGT are raised, liver is the source. When GGT is normal, the raised ALP is probably from bone.

Occasionally, however, the answer is not obvious. Heat stability is a method of isoenzyme analysis, the ALP being measured before and after 10 mins incubation at 56˚C.

%ALP remaining after incubation	Likely origin of raised ALP
< 20%	mainly bone
25–55%	mainly liver and/or intestine
> 90%	placenta

Allergy

Tests used in the diagnosis of IgE-mediated allergy
Once evidence of hyperreactivity is established, allergy testing plays a significant role in the diagnosis of allergy. It is important to note that an allergy test cannot have 100% specificity and sensitivity. Changing the cut-off level to improve one parameter will often worsen the other parameter. Hence cut-off levels are set to maximise both sensitivity and specificity.

Total IgE
The majority of patients with allergies have elevated total IgE levels. Higher serum IgE levels are seen in hyperreactivity diseases in which larger parts of skin and/or mucosa are involved. Patients with atopic dermatitis tend to have higher IgE levels than asthmatics who, in turn, have higher levels than patients with allergic rhinitis. In the absence of an increased IgE, further investigations for IgE allergy are likely to be unproductive. Therefore, serum IgE level can be used as a simple assessment of a patient's allergic profile.

Allergen-specific IgE
Specific IgE may be determined from a range of allergens using a non-isotopic variation of the radioallergosorbent test (RAST). RAST results are not affected by antihistamine or corticosteroid intake. The sensitivity and specificity of the RAST varies with the allergen and the allergy site. For inhalant allergies, the sensitivity of the RAST is 60–80% and the specificity is higher than that of skin tests, often as high as 90%. Please ring our Doctor's Service Centre (DSC) with any further enquiries.

Skin tests
Skin tests for a range of allergens are usually performed on the volar aspect of the forearm. Antihistamine intake may cause false negative results.

As with the RAST, skin test sensitivity and specificity varies with the allergen and allergy site. Generally, the sensitivity of skin tests is higher than that of the RAST. However, the specificity of the test is often lower (70–80%).

Alopecia

The common age and gender related variant does not warrant laboratory investigation but specific aetiologies to be considered include:

- Hypothyroidism
- Hyperthyroidism
- Drugs: cytotoxics, lithium
- Dermatological pathology (*see Skin Biopsy*)
- Any severe illness
- Zinc deficiency
- Warfarin hypersensitivity (rare)
- Polycystic ovary syndrome.

When associated with a severe but correctable illness, the alopecia is likely to be transient.

Alpha–1–antitrypsin

Specimen: Serum – Gel
Genotype Whole blood – EDTA.

Reference ranges: Supplied with report

AAT deficiency in its severe forms predisposes to early onset emphysema and juvenile cirrhosis and should be sought in these situations.

Sometimes the deficiency is detected as an absent or markedly reduced alpha–1 band on routine serum protein electrophoresis.

Genetically there is one normal gene, M, and two abnormals, S and Z

Genotype	Prevalence (%)	Reduction AAT level (%)
MM	88	0
MS	7	20
MZ	4	40
SS	1	40
SZ	0.1	70
ZZ	0.03	90

ZZ and, to a lesser extent, SZ are associated with emphysema and cirrhosis, and both smoking and alcohol should be strongly discouraged. The importance of MS, MZ and SS is uncertain but avoiding smoking is a wise precaution.

Tests for AAT deficiency:

Protein electrophoresis — a crude screen test but an absent or reduced alpha–1 band, or a band of slow mobility, is often the first clue to AAT deficiency.

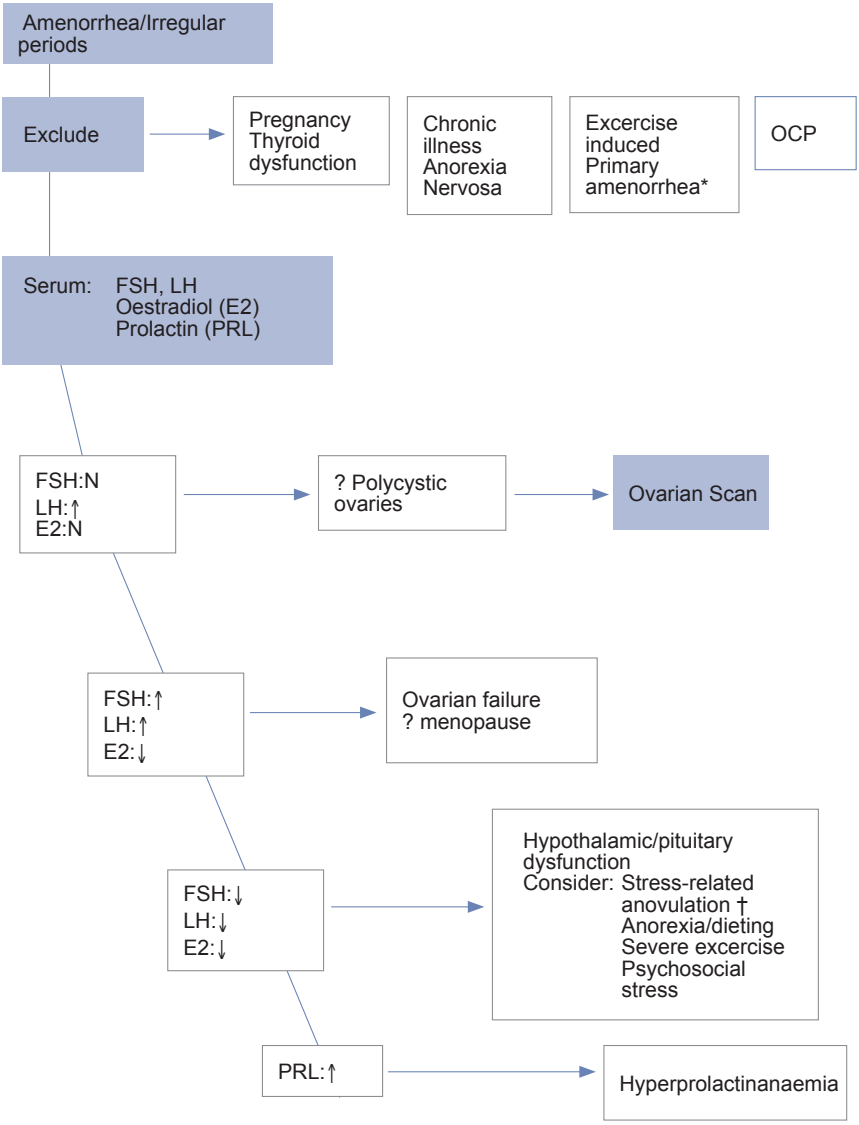
AAT quantitation–a result of < 1.0 g/L is suspicious and requires phenotyping which gives the best measures of risk.

Aluminium

Specimen:	Whole blood – Trace element tube Urine – 24-hour urine (nil preservative) or 50 mL random urine.
Reference Range:	Supplied with report

Amenorrhoea

EVALUATION ALGORITHM



* Chromosome abnormalities/ovarian failure more common in women who have never had a spontaneous period.
† FSH:LH ratio often > 1.0

Amiodarone

Specimen: Whole blood – Lithium heparin tube
Wait at least 3 months after commencing therapy before sample collection, unless toxicity is suspected.
Please provide the date that therapy started.
Collection just before next dose (at least 8 hours post dose).

Reference Range: Supplied with report

Amitriptyline

Specimen: Plasma – Lithium heparin
Trough level is taken before next dose (within one hour).

Reference Range: Supplied with report

Ammonia

Specimen: Plasma – EDTA
Spin and separate plasma within 20 minutes of collection and test to be performed within 2 hours of collection.

Reference Range: Supplied with report

Amphetamines (Drug Screen)

Specimen: Random urine

Reference Range: Not detected

See *Drug Screen*

Amylase

Serum

Specimen: Serum – Gel

Reference Range: 20–100 U/L

Amylase, found mainly in pancreas and salivary glands, is used in the differentiation of acute pancreatitis from other causes of the acute abdomen.

In acute pancreatitis, amylase typically rises above 400 U/L, returning to normal in about 4 days.

The enzyme pattern is inconsistent, however, and failure to detect an elevation does not preclude the diagnosis even when there is severe infarction.

Acute peritonitis, causing inflammation of the serosal surfaces of the pancreas and other organs, can elevate amylase but usually not above 400 U/L.

Other causes of hyperamylasaemia include mumps, diabetic ketoacidosis, biliary tract disease, renal insufficiency, shock, some drugs (particularly narcotics), hypoxia, pelvic infection, macroamylasaemia.

Chronic pancreatitis does not raise amylase except sometimes during acute exacerbations.

In macroamylasaemia, as in other macromolecular enzyme variants, a consistently elevated enzyme level is found in a well person. A definitive diagnosis can be made using the amylase/creatinine clearance ratio (ACCR).

$$\text{ACCR}(\%) = \frac{\text{urine amylase}}{\text{serum amylase}} \times \frac{\text{serum creatinine}}{\text{urine creatinine}} \times 100$$

In macroamylasaemia, the ACCR is < 2%. The usual reference range is 2–5%.

Urine

Specimen: Random or 24–hour urine (nil preservative)

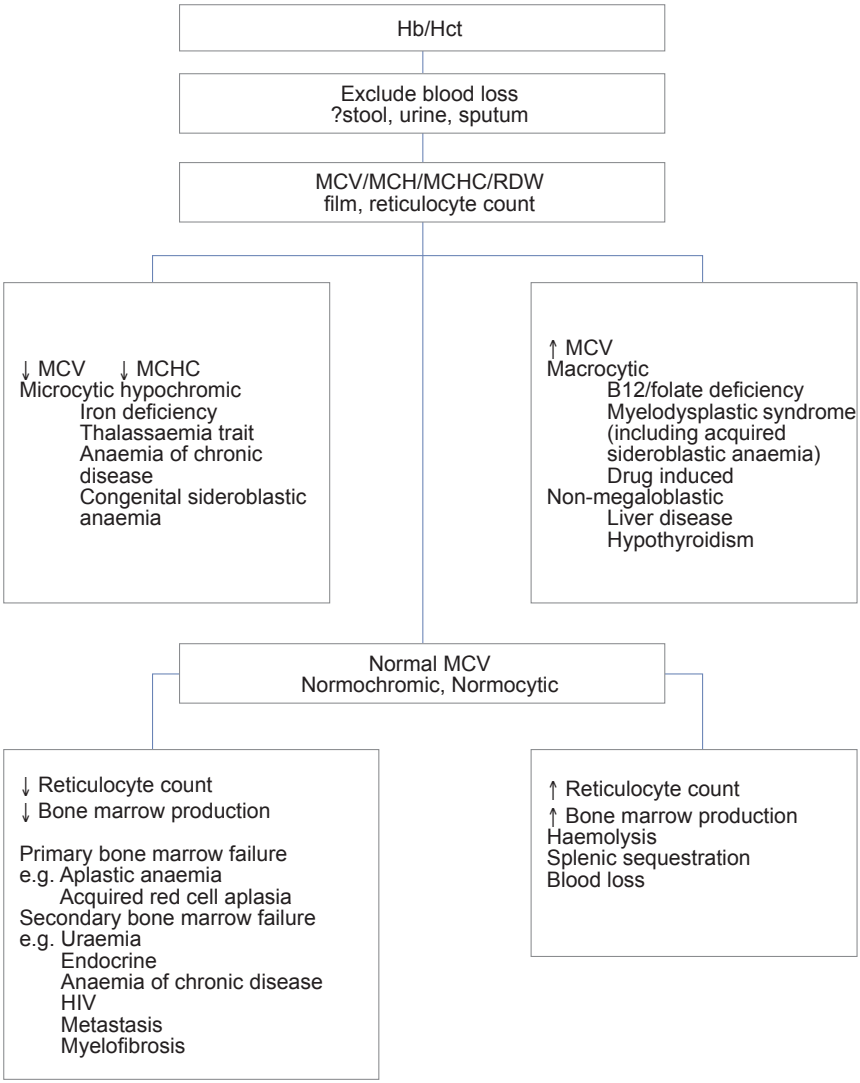
Reference Range: Supplied with report

Urine amylase is elevated in all conditions where serum amylase is raised except renal failure and macroamylasaemia. It is not often used except perhaps in the diagnosis of macroamylasaemia where the ACCR is calculated.

Anaemia

Anaemia Investigation

An Approach to the Investigation of a Decreased Haemoglobin



Anaerobic infections

Anaerobic organisms, which are widely distributed in soil and as part of the normal flora of mouth, gut and vagina, grow readily in deeply–placed abscesses in the abdomen, wounds, pleural cavities, brain, oropharynx and also in peritonitis or endometritis. Foul–smelling discharge indicates anaerobic infection.

Routine cultures do not grow anaerobes which require special media and an oxygen–reduced environment. Anaerobic cultures are set up only on specific request or when clinical details indicate the need for them. Swabs are not the ideal specimen, tissue obtained at debridement being the ideal. Aspirated pus or fluid is best collected and transported in a syringe which should not be refrigerated. IUCDs (intrauterine contraceptive devices) are routinely cultured for *Actinomyces*.

Common anaerobic species are *Bacteroides*, *Fusobacterium*, *Actinomyces* and *Clostridium*.

Most of the anaerobes we isolate will be susceptible to amoxycillin–clavulanate, clindamycin and metronidazole.

ANCA (Antineutrophil Cytoplasmic Antibodies)

Specimen: Serum – Gel

Reference Range: Not detected

These autoantibodies are directed against the cytoplasm of neutrophils.

Androstenedione

Specimen: Serum – Gel

Reference Range: Supplied with the report

Androstenedione is secreted by the adrenal cortex, testis and ovary and is a precursor of testosterone and oestrone. In premenopausal women, the adrenal cortex and ovaries contribute equally but after the menopause androstenedione is almost entirely of adrenal origin.

In hirsutism and polycystic ovary syndrome it can be elevated but testosterone is the preferred test.

Androstenedione can be markedly elevated in adrenal hyperplasia and is used in the investigation of virilism.

Angiotensin Converting Enzyme (ACE)

Specimen: Serum – Gel

Reference Range: Supplied with report

Although ACE levels can be used in the diagnosis of sarcoidosis (two thirds of patients with active disease have elevated ACE), elevations are also found in other chronic inflammatory disorders, giving the test poor specificity.

Anion Gap

Specimen: Serum – Gel

Reference Range: 10–22 mmol/L

The anion gap is a simple calculation which, if increased above 16 mmol/L, gives a crude (very crude) indication of metabolic acidosis as in salicylate or methanol poisoning, ketoacidosis, lactic acidosis, renal acidosis.

The calculation and its derivation are as follows:

In serum, total anions = total cations

$C1^- + HCO_3$ + acid anions (the anion gap) = Na^+ (the major cation)

Anion gap = $Na^+ (C1^- + HCO_3)$

For example a patient with salicylate poisoning might have

$Na^+ = 146$ $C1^- = 100$ $HCO_3 = 16$

The anion gap, at 30 mmol/L is increased, with acidic salicylate anions being the cause of the increased gap.

Ankylosing Spondylitis

A seronegative spondyloarthropathy causing low back pain and reduced spinal mobility due to sacroileitis and spondylitis. Typically it occurs in younger males with 95% positivity for HLA B27.

Anorexia Nervosa and Bulimia

Laboratory abnormalities include:

- Hypokalaemia is common in both conditions and can be profound (< 2.0) in bulimia due to the combination of vomiting and use of laxatives. In this situation there may be a metabolic alkalosis with raised serum bicarbonate.
- Anaemia (normocytic).
- Low FSH/LH accompanying the amenorrhoea which is almost invariable in anorexia.
- Hypoalbuminaemia, particularly when there is oedema.
- Hypocalcaemia is uncommon but is found occasionally.
- Cortisol may be elevated.

Other wasting disorders need to be excluded:

- glucose to exclude diabetes
- TSH/T4 to exclude hyperthyroidism
- Addison's disease causes wasting but is rare in young women under the age of 25 which is the common age group for eating disorders.

Antenatal Testing

Routine antenatal care involves looking for several diseases or maternal conditions that can affect either mother or baby.

The recommended tests can be considered at different stages throughout pregnancy.

Before 20 weeks:

- Full Blood Count (FBC)
- Blood group
- Red cell antibody screen
- Rubella antibodies
- Hepatitis B (HbsAg)
- Hepatitis C (HCV) antibody
- HIV antibody
- Syphilis serology
- Urine microscopy and culture
- Cervical cytology if indicated.

At 26–28 weeks:

- Red cell antibody screen
- Glucose load.

At 30–36 weeks:

- Full Blood Count
- Group B streptococcus screening may be indicated.

If high risk for infection:

- Urine or genital swab for Chlamydia trachomatis, gonococcus PCR
- Viral swab for herpes simplex PCR.

Additional tests if appropriate:

- 1st trimester screening
- 2nd trimester screening.

If routine antenatal screening returns a positive result, or if the patient is felt to be at risk for any other reason, further prenatal testing may be required.

It is preferable to specify the individual test required. However, if a request is received for ‘antenatal serology’ or ‘antenatal screen’ then the following tests will be performed: Full Blood Count (FBC), Blood group and Red cell antibody screen, Rubella antibodies, Hepatitis B surface antigen (HbsAg) and syphilis serology. Please note: Hepatitis C and HIV serology tests require patient counselling and consent, and need to be individually specified on the request form.

Please note labelling requirements.

1. All pre- transfusion and antenatal and perinatal specimens must be labelled **with surname, given name(s), date of birth, date and time of collection**
2. The **person collecting the blood** is to **sign** or **initial** the **tubes**
3. The **person collecting the blood** is to **sign** a **declaration** on the **request form** similar to the following:

   “I certify that I collected the accompanying sample from the above patient whose identity was confirmed by enquiry and/or examination of their name band and that I labelled the sample immediately following collection.”

See *Pre-Natal Testing*

Antibody Screen (Red Cell)

Specimen: Whole Blood EDTA

See *Coomb's test*

Anticonvulsants

Trough levels are usually preferred. Trough levels are taken just before the next dose. Peak levels may be collected 3–8 hours after the last dose. Timing of peak levels varies with the individual drug being measured. Carbamazepine peak level 3 hours post dose, Ethosuxamide peak level 2–4 hours post dose, Phenobarbitone peak level 1–3 hours post dose, Phenytoin peak level 4–7 hours post dose, Primidone peak level 1–3 hours post dose.

Drug	Type of Serum specimen	Half–life (hours)
Carbamazepine (Tegretol)	Gel	20
Clobazam (Frisium)	Plasma – Lithium heparin	
Clonazepam (Rivotril)	Plasma – Lithium heparin	40
Phenobarbitone	Gel	100
Phenytoin (Dilantin)	Gel	30
Primidone (Mysoline)	Plasma – Lithium heparin	
Valproic acid (Epilim)	Gel	13

Indications for monitoring anticonvulsants vary between the different drugs.

Suggested guidelines:

- Monitoring initial stabilisation or change of dose–phenytoin, carbamazepine, phenobarbitone
- Suspected toxicity–all drugs
- Suspected non–compliance–all drugs
- Failure to control seizures–all drugs
- Ongoing routine monitoring–phenytoin only, and even this may not be essential.

The therapeutic range is a guide only. Levels below the range may control seizures; levels above the range may not be toxic and may be necessary for control. When changing doses, retesting should be delayed a week or so till a new equilibrium develops.

Phenytoin, carbamazepine and phenobarbitone are potent inducers of hepatic enzymes thereby raising serum levels of GGT and ALP. The raised enzymes are regarded as an acceptable side–effect.

Primidone is measured as phenobarbitone which is the active metabolite.

Antidepressant Drugs, tricyclic

Therapeutic monitoring These drugs are not monitored routinely but estimations may be useful where there is lack of therapeutic response or uncertain compliance.

Drug	Type of Specimen
Anafranil	Plasma – Lithium heparin
Amitriptyline	Plasma – Lithium heparin
Desipramine	Plasma – Lithium heparin
Doxepin	Plasma – Lithium heparin
Imipramine	Plasma – Lithium heparin
Nortriptyline	Plasma – Lithium heparin

Reference ranges supplied with report.

Antidiuretic Hormone (ADH)

Specimen: Plasma – EDTA x 2

Reference Range: Supplied with report

Antimitochondrial Antibodies (AMA)

Specimen: Serum – Gel

Reference Range: Supplied with report

Antimitochondrial antibodies are seldom found in normal people. Their main use is in the diagnosis of primary biliary cirrhosis–90% of patients are positive. They are sometimes present in chronic active hepatitis and occasionally in other autoimmune disorders.

Anti Mullerian Hormone

Specimen: Serum – Gel

Reference Range: Supplied with report

Antinuclear Antibodies (ANA)

Specimen: Serum – Gel

Interpretation:	
< 1:40	Negative
1:40 or 1:80	Borderline
> 1:160	Consistent with autoimmune disease but not diagnostic

ANA in SLE (Systemic Lupus Erythematosus)
The main indication for ANA is as a screen test in suspected SLE where it will be positive in 95% of cases. A negative ANA makes SLE unlikely. A positive ANA requires follow–up by anti–DNA, which has a much higher specificity for SLE, and ENA to check specificity of the antibody.

ANA in healthy people
The exact incidence of positive ANAs in healthy populations depends on gender (commoner in women), age–group (rises with age), test methodology, ethnicity etc. and for this reason precise figures are not available.

A commonly quoted overall incidence is 5%.A recent study narrowed this to 5% in women aged 40–50. Another quotes 20% in women over the age of 60 rising to 30% over age 80.

The only certain statement is that positive ANAs are fairly common in healthy people and should never, as an isolated finding, be represented as possibly indicating SLE which requires additional laboratory and clinical features before the diagnosis can be made.

See *Systemic Lupus Erythematosus*

ANA in other autoimmune disorders

Incidence of positive ANA in other conditions:

- Rheumatoid arthritis 30%
- Sjogren's syndrome 60%
- Limited scleroderma (CREST) 50%
- Diffuse scleroderma (systemic sclerosis 80%
- Mixed connective tissue disease (MCTD) 80%
- Polymyositis 30%
- Autoimmune liver disease < 10%

Drugs causing positive ANA and sometimes a lupus–like illness

- Procainamide–responsible for most drug–related SLE
- Anticonvulsants
- Isoniazid
- Hydralazine
- Chlorpromazine
- Methyldopa.

The clinical manifestations of drug–related SLE are mild and subside within six weeks of drug withdrawal.

ANA patterns

ANA is detected by adding patient serum to a layer of cells on a slide, then adding a fluorescent detection system. If ANA is present it shows up as a fluorescent pattern illuminating various nuclear structures. Usually the pattern is not specific for a particular disorder but associations have been suggested:

- Nucleolar pattern
- Centromere
- Speckled
- Peripheral, diffuse (homogeneous)
- Diffuse scleroderma
- Limited scleroderma, CREST
- MCTD, Sjogren's, polymyositis, RA
- SLE.

Their presence indicates vasculitis including:

- Wegener's necrotising granulomatous vasculitis
- Systemic vasculitis
- Focal necrotising glomerulonephritis with vasculitis.

An initial ANCA screen test has 95% sensitivity for these conditions and is followed by the more specific antiproteinase 3 (PR3) and myeloperoxidase (MPO) tests.

Antiphospholipid Antibody Syndrome (APS)

A syndrome characterised by widespread, recurrent arterial and venous thromboses causing ischaemia in the affected organs. Clinical presentations include:

- Recurrent fetal loss, frequently mid–trimester with placental infarcts
- Recurrent arterial or venous thrombosis
- Stroke at a young age
- Other neurological and cardiological presentations.

APS is found frequently in SLE. When it occurs independently it is known as the primary antiphospholipid syndrome.

Markers used in the diagnosis of APS are autoantibodies directed against phospholipids, the main ones being:

- Cardiolipin antibodies are used as the initial test when APS is suspected
- Lupus anticoagulant
- Biological false positive RPR/VDRL.

The aetiology of these antibodies, which may be transient, is unknown but the presence of elevated titres of anticardiolipin antibodies and/or the lupus anticoagulants, six months after an episode of venous thromboembolism is a predictor for an increased risk of recurrence with probable benefit from long–term oral anticoagulation therapy.

See *Cardiolipin Antibodies*
Coagulation Studies
Lupus Anticoagulant/Inhibitor

Antithrombin III (ATIII, antithrombin)

Specimen: Plasma – Sodium Citrate x 2

Reference Ranges: Supplied with report

Antithrombin III is a non–vitamin K dependent physiological inhibitor of coagulation. It is essential for the anticoagulant effect of heparin.

Deficiency of ATIII, transmitted as an autosomal dominant, is associated with venous thrombosis. It is a strong but uncommon risk factor, which accounts for < 5% of patients with thrombophilia. The approximate incidence of the deficiency state is 1:5,000. Thrombosis occurs in 50% of patients before age 40. It is usually an indication for life long anticoagulant therapy after a thrombotic event.

ATIII can be spuriously reduced in the presence of heparin and after a thrombotic event, as well as in nephrotic syndrome, liver disease and pregnancy. Elevated levels are of no clinical significance.

Apolipoproteins

Specimen: Serum – Gel

Reference Ranges: Supplied with report

Cholesterol and triglyceride particles in serum are coated with apoproteins to render them soluble. Quantitation of the various apoproteins provides a more sophisticated basis for classifying the hyperlipoproteinaemias.

Apolipo–Protein–E Genotyping

Specimen: Whole blood – EDTA

Reference Ranges: Supplied with report

Apolipo–Protein “a”, A1, B

Specimen: Serum – Gel

Reference Ranges: Supplied with report

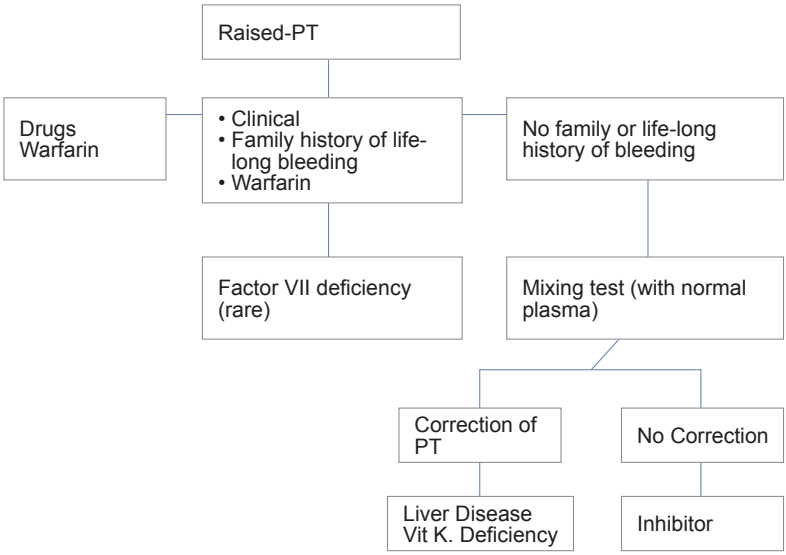
APTT (Activated Partial Thromboplastin Time)

Specimen: Plasma – Sodium Citrate
Sodium Citrate tube must be filled to capacity.

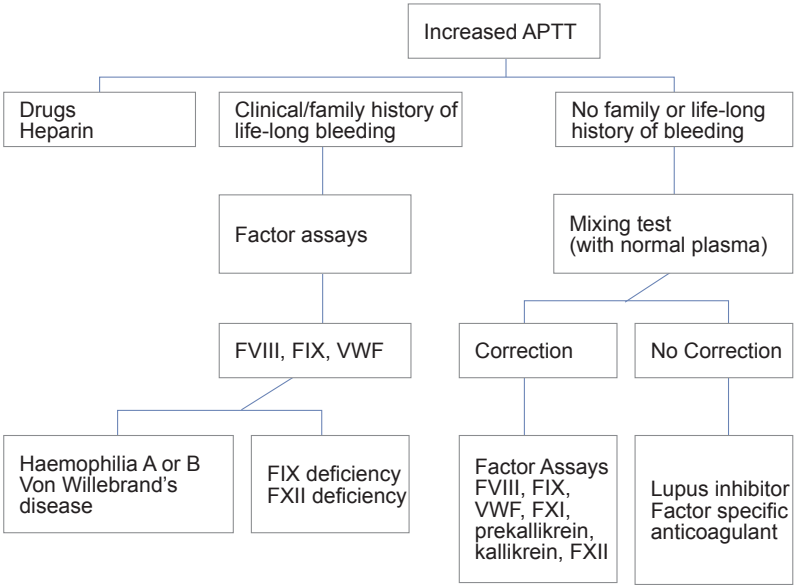
Reference Range: 24–37 secs

INVESTIGATION OF COAGULATION DISTURBANCES

Investigation of an increased Prothrombin Time (PT)



Investigation of Prolonged Activated Partial Thromboplastin Time (APTT)



The APTT is a basic test used to detect abnormalities of coagulation involving the intrinsic pathway. It is used to screen for deficiencies of plasma coagulation factors (except Factors VII and XIII) or for the presence of an acquired inhibitor. The mixing test is performed to distinguish between a deficiency and an inhibitor.

The APTT is usually sensitive to factor deficiencies < 40% normal. It is most commonly prolonged by deficiencies of contact factor FXII, FVIII (reduced in classical haemophilia and von Willebrand's disease) and Factor IX (haemophilia B or Christmas disease).

Marked prolongation of the APTT (> 80 secs) is usually due to haemophilia if the patient presents with bleeding, or due to Factor XII deficiency if the patient is asymptomatic.

The lupus anticoagulant, found in the antiphospholipid antibody syndrome, can prolong the APTT markedly. It is usually associated with an increased thrombotic tendency rather than bleeding.

Heparin therapy

The APTT is used to monitor standard or unfractionated heparin but not low molecular weight heparin (LMWH) therapy.

Reference Ranges: APTT ratio: 1.5–2.5

Arbovirus

(Ross River, Barmah Forest Dengue Serology)

Specimen: Serum – Gel

Reference Ranges: Supplied with report

Arsenic

Specimen: Occupational exposure – Random urine
Non-occupational exposure
• 24-hour urine (nil preservative) or random urine or
• Whole blood – EDTA.

Reference Range: Supplied with report

Urine is the preferred specimen for toxicity and occupational monitoring. This method measures total arsenic. Avoid seafood 5 days prior to collection to exclude non–toxic organo arsenic compounds.

Arterial blood gases

Specimen: 3 mL arterial blood in heparinised syringe transported on ice
Sample should be analysed within 15 minutes (Hospital in-patients).

Includes: pO2, pCO2, bicarbonate, base excess, pH and oxygen saturation

Reference Range: Supplied with report

Arthritis

For initial investigation (screen), the following tests are recommended:

Antinuclear antibodies (ANA), C- Reactive Protein (CRP), ESR and Rheumatoid Factor (RF).

Some of the diagnoses to be considered include:

Septic arthritis – immediate joint aspiration (*see Synovial aspirate*) will give definitive diagnosis of septic arthritis and differentiate it from gout.

Gout and pseudogout

Rheumatoid arthritis, SLE and other connective tissue diseases

Reactive Arthritis

Reactive arthritis is an aseptic arthritis that develops after an infection elsewhere in the body. Reiter's disease, which includes arthritis, conjunctivitis, and urethritis, is an example of a reactive arthritis. 60–90% of affected patients are positive for HLA-B27 antigen.

- Post–enteric
  - Yersinia
  - Chlamydia
  - Campylobacter
  - Salmonella
  - Brucella
- Post–venereal
  - Chlamydia
  - Ureaplasma

Specific Viral Arthritis

- Arbovirus
  - Ross River
  - Barmah Forest

Arthritis: Infectious

Serology: RRV, BFV, Parvovirus, Rubella, Group A Streptococcus, Yersinia, Campylobacter, Shigella

Synovial fluid: MCS including cell count and crystal examination

Arthritis: Non-infectious

ANA, Anti-DNA, Anti-ENA, CRP, HLA B27, RF, CCP Ab

ASOT (Antistreptolysin O titre)

Specimen: Serum – Gel

ASOT = Antistreptolysin O titre

ADNAse = AntiDNAse

These tests are used when searching for evidence of recent streptococcal infection in suspected post–streptococcal glomerulonephritis (PSGN) or rheumatic fever (RF).

Serial tests should be done, looking for rising titres. A high level is more likely to be significant and sustained levels may indicate persisting infection. PSGN or RF can easily be over–diagnosed when an isolated, equivocal titre is used as evidence.

See *Rheumatic Fever*

Aspartate Aminotransferase (AST)

Specimen: Serum – Gel

Reference Range: Males 3–40 U/L
Females 10–35 U/L

See *Liver function test / interpretation*

Aspergillus

A fungus common in soil and decaying material and the cause of an invasive disease in immunocompromised patients. Diagnosis is by demonstration of hyphae in tissue biopsy, sputum or culture.

We often recover *A. fumigatus* and occasionally other Aspergillus species from ear swabs taken from patients with otitis externa. There is no easily applied antifungal agent for this situation. Management requires thorough debridement and keeping the ear dry.

Allergic pulmonary aspergillosis is characterised by asthma and eosinophilia. The total IgE is often above 1000IU/L. Aspergillus precipitins should be tested for and skin prick testing arranged.

Aspirin

Specimen: Plasma – Lithium heparin

Reference Ranges: Supplied with report

Ativan (Lorazepam)

Specimen: Plasma – Lithium heparin

Reference Range: Supplied with report

Autoantibodies

Specimen: Serum – Gel

The list of autoantibodies used in the diagnosis of autoimmune disorders continues to lengthen.

Specificities for particular diseases are seldom absolute and low titre autoantibodies can be found in apparently disease–free people. Observation over months or years may show regression, no change or progression to clinical disease.

Each autoantibody is described in more detail under its own alphabetic entry.

Please specify specific tests required according to the patients clinical presentation. If an 'Autoantibody profile' is requested, the following tests will be performed: Antimitochondrial antibodies (AMA), Smooth muscle antibodies (SMA), Anti Nuclear Antibodies (ANA), Gastric parietal cell antibodies (GPC) and Thyroid antibodies (TA).

Tissue	Disease	Autoantibody
Adrenal cortex	Addison's disease	Adrenal
Islet cells	Type I diabetes	GAD 65 IA2 Islet cell (ICA) Insulin (IAA)
Thyroid	Hypothyroidism Grave's disease	Microsomal Thyroglobulin TSH receptor
Gastric	Pernicious anaemia	Intrinsic factor Parietal cell (GPC)
Small intestine	Coeliac disease	Gliadin (AGA) Tissue Transglutaminase (TTG) Reticulin
Tissue	**Disease**	**Autoantibody**
Liver	Autoimmune hepatitis 1° biliary cirrhosis	Smooth muscle (SMA) Liver kidney microsomal (LKM) Mitochondrial (AMA) MUSK
Neuromuscular	Myasthenia gravis	Acetylcholine receptor Skeletal muscle
Kidney	Goodpasture's syndrome Crescentric glomerulonephritis	Glomerular basement membrane
Testis	Seminal infertility	Sperm
Skin	Pemphigus	Pemphigus

Tissue	Disease	Autoantibody
Haematological	ITP Post–transfusion Antiphospholipid (APS)	Platelet Red cell Lymphocyte Cardiolipin Beta 2 glycoprotein
Connective tissue disease	Rheumatoid arthritis SLE Polydermatomyositis Scleroderma Mixed CTD Sjogren'syndrome Vasculitis	Rheumatoid factor (RF) ANA ds DNA ENA CCP Salivary Duct Neutrophil cytoplasm (ANCA)

Avian Precipitins (Bird Fanciers' Disease/Lung)

Specimen: Serum – Gel

Reference Range: Supplied with report

B

Bacterial Swabs

Bacterial specimens are collected for identification of microorganisms and for antibiotic susceptibility testing. The quality of the specimen is all–important and will determine whether a pathogen is successfully isolated or whether a false negative culture occurs.

Human pathogenic bacteria like a moist environment, close to body temperature (37ºC) with no exposure to toxic chemicals. Conversely they are killed by:

- Drying
- High Temperature
- Low Temperature
- Sunlight
- Antiseptics
- Antibiotics.

To provide an optimum environment for swabs, we place them in transport medium which should be stored at room temperature rather than at 4ºC in the fridge. Gonococci, for example, survive 24 hours in transport medium at room temperature but only 2 hours at 4ºC.

An exception is urine swabs which should be stored in the fridge to prevent overgrowth of contaminants.

Details about collecting swabs from specific sites will be found under their own alphabetical headings.

- Cervical
- Ear
- Eye
- Skin
- Sputum culture
- Throat
- Urethral
- Vaginal.

Bacteroides

An important group of anaerobic gram–negative bacilli. *B. fragilis*, which is part of the normal flora or the bowel, has the ability to form abscesses when released into the peritoneal cavity. Specimens from intra–abdominal sites must always be cultured anaerobically as well as aerobically. Septicaemia and puerperal or post–abortion sepsis often include *B. fragilis* as a major pathogen.

Bacteroides fragilis produces beta–lactamase which inactivates penicillin and amoxycillin. Amoxycillian–clavulanate, metronidazole and clindamycin are the most effective agents.

Bacteroides other than *B. fragilis* are grouped as Bacteroides species. Some may be sensitive to penicillin. Antibiotic susceptibility testing should be discussed with a clinical microbiologist.

Some species in the genus have recently been reclassified as Prevotella and Porphyromonas species.

Barmah Forest Antibodies

Specimen: Serum – Gel

Reference Range: Supplied with report

Basophils

Specimen: Whole blood – EDTA

Reference Range: Adults < 0.3 x 10^9/L

The basophil count is part of a routine blood count.

Basophilia is rare as an isolated abnormality. Increases occur in chronic myeloid leukaemia and other myeloproliferative disorders including polycythaemia rubra vera.

See *Blood Count*

Bence Jones Protein (BJP)

Specimen: Random urine

Reference Range: Not present in normal urine

In 1848, Bence Jones described a protein in the urine of myeloma patients that precipitated at 50ºC and redissolved on boiling. It consists of free light chain fragments (kappa or lambda) derived from IgG and IgA paraproteins found in serum in myeloma and occasionally from an IgM lymphoma–associated paraprotein. Because free light chains are small molecules they pass through the glomerular filter whereas the larger paraprotein molecules are usually retained in serum.

Presence of BJP in urine is an indication that a paraprotein is malignant rather than benign. In Bence Jones (light chain) myeloma there is no paraprotein in serum but a heavy band of free light chains in urine which means that testing for myeloma must include urine as well as serum electrophoresis.

BJP is detected in two ways:

- As an abnormal band found on EPP (electrophoresis) of concentrated urine
- As a kappa or lambda arc on IMF (immunofixation) of concentrated urine

Benzodiazepines (Drug Screen)

Specimen: Random urine

Reference Range: Not detected

See *Drug Screen*

Beta(β) HCG

Specimen:	Quantitative HCG	Serum – Gel
	Qualitative pregnancy test	Serum – Gel
	or	Random urine
Reference Range:	Non–pregnant female or male serum : < 5 IU/L	

βHCG in normal pregnancy

A single measurement of HCG answers the question whether a pregnancy is present or not.

Adding a second HCG or a series and plotting them on the HCG graph gives valuable information on the health of the embryo.

In a normal pregnancy, the HCG doubles every 1–2 days between weeks 3–15 and the trajectory rises parallel to the graph's percentile lines with remarkably little deviation.

A plot which is rising parallel to the percentile lines but outside the 5^{th}–95^{th} range indicates a healthy pregnancy with incorrect dates.

A graph rising more slowly, or flattening, usually indicates the onset of spontaneous abortion or an ectopic pregnancy and is an indication for proceeding with vaginal ultrasound looking for an intrauterine gestational sac.

βHCG after spontaneous or induced abortion

After fetal loss or death, the HCG falls over a period of 1–5 weeks depending on its initial height and the completeness of elimination of placental tissue. The starting HCG depends on gestational age and health of the conceptus.

βHCG in hydatidiform mole and choriocarcinoma

The abnormal trophoblastic tissue produces large amounts of HCG whose levels are usually either high normal or clearly above the 95th percentile, sometimes as high as 1,000,000 IU/L. Ultrasound will separate a hydatidiform mole from a multiple pregnancy which can also have an HCG level above the 95th percentile.

Absolute HCG levels are more method–dependent in trophoblastic disease and tumours than in normal pregnancy where levels should be repeatable between laboratories with different methods. In tumours, there are varying amounts of free beta chains and beta fragments, and different methods measure these to a varying extent. For this reason, when monitoring a tumour, specimens should be sent to the same laboratory.

βHCG in other tumours

Ovarian and testicular germ cell tumours commonly produce HCG and a variety of malignancies from other sites can occasionally secrete HCG. Levels are used for monitoring treatment.

Beta–2–microglobulin

Specimen: Serum – Gel
Urine – random collection.

Reference range: Supplied with report

Beta–2–microglobulin is elevated in autoimmune conditions where there is lymphocyte activation or destruction.

- Multiple myeloma – used for monitoring and prognosis
- HIV–levels above 4.0 g/L are more likely to progress to AIDS
- Autoimmune disease
- Viral infections
- Lymphoid neoplasm
- Renal impairment.

Bicarbonate

Specimen: Serum – Gel

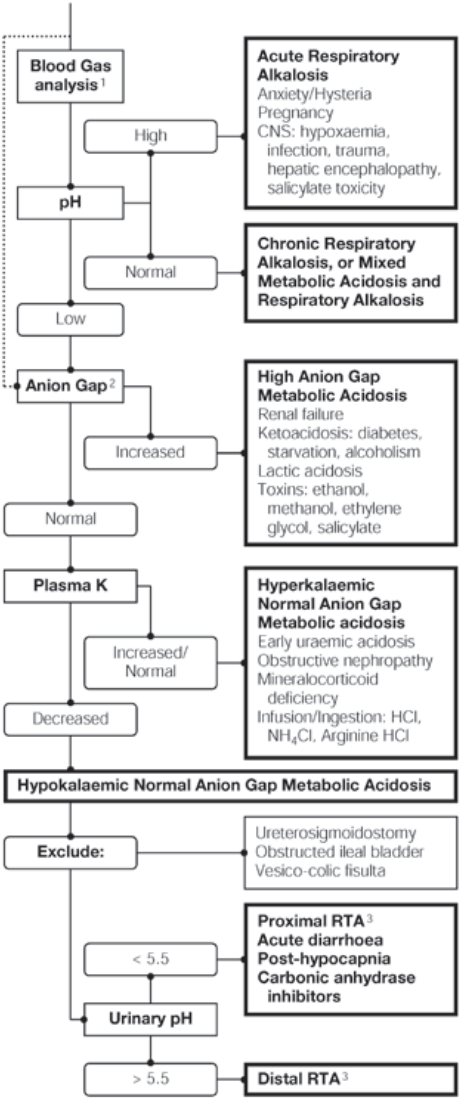
Reference range: Adults 20–32 mmol/L

Also known as total CO_2, this is a crude acid–base measure. In metabolic acidosis, bicarbonate is reduced, e.g. in diabetic ketoacidosis. In metabolic alkalosis, it is increased – as in pyloric stenosis or the compensated respiratory acidosis of chronic obstructive airways disease.

See flow charts on next page

Causes of low plasma HCO_3

Flowchart for assessment of low plasma bicarbonate



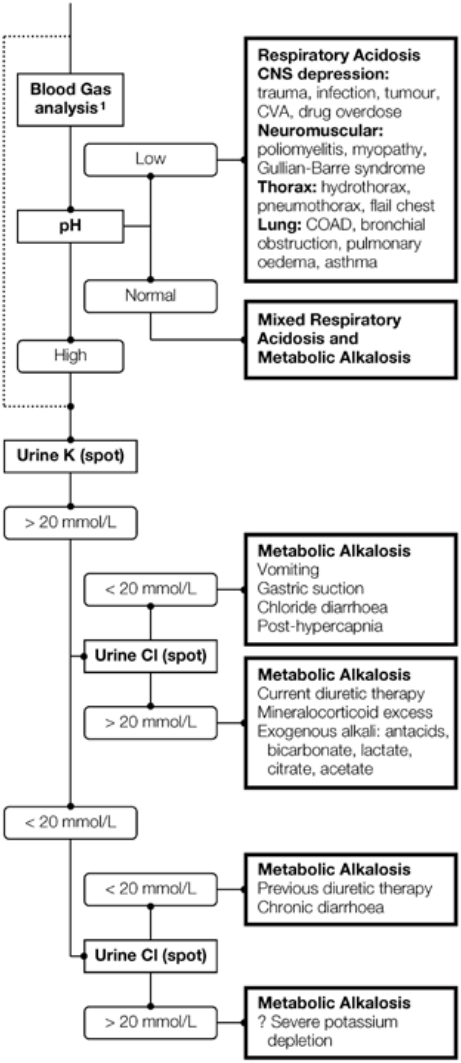
1 If blood gases are unavailable exclude Respiratory Alkalosis on clinical grounds

2 Anion Gap = $(Na + K) - (Cl + HCO_3)$
Reference range: 8–16 mmol/L

3 RTA: Renal Tubular Acidosis

Causes of high plasma HCO_3

Flowchart for assessment of high plasma bicarbonate



1 If blood gases are unavailable exclude Respiratory Acidosis on clinical grounds and perform spot urinary K and Cl estimations.

Bilirubin

Specimen:	Total: Serum – Gel (protect from light) Conjugated and unconjugated: Serum – clot or gel (protect from light).
Reference Range:	Total: Adult 4–20 µmol/L
See	*Gilbert's Syndrome* *Jaundice* *Haemolysis*

Hyperbilirubinaemia in infants

I. Benign Jaundice

- **Physiological jaundice**

This is due to breakdown of fetal red cells, "immaturity" of the liver's glucuronyl transferase system and increased bilirubin resorption from bowel meconium in infants as maturation is completed during the first days and weeks of life.

The bilirubin reaches a peak in 5 days (sometimes as high as 300 umol/L) and in bottle–fed infants disappears in 2 weeks.

- **Jaundice of breast–feeding**

This condition, an extension of and inseparable from physiological jaundice, is due to the presence of maternal hormone in breast milk inhibiting the maturation of glucuronyl transferase.

Bilirubins can stay high for a long time. In about 2–5%, levels reach 250–350 umol/L by the 3rd week, remain high for 3–4 weeks, falling to normal over another 1–3 months.

Despite being yellow, the infant thrives, feeds and grows. Changing to bottle–feeding results in a prompt and sustained fall in bilirubin levels and cessation of breast–feeding for 24–48 hours, with bilirubins before and after, can be used as a diagnostic test whilst maintaining lactation by manual expression.

II. Pathological jaundice

- **Haemolytic disease of the newborn**

Rhesus incompatibility

The most severe forms of this previously dreaded disease were due to Rhesus incompatibility between mother, (Rh –ve and with anti–Rh antibodies) and baby (Rh +ve). The Rhesus system was first described in 1940. Before that time, and until modern methods of prevention and treatment were developed, these babies were born dead or anaemic with jaundice at birth increasing rapidly in the first days of life to cause long–term brain damage in the form of kernicterus. By the late 20th century, kernicterus had almost entirely disappeared but the fear of it lingers on.

Haemolytic disease is caused by anti–Rh antibodies in an Rh –ve mother who has been sensitised during a previous pregnancy with an Rh +ve baby whose red cells have spilled into the maternal circulation at the time of placental separation. Over the past 40 years this sensitisation process has been prevented by injecting the mother at delivery with enough anti–Rh immune globulin to render the fetal cells non–antigenic.

Sensitisation of the mother by an incompatible blood transfusion, unavoidable before 1910, is now an exceptionally rare event.

Despite the success of preventive measures, haemolytic disease is still occasionally found.

The at–risk mother is Rh –ve and will have red cell autoantibodies when screened antenatally, usually anti–D, sometimes anti–c or anti–E, least commonly anti–C or anti–e. For the fetus to be at risk, the father must possess antigen to which antibody is targeted.

Haemolytic disease in the at–risk infant is diagnosed by examining cord blood at birth, looking for

- Rh +ve infant
- Positive Coomb's test
- Hyperbilirubinaemia.

ABO incompatibility and other blood groups
This is more common but rarely severe enough to require exchange transfusion. The at–risk situation is found in 20% of pregnancies. Usually the mother is Group O and the infant Group A or B. Prior sensitisation is not required and haemolysis can occur in the first pregnancy.

Maternal antibody tests are of no use and often the direct Coomb's test on cord blood is negative.

Other blood groups include Kell (K), Duffy (Fy^a, Fy^b) and Kidd (Jk^a, Jk^b). For these incompatibilities, maternal antibodies will be present and the direct Coomb's on cord blood positive.

- **Neonatal bacterial infections**

Septicaemia is life–threatening and the infant is obviously unwell. Urinary tract infections are more easily overlooked, the infant being less obviously unwell to begin with.

- **Hypothyroidism, cystic fibrosis and galactosaemia**

Tested on all neonates by way of the Guthrie Card blood spots

- **G–6–PD deficiency**

The vitamin K given routinely to neonates elevates bilirubin in G–6–PD hetero–and homozygotes. On rare occasions homozygous infants (usually males of SE Asian or Mediterranean origin) develop a severe haemolytic process in the neonatal period, especially if exposed to certain drugs.

- **Biliary atresia and neonatal hepatitis**

The infant is often clinically well during the first few weeks but thereafter there is an increasing obstructive jaundice shown by rising total bilirubin, a raised level of conjugated bilirubin, pale faeces and dark urine.

An infant still jaundiced at 3 weeks should have its conjugated bilirubin level measured–over 20 umol/L of conjugated bilirubin is abnormal and an indication for further investigation.

When and how often should bilirubins be measured?
As always, clinical judgement and experience are more important than rigid guidelines.

A healthy term baby, growing and feeding well, without visible jaundice, does not need to have its bilirubin measured.

Premature babies need to be watched more carefully than full–term babies, particularly during the first 3–5 days.

Jaundice in the first 24 hours of life is not benign. If the jaundiced baby is unwell, the underlying cause must be identified urgently.

Biopsy Specimens

See *Histopathology, skin biopsy*

Blastocystis hominis

A protozoan parasite whose status as a cause of diarrhoea is still unresolved. It can be observed in the stool of asymptomatic people. Metronidazole, 400–800mg 8-hourly for 10 days, may be effective treatment.

Blood Banking

Autologous transfusion

Autologous transfusion involves the donation and storage of blood from patients who will be undergoing elective surgery. If required, these patients can be transfused with their own blood at surgery. The procedure has become more widespread as concern over transfusion–transmitted diseases has grown.

1–4 units can be collected pre–operatively at Red Cross House at Woden.

All of these units undergo the same routine screening procedures that are carried out on all blood collected at the Red Cross. The units are then sent to the laboratory for storage and crossmatching prior to surgery.

The patient must return to a Collection Centre for a crossmatch specimen to be collected within five days of the proposed surgery. All of the units that may be transfused must be crossmatched against this specimen. When required, the crossmatched units are dispatched to the appropriate hospital or clinic.

Crossmatching requests

Specimen: Whole Blood (EDTA) x 2. Blood should be taken up to 10 days prior to surgery. If patient is pregnant or has been pregnant in the last three months or has been transfused in the last 3 months, blood should be taken no more than 72 hours prior to surgery.

Specimens must be carefully labelled. The following details must be clearly written on the tube; patients family name, given name, medical record number and/or date of birth, date and time of collection and signature of the collector.

Specimens are to be accompanied by a transfusion sheet which must contain the following details : patients family name, given name, medical record number and/or date of birth, date and time of collection.

There must be two signatures in the declaration section of the transfusion sheet:

1. The person who collected the blood and;
2. The patient, or if the patient is unable to sign, a witness who can attest to the patient's identity.

The declaration should read as follows:

"I certify that I collected the accompanying sample from the above patient whose identity was confirmed by enquiry and/or examination of their name band and that I labelled the sample immediately following collection."

Please provide any relevant clinical information on the transfusion sheet including previous transfusion, obstetric history and any known antibodies.

The request form and specimen tube will be checked in the laboratory to ensure validity. Printed sticky labels are acceptable for specimen identification when there is a signature from the collector on the label.

Emergency transfusion

A pre–transfusion blood sample must be obtained, labelled with the patient's name, Medical Record Number, date of birth and date of collection and sent to the laboratory as quickly as possible. Please contact the Main Laboratory on 02 6285 9811 when blood is required urgently. A correctly filled out transfusion form is also required.

In an emergency there are units of O negative packed red cells in the theatre fridge's for immediate use. The laboratory must be notified immediately if these units are used.

Group and hold (G&H) blood for transfusion

Specimen: Whole blood (EDTA) x 1

It may be adequate to perform a blood group and antibody screen only ("Group and Hold"). Blood can then be crossmatched and released promptly if required.

When a Group and Hold has been performed, red cell units can be issued within 30 minutes. Without prior group and hold, a full procedure of blood group, antibody screen and crossmatch will take approximately 50–60 minutes.
If antibodies are present in the antibody screen, fully compatible units may take even longer to locate.

Routine crossmatching

Specimen: Whole blood (EDTA) x 2

Blood should be taken up to 10 days prior to surgery. If patient is pregnant or has been transfused in the last 3 months, blood should be taken no more than 72 hours prior to surgery.

Specific blood products

Antihaemophilia Factor (AHF)

AHF is indicated for the management of bleeding episodes and surgical and dental procedures in patients with haemophilia A (Factor VIII C deficiency) and moderate to severe von Willebrand's Disease (vWD).

Cryoprecipitate

Cryoprecipitate is the cold–insoluble portion of plasma remaining after FFP has been thawed between 1°C and 6°C. It contains the following factors– fibrinogen, Factor VIII C, von Willebrand factor, Factor IX, Factor XI, Factor XII and Fibronectin. Cryoprecipitate is available through the Red Cross Blood Bank, in consultation with the laboratory.

Fresh Frozen Plasma (FFP)

FFP contains all coagulation factors plus complement and is stored at -25°C in the laboratory. FFP is indicated for use in control of bleeding in patients with multiple coagulation defects such as liver disease, dilution coagulopathy and in some instances of warfarin overdose or if warfarin therapy needs to be reversed.

Platelets

Standard platelet concentrates are available and should be ordered in advance. For routine purposes at least 24 hours notice is desirable. In an emergency situation platelets can be provided on request.

The following MSBOS is taken from ANZSBT Guidelines for pretransfusion laboratory practice 5th edition March 2007 and is intended as a guide only.

Recommended Maximum Surgical Blood Order Schedule (MBOS)

General Surgery

Abdomino-perineal Resection	2
Amputation	
• Below knee	G&S
• Above knee	G&S
Anterior Resection	2
Appendicectomy	Nil
Apronectomy/Lipectomy	G&S
Bowel Resection	2
Breast Surgery	
• Lumpectomy	G&S
• Simple Mastectomy	G&S
• Radical Mastectomy	G&S
Burns Debridement	IA*
(*IA = Individual Assessment)	
Cholecystectomy	G&S
Colectomy (formation or closure)	G&S
Gastrectomy	2
Gastric Stapling	G&S
Haemorrhoidectomy	Nil
Hiatus Hernia Repair	
• Transthoracic	2
• Abdominal	G&S
Incisional Hernia Repair	Nil
Laparotomy	G&S
Lipectomy	G&S
Lumbar Sympathectomy	G&S
Pancreatectomy	2
Parotidectomy	G&S
Splenectomy	2
Thyroidectomy	G&S
Vagotomy and Drainage	G&S
Varicose Veins Stripping	Nil
Ethmoidectomy	Nil
Mastectomy	G&S
Mastoidectomy	G&S
Rhinoplasty	G&S
Tonsillectomy	G&S
Trachoestomy	G&S

Recommended Maximum Surgical Blood Order Schedule (MBOS)

Gynaecological Surgery	
Caesarean	G&S
Colposuspension	G&S
Cone Biopsy	Nil
D & C	Nil
Ectopic	G&S
Hysterectomy	G&S
Laparoscopy	Nil
Myomectomy	G&S
Ovariancystectomy	G&S
Termination of Pregnancy	G&S
Tubil Ligation	Nil
Vaginal Repair	G&S
Vulvectomy	G&S
Orthopaedic Surgery	
Arthroscopy	Nil
Arthrotomy	Nil
Femoral Nail Removal	Nil
Fractures	
• Femur	2
Harrington's Rods	4
Hip Replacement	3
Knee Replacement	G&S
Laminectomy	G&S
Menisectomy	Nil
Putti-Platt	G&S
Spinal Fusion	2
Synovectomy (Knee)	G&S
Thoracic Surgery	
Lobectomy	2
Pleurectomy	2
Pneumonectomy	4
Thymectomy	2
Urological Surgery	
Cystoscopy/otomy	Nil
Cystectomy	4
Nephrectomy	G&S
Nephrolithotomy	G&S
Prostatectomy	
• Open	2
• Transurethral resection; TURP	G&S
Pyelolithotomy	G&S
Transurethral Resection of Prostate	G&S
Ureterolithotomy	G&S
Vascular Surgery	
Aortic Aneurysm – Elective	4
Aorto-femoral Bypass Graft	4
Aorto-iliac Bypass Graft	4
Av Shunt	Nil
Carotid Endarterectomy	G&S
Femoro-popliteal Bypass Graft	2
Ilio-femoral bypass Graft	4
Sympathectomy Lumbar	G&S

Blood Count (FBC)

Specimen: Whole blood – EDTA: Adult tube = 4.5 mL blood
: Paediatric tube = 0.5 mL blood

Reference ranges vary widely with age and sex, and detailed reference ranges are supplied with reports.

Includes Haemoglobin, Haematocrit, Red cell count, Mean cell volume, Mean cell haemoglobin, Mean cell haemoglobin concentration, White cell count and differential, platelet count and film examination where indicated.

Blood Cultures

Specimen : Whole blood taken by aseptic technique

Generally, a blood culture collection requires two culture bottles (aerobic and anaerobic).

If blood culture for Mycobacteria is required please contact the Microbiology Department on 02 6285 9846 for special collection details.

Careful aseptic technique is required. The bottles must be labelled (including time of collection) and if there is any delay in delivery, they should be kept at room temperature until pick–up.

Collect 10–20 mL into two culture bottles. The volume of blood collected is directly proportional to the success of isolating an organism. In cases of suspected endocarditis, three sets over 24 hours are recommend to optimise isolation of the organism. If antibiotics need to be started rapidly, collect only one set of blood cultures prior to starting therapy.

Blood cultures are examined continuously and incubated routinely for five days.

Blood Group

Specimen: Whole blood – EDTA

ABO and Rh (D) phenotype are done routinely. The frequencies of the four main groups of the ABO system in the caucasian population and their naturally occurring antibodies are:

ABO Group	Genotype	Frequency (%)	Naturally occurring antibodies
O	OO	46	anti A, anti B
A	AA/AO	40	anti B
B	BB/BO	9	anti A
AB	AB	5	none

Blood Group and Antibody Screen

Specimen: Whole blood – EDTA
If pregnant please note gestation on the request form.

Blood Group and Direct Coomb's Test

Specimen: Whole blood – EDTA

Blood Group and Hold Serum (G&H)

Specimen: Whole blood – EDTA
Transfusion form and special collection requirements.

See *Blood Banking*

Blood Group Phenotyping

Specimen: Whole blood – EDTA

Bone Marrow

Aspirate and trephine

Bone marrow trephines and aspirates are performed routinely at our main laboratory at 2 Makin Place, Deakin. Please phone the laboratory on 02 6285 9867 to arrange a booking. Results will be available as soon as possible.

Patient information sheets available upon request from:
Collection Centres, Capital Pathology website (www.capitalpath.com.au) and from the main laboratory.

Bone Turnover Markers

See *Deoxypyridinoline Cross Links (DPD)*
Hydroxyproline

Bordetella pertussis

Antibodies

Specimen: Serum – Gel

Reference Range: Supplied with report

Antibody (IgA) detection methods are available for diagnosis. For assessing current illness IgA detection is more reliable later in the disease course. In young children and infants IgA response may take several weeks to develop, so repeat testing may be necessary.

PCR

Polymerase Chain Reaction (PCR) requires a plain dry swab with metal wire , which can be taken as a nasopharyngeal or high oropharyngeal swab. The PCR is highly sensitive, and is best performed in the early stage of the illness (say the first two weeks of illness).

Culture

The culture for Bordetella requires a nasopharyngeal or oropharyngeal swab in transport medium. Culture is not as sensitive as PCR for isolating the causative organism. PCR is the preferred test in the early disease phase.

Borrelia burgdorferi Antibodies (Lyme Disease)

Specimen: Serum – Gel

Reference Range: Supplied with report

Brain Natriuretic Peptide (BNP)

Specimen: Serum – Gel

Reference Range: Supplied with report

BNP is a peptide produced by cardiac atrial cells in response to atrial stretch. The physiological response to volume overload sensed by atrial stretch receptors is to induce diuresis by the natiuretic action of BNP on the kidney. BNP can be used in the assessment of dyspnoea. A normal BNP almost excludes cardiac failure as the likely cause for shortness of breath, whereas a markedly elevated BNP makes cardiac failure the most likely cause. Intermediate values for BNP can be found in other conditions including renal failure, cor pulmonale and pulmonary hypertension.

Breast Aspirate Cytology

Breast Cyst Aspirate – *See Cyst Fluids*

Solid Lesions – *See Fine Needle Aspirate (FNA)*

Bronchial Specimens

Bronchial brushings
This specimen is obtained during bronchoscopy. The brush is placed in a 60mL specimen container containing saline and vigorously moved around against the sides of the container to dislodge the adherent cells.

Remove the brush and secure the lid of the container. Specimen should be refrigerated and transported to the laboratory as soon as possible.

Bronchial washings
Sterile saline (10–20mL) is pipetted down the flexible bronchoscope and sucked out again into a disposable plastic reservoir.

Specimen should be refrigerated and transported to the laboratory as soon as possible.

Brucella Antibodies

Specimen: Serum – Gel

Reference Range: Supplied with report

Brucellae are gram–negative bacilli infecting domestic animals, particularly cattle and pigs. Human infection occurs in farmers and meat–workers as an occupational hazard or by ingestion of contaminated milk products.

Over the past 30 years, brucellosis has been virtually eliminated from Australia. Cattle and human brucellosis, when found, will almost invariably have been contracted abroad.

Agglutination and Coomb's antibody tests detect both IgM and IgG antibodies. Results may be negative in the early stages of acute disease and sometimes antibodies are not detected at any stage. Seroconversion, however, or a > 4–fold increase in antibody titre, strongly suggests active infection.

Distinguishing previous resolved infection from chronic continuing infection is impossible using antibody tests alone.

Calvary
John James Hospital

'A Caring Community'

Providing Maternity, Surgical, Medical and Rehabilitation services to Canberra and surrounding communities

For a detailed Speciality Directory please email Robbyn Nedeljkovic
info@cjjh.com.au or 02 6281 8114

www.calvaryjohnjames.com.au

In the Tradition of the Sisters of the Little Company of Mary
with the values of hospitality, healing, stewardship and respect

C

C1 (Esterase) Inhibitor

Specimen: Serum – Gel
Specimen should be spun, separated and frozen immediately.

Reference Range: Supplied with report

CA 15–3

Specimen: Serum – Gel

Reference Range: Supplied with report

Mainly used for monitoring established breast malignancy.

Elevations are found in other benign and malignant conditions.

CA19–9

Specimen: Serum – Gel

Reference Range: < 40 kU/L

Elevated in pancreatic carcinoma. May be elevated in other gastrointestinal malignancies.

CA 125

Specimen: Serum – Gel

Reference Range: Age related reference ranges supplied with report

Mainly used for monitoring treatment and progress in established ovarian cancer.

Elevated levels may be found in other malignancies, or occasionally other non-malignant conditions including endometriosis, Pelvic Inflammatory Disease (PID), cirrhosis, renal failure and pregnancy.

C-Peptide

Specimen: Serum – Gel
Patient must be fasting.

Reference Range: Supplied with report

Cadmium

Specimen: Whole blood (EDTA)
Or 24-hour urine (nil preservative) or random urine.

Reference Range: Supplied with report

Used where toxicity is a possibility in workers involved with cadmium in industry.

Eating contaminated shellfish may raise levels. If in doubt, repeat after two weeks' abstention from seafood.

Calcitonin

Specimen: Serum – Gel. Spin, separate and freeze immediately.

Reference Range: Supplied with report

Calcitonin is a hypocalcaemic hormone produced naturally by the thyroid. There are no clinical excess or deficiency states.

Calcitonin's clinical use is as a marker for medullary thyroid carcinoma either as an isolated tumour or as part of Multiple Endocrine Neoplasia (MEN), type II, along with phaeochromocytoma and parathyroid tumours.

See *MEN (Multiple Endocrine Neoplasia)*

Calcium, serum

Specimen: Serum – Gel

Reference Range: Adults 2.10–2.60 mmol/L

A protein correction is routinely applied:

Adjusted Ca^{++} = observed Ca^{++} + {(43-Albumin) x 0.025}

Hypercalcaemia

The two most important causes are:

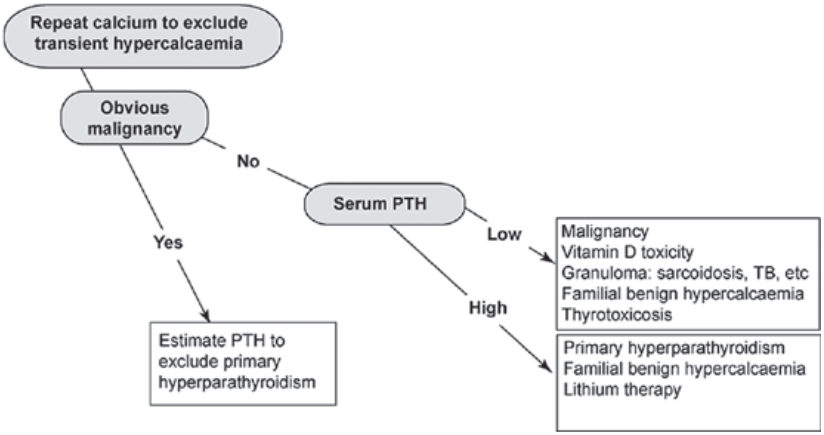
- **Hyperparathyroidism due to parathyroid adenoma**
  The parathyroid hormone (PTH) level is increased.
- **Malignant disease**
  Particularly metastatic tumour in bone, multiple myeloma or cancer of lung, breast or urinary tract, producing an ectopic PTH–like hormone, PTHrP (PTH related Peptide).

Other causes of hypercalcaemia include:

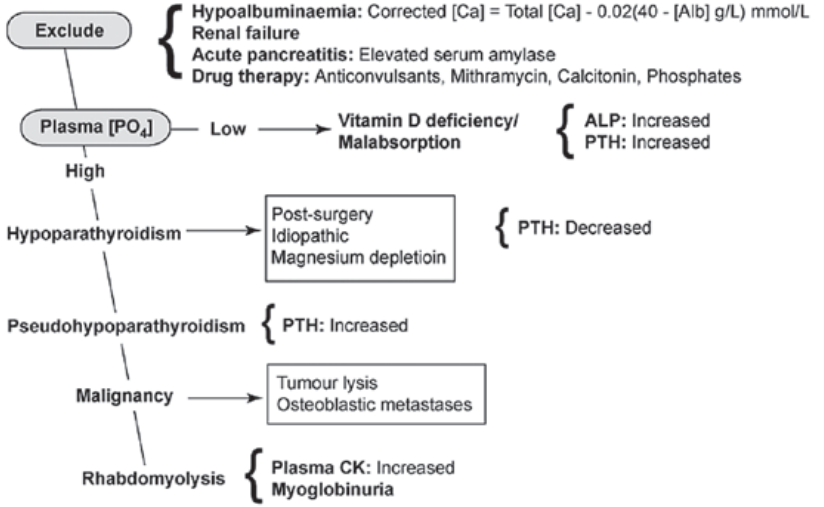
- Drugs – Vitamin D
  – Thiazides
  – Lithium
  – Antacids
- Thyrotoxicosis
- Sarcoidosis
- Familial hypocalciuric hypercalcaemia
- Renal transplantation
- Paget's disease with immobilisation

- **Hypocalcaemia**
- Chronic renal failure
- Vitamin D deficiency
- Hypoparathyroidism (usually post–thyroidectomy)
- Drugs – Anticonvulsants
  – Frusemide
  – Biphosphonates
  – Oral phosphate
- Malabsorption
- Pancreatitis
- Hypomagnesaemia.

Hypercalcaemia



Hypocalcaemia



Calcium, urine

Specimen: 24 urine (nil preservative) or random urine

Reference Range: Supplied with report

Some patients with recurrent renal calculi have a high urinary calcium output – idiopathic hypercalciuria. Serum calcium must be checked to exclude hypercalcaemia.

Campylobacter jejuni/coli

Specimen: Faeces is the usual specimen for acute infection
Serum – Gel for serology

Infections with Campylobacter jejuni or Campylobacter coli cause 60% of bacterial diarrhoea in the community. These organisms cause an acute enterocolitis which can be associated with intense abdominal pain. The average incubation period is 3 days; most patients recover within a week. Antibiotic treatment is usually not required but for severe infections erythromycin is effective. Treatment with quinolones is not recommended because resistance to them emerges commonly and rapidly. Untreated patients may excrete Campylobacter in their faeces for 2–3 weeks but transmissibility is low and it is not usual to put restrictions on otherwise healthy food handlers who are excreting the organism.

Candida

Candida albicans, the principal pathogenic yeast in humans, is a member of the normal flora of the gut, respiratory tract and vagina. It can gain dominance under certain conditions such as diabetes, antibiotic use and suppression of the immune system. Other species of Candida which are occasionally isolated cause the same type of infection as *C. albicans*. For most, the treatment is the same as for *C. albicans*.

The main sites and types of infection are:

- **Vulvo–vaginitis,** predisposing factors are antibiotics, diabetes, pregnancy and progestagens. Some individuals suffer from recurrent thrush for no detectable reason. A random glucose should be checked to exclude diabetes.

  Specimen: Vaginal swab in transport medium

- **Skin,** Infection occurs in warm moist areas such as the groin, perianal region, axillae, the breasts or in interdigital webs. It is often seen in those who frequently immerse their hands in warm water, such as dishwaters.

  Specimen: Skin swab in transport medium

- **Nails**, *Candida* can cause a painful red swelling of the nail fold resembling pyogenic paronychia. This may progress to nail involvement (onychomycosis).

  Specimen: Skin swab in transport medium, nail scrapings for mycology will grow Candida if present

- **Mouth**, Infections are found mainly in infants and show up as white adherent patches. Laboratory identification is not usually necessary but a swab will grow the yeast.

- **Systemic candidiasis,** Found in immunocompromised patients or in association with prostheses.

Cannabinoids (Drug Screen)

Specimen: Random urine (nil preservative)

Reference Range: Not detected

Cannabis is rapidly absorbed into fat depots with cannabinoids remaining detectable in the urine for up to 1–2 weeks after a single exposure. In chronic users, cannabinoids remain detectable for up to 6 weeks after cessation.

See *Drug Screen*

Carbamazepine (Tegretol)

Specimen: Serum – Gel
Trough level should be taken just before next dose (within one hour).
Peak level should be collected 3 hours post dose.

Therapeutic range: 15–40 umol/L

Carboxyhaemoglobin (carbon monoxide)

Specimen: Whole blood – EDTA

Reference Range: Supplied with report

The affinity of CO for Hb is 200x that of oxygen.

CO toxicity is mainly due to deliberate or accidental exposure to car exhaust fumes.

Levels fall about 15% per hour in air after removal of the CO source. Oxygen treatment, particularly hyperbaric, accelerates clearance of CO.

Carcinoembryonic Antigen (CEA)

Specimen: Serum – Gel

Reference Range: Age related reference ranges supplied with report

Cardiac enzymes / markers

See *Troponin*

Cardiolipin antibodies (aCL, ACA)

Specimen: Serum – Gel

Reference Range: Supplied with report

High levels of the IgG antibody are found in:

- Antiphospholipid syndrome
- SLE
- Other autoimmune disorders
- In some healthy people.

A first indication of aCL may be a “false positive” VDRL or RPR test during routine antenatal screening.

Low titre antibodies may be transient and are of uncertain significance.

See *Antiphospholipid Antibody Syndrome (APS)*

Carotene

Specimen: Serum – Gel. Protect from light

Reference Range: Supplied with report

Carotene levels are used in the diagnosis of carotenaemia, an orange–yellow colouration of the skin (but not conjunctivae) that can look like jaundice. The usual cause is a high intake of vitamin A precursors in carrot or other coloured fruit or vegetable juice but some systemic illnesses, including hypothyroidism, diabetes, liver and renal disease, can cause carotenaemia. Because carotene is lipid–soluble, hyperlipidaemias can give elevated levels. Low values have been used as an indicator of malabsorption but specificity and sensitivity are poor.

Catecholamines

Specimen: 24-hour urine
Plasma special tubes available (Plasma Metanephrine).

Reference Range: Supplied with report

Indications

Phaeochromocytoma
25% of the population have hypertension but only a fraction of these can or should be screened for phaeochromocytoma. Particular indications are:

- Symptoms, including sweating attacks, severe headaches, palpitations, nervousness, chest pain; flushing attacks are very uncommon
- Episodic hypertension (not always present)
- Moderate or severe hypertension in pregnancy or in young people
- Adrenal mass.

Neuroblastoma
These usually present as an abdominal mass in children under the age of five.

Number of specimens
When clinical suspicion is low, a single normal result is sufficient but when suspicion is high, up to three specimens should be tested, preferably collected during or just after symptoms.

Interpretation

The typical Phaeochromocytoma gives an elevated noradrenaline level. Occasionally the adrenaline level is elevated but usually it and the dopamine are near normal. In malignant phaeochromocytomas (10% of the total), dopamine tends to be elevated as well as noradrenaline.

The typical neuroblastoma shows a huge increase in dopamine.

Non–tumour elevations can be caused by anxiety, stress and exercise, particularly in the case of dopamine where elevations can be up to twice the upper limit. Small, isolated elevations of dopamine can usually be ignored.

Essential hypertension can be associated with a minor increase in noradrenaline, usually less than twice the upper limit.

Medication

Noradrenaline is increased by amphetamines, alpha– and beta–blockers, vasodilators, theophylline, phenothiazines and tricyclic antidepressants.

Noradrenaline is decreased by clonidine, methyldopa and bromocriptine.

Dopamine is massively increased by levodopa.

Cat Scratch Disease (CSD)

Specimens:

1. Serology
   - Paired sera, 2–3 weeks apart.
2. Histology, cytology
   - Formalin–fixed tissue, usually lymph node, for histology
   - FNA material for cytology.
3. Culture (slow–up to 28 days)
   - Whole blood – EDTA
   - Tissue or FNA in sterile container.

Cat scratch disease (CSD) is a self–limiting febrile illness with localised tender lymphadenopathy caused by *Bartonella* (previously *Rochalimaea*) *henselae* which is transmitted when a bacteraemic cat or kitten bites or scratches a human. In the United States, where one third of households have a cat, the annual incidence of CSD is 100 cases/million population, 80% of them in children.

Although CSD was first reported in 1931, it is only recently that *B. henselae*, a fastidious slow–growing, gram–negative rod, was identified as the pathogen responsible. Fleas transfer the organism from cat to cat and flea control is a useful preventive measure.

Severe systemic illness can occur in immunocompromised individuals.

Treatment

Pain relief may be required. Steroids are generally ineffective. There are no controlled trials to help choose an antibiotic but for those with prolonged fever and/or severe lymphadenitis, erythromycin, doxycycline, clarithromycin, ciprofloxacin, aminoglycosides and sulphamethoxazole/trimethoprim have been used. Therapy should be discussed with an infectious disease specialist.

Cerebrospinal Fluid (CSF)

Collect specimen in a sterile plastic CSF tube (These are available from the Stores Department. Glass tubes cause cellular distortion). Specimen must be received by the laboratory **within one hour** of collection due to rapid cellular deterioration.

Please phone a Courier on 02 6285 9877 for an urgent specimen pick up.

Ceruloplasmin (Copper Oxidase)

Specimen: Serum – Gel

Reference Range: Supplied with report

Ceruloplasmin is reduced in hepatolenticular degeneration (Wilson's disease), and Menk's kinky (steely) hair syndrome.

Oestrogens, anticonvulsants and inflammation cause elevations–ceruloplasmin is a late acute phase reactant. 95% of serum copper is carried by ceruloplasmin.

See *Copper*

Cervical Cytology

Pap smear

The quality of the Pap smear specimen is of critical importance if false negative cytology reports are to be minimised.

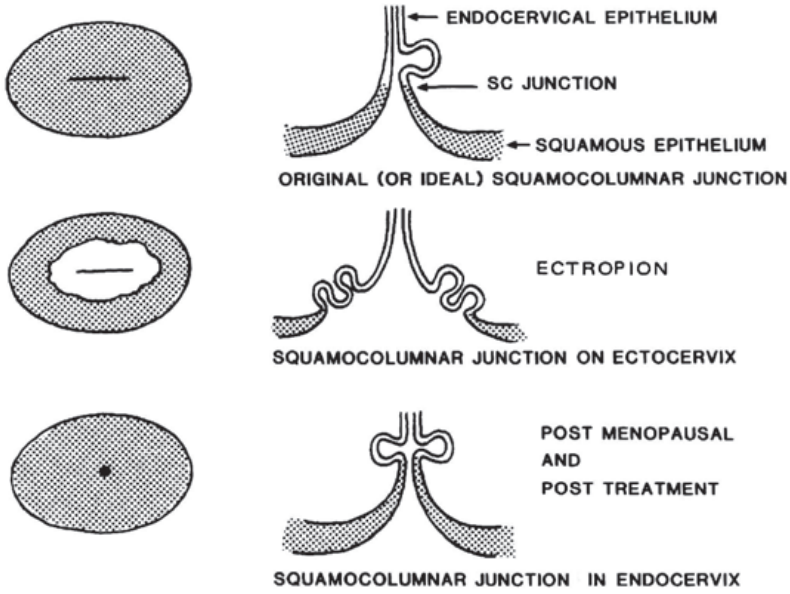
There are two main reasons for the occurrence of **false negative** Pap smear reports. One is laboratory error and the other is sampling error. At Capital Pathology we have put in place numerous quality assurance measures to minimise laboratory false negatives. Sampling errors can be reduced by taking an optimal sample. This also helps the laboratory in the interpretation of the smear.

Specimen collection

Principles of Sampling for Cervical Cytology

The aim of cervical cytology is to detect all cervical pre–cancer. The great majority of precancerous lesions which we need to detect arise in the transformation zone of the cervix, an area that is constantly "transforming" during a woman's reproductive life. The actual site will vary depending on numerous factors such as hormone influences or previous treatment. In this area the cells change from glandular to squamous cells, a process called metaplasia. Metaplastic cells are the transforming cells, which often have microscopic features of both squamous and glandular epithelium.

The squamous precursor lesions which occur in this area are patchy and they will not be visible with the naked eye. Glandular precursor lesions often occur in the same region but may also arise further up the endocervical canal. It is critical then that these areas are sampled.



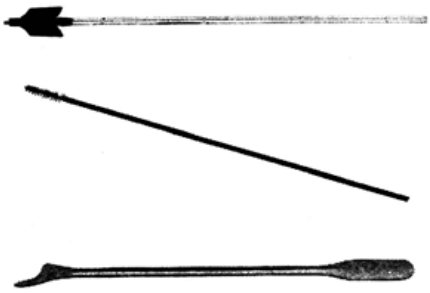
To the clinical observer the transformation zone may appear as a large eversion or ectropion. It is important to recognise this as a normal process.

Recommended technique for taking a cervical smear

Which instrument?

The appropriate instrument is chosen depending on the appearance of the woman's cervix and upon individual experience and preference.

- **The Cervex Sampler** (Cervex Brush®) is a broom-like instrument that consists of numerous specially designed filaments shaped to peak centrally, with shoulders graded to fit the shape of the cervix.
- **The Cytobrush** (cervical brush) is like a miniature bottle brush with small sharp bristles arranged in a spiral around a twisted wire.
- **Spatulas** should be plastic and are flat instruments shaped to fit the cervix.



(from top to bottom) Cervex Sampler, cytobrush (cervical brush), plastic spatula

The procedure

Cervex Sampler
Cervex Sampler has a peaked central portion and sloping shoulders. It is applied to the cervix with the peaked central portion situated in the endocervical canal. Then with some pressure it is rotated through the full 360° two to three times.

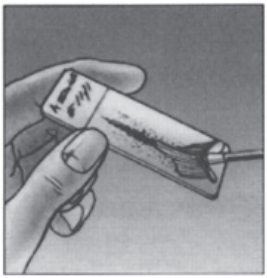
Plastic Spatula / Cytobrush Combination
If choosing a spatula it should optimally be used as a combined technique with a Cytobrush. The Cytobrush should never be used alone and its use is contra–indicated in pregnancy. In postmenopausal women and women previously treated for cervical precancer, the combined spatula and endocervical brush (Cytobrush) method is appropriate.

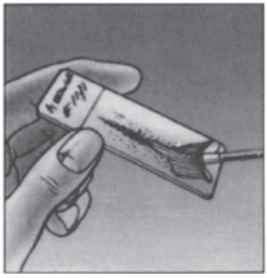
Visualise the Cervix
It may be difficult in some patients to visualise the cervix but every attempt needs to be made to get a clear view of the transformation zone, external os and into the endocervical canal. The cervix should be visualised under direct illumination with a speculum in–situ. However, when inserting the speculum it is preferable not to use lubricant gel as this may cause an unsatisfactory result. Warm water will suffice.

Making The Pap Smear

It is very important to make sure that whatever instrument is used, the sample is spread evenly and as smoothly as possible along the whole of the glass slide.

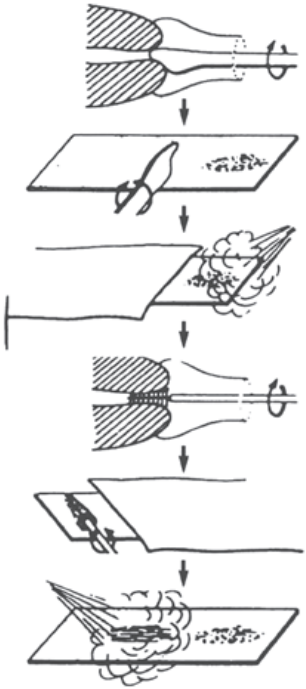
Cervex Sampler Technique





Prepare the conventional smear by 'smearing' first one side of the brush on the glass slide, and then the other. Apply fixative immediately. If desired a cervex sampler can then be used for *ThinPrep*, HPV, Chlamydia, and Gonorrhea.

Spatula Cytobrush Technique



Recommended spatula cytobrush technique. Most practitioners prefer to take the spatula/ectocervical sample first.

Fixing the Smear
The smear needs to be fixed as quickly as possible to ensure optimal cell fixation. Pump spray fixative is recommended. Fixative by immersion in 95% alcohol for 20 minutes is also acceptable. With each technique, the smear needs to be completely covered. Rapid fixation following smear taking is essential to prevent air–drying, which can occur within seconds. Sub–optimal fixation is the single most common factor precluding reliable cytological assessment.

Labelling the Slide
This may be done before or after taking the smear but it is crucial for accurate patient identification. The slide must be labelled in pencil (ink will wash off during the normal slide staining and processing) with the patient's surname, given name and date of birth. Stick–on labels are unsuitable as these may become detached during processing.

One Slide or Two
Generally it is preferable to use a technique which makes only one slide. If two instruments are used, optimal fixation is achieved by smearing the first half of the slide, covering the unsmeared half with a piece of cardboard and then immediately spray fixing the smeared half. The cardboard is then removed from the second half of the glass slide and the second instrument is smeared on this area. The whole smear can then be spray fixed. Spray fixing the first half again will not affect the final result, but a delay in spray fixation will be detrimental.

Blood
It is best not to have too much blood on a Pap smear, but sometimes it is unavoidable. If bleeding should occur during the taking of the smear it is best to proceed, making sure that squamous and glandular cells are sampled. Smears can be read through a bloody background but the smear will need to be repeated if it consists only of blood. The addition of *ThinPrep* may be worth considering if the smear is bloody.

Mucus
Gently remove any excess mucus from the cervix with a swab. Putting too much mucus on the slide will result in too scant a sample for accurate diagnosis and will preclude adequate fixation of the specimen.

Thinprep

ThinPrep is a slide preparation technique that has been shown to be associated with an increase in diagnostic accuracy of Pap smears.

ThinPrep is a simple procedure, which some studies have shown reduces the risk of false negative results. The fully automated slide preparation procedure produces consistent, easy to interpret slides, reducing the incidence of "unsatisfactory" reports, leading to fewer women being recalled for rescreening.

After preparing a conventional Pap smear, immerse the head of the sampling device in the *ThinPrep* vial and agitate the device to release the rest of the cell sample. Do not break off the head of the device. The *ThinPrep* vial is then transported to the laboratory where a *ThinPrep* slide is prepared. Gentle mechanical dispersion of the solution frees diagnostically important cells from blood, mucus and cell debris, and a filtration process results in the collection of epithelial cells which are placed on a slide in a thin layer prior to staining and microscopic assessment. Lubricant gel may cause a *Thinprep* slide to be unsatisfactory.

The resultant *ThinPrep* slide has a representative sample of cells, relatively free of mucus, erythrocytes and artefacts.

Some studies suggest that *ThinPrep* facilitates the identification of more high–grade lesions than conventional Pap smears.

ThinPrep is available in conjunction with the conventional Pap smear for a cost of $45. It has no associated Medicare rebate.

When *ThinPrep* is ordered, according to government regulations, the conventional Pap smear is not available as a bulk bill test, but can be billed at the rebate level.

If *ThinPrep* is required, simply endorse the request form by ticking the *ThinPrep* Pap test box. The *ThinPrep* vial can be used for chlamydia and gonorrhoea testing as well as for HPV testing.

A *ThinPrep* collection Protocol sheet is available from Client Services Department.

Please discard any expired *ThinPrep* vials and contact Stores Department for new supplies.

HPV Testing

See *Human Papillomavirus (HPV)*

Pap Smear Tracking Services

A number of supplementary routine and optional measures are provided to assist surgeries and their patients. These services may afford valuable and convenient support for practice follow–up procedures. Please contact the Cytology Department for any further information.

Tear–off Letters Addressed to Patients

Tear–off letters to the patient, indicating her result and appropriate follow–up, are attached to the bottom of the Pap smear result report. The patient's address is printed on the back of the tear–off section, ready for insertion into a window envelope.These letters are for doctors to send to their patients if they wish to do so.

Statistical Analysis of Results

Surgeries are routinely provided with a 12 monthly statistical analysis of the Pap smears taken. The number and percentage of smears which fall into the negative and all diagnostic categories are provided. Endocervical cell pick–up rates are also given. The overall laboratory percentages for all categories are provided for comparison.

A list of patients with abnormal smears in the 12 monthly period is attached to the statistical report.

Similar information can be provided on request for audit purposes.

Pap Smear Reminder System

Capital Pathology utilises a Pap smear reminder system for all patients with a smear repeat recommendation of less than two years. The aim of this service, in addition to other quality assurance measures, is to ensure that the number of women missing the Pap smear “safety net” is minimised. All letters are sent to the requesting medical practitioner.

A reminder letter that will be sent to medical practitioners listing all patients with abnormal or unsatisfactory results requiring repeat Pap smears.

Follow–up letters for patients who have had smears on which colposcopy was recommended. Doctors are routinely sent questionnaires requesting information on the diagnostic outcome for patients for whom colposcopy was recommended. (As part of our quality assurance protocols, we are required by government regulations, to correlate all high grade or inconclusive Pap smear results with any further clinical information.)

Additional Pap Smear data lists are available as required. Please contact Cytology Department to discuss further on 02 6285 9867.

Capital Pathology has been working closely with the ACT Cervical Cytology Register since its inception and is involved in its management and advisory committee functions. Unless the patient has opted off the Pap test register, her Pap test results will be automatically sent to the Pap Test Register (as required by legislation).

Capital Pathology also works closely with the NSW Pap Test Register.

Cervical Swabs

See *Chlamydia trachomatis*
Neisseria gonorrhoea

Chlamydia pneumoniae

Specimen: Serum – Gel
Preferably paired sera.

Reference Range: Supplied with report

C. pneumoniae is recognised as an important cause of atypical pneumonia, second only to *Mycoplasma*. Like the other *Chlamydia* it is an obligate intracellular bacterium.

Spectrum of disease
Many adults have antibodies indicating that the infection is common, initial infection being typically at age 5–15 years. It can cause pneumonia, severe pharyngitis, hoarseness, fever, cervical lymphadenopathy. Infection in young adults is usually of mild to moderate severity but can be sub–clinical or, in immunocompromised patients, severe. Incubation period averages 21 days and infection can recur. Its most recent (and surprising) association is with coronary artery disease, suggested by seroepidemiologic studies and finding the organism in atheromatous plaques.

Laboratory diagnosis is typically based on serology.

Chlamydia psittaci

Specimen: Serum – Gel
Preferably paired sera.

Reference Range: Supplied with report

C. psittaci is a pathogen endemic in all bird species. When a human inhales dust from fomites from infected birds, they can develop an infection which may present as an atypical pneumonia, headache, fever, rash, myalgia. Severity ranges from mild to moderate, occasionally severe.

Psittacosis is largely confined to bird–fanciers (the parrot family particularly) and poultry–handlers.

Laboratory diagnosis is based on antibody findings.

Chlamydia trachomatis

PCR (Polymerase Chain Reaction) testing is the diagnostic technique routinely used by Capital Pathology to detect *Chlamydia trachomatis*.

This procedure uses nucleic acid probes which are specific for all serovars of *C. trachomatis*. The main advantage of this technique is through the step of PCR target amplification. Here, specific DNA sequences of the cryptic plasmid are amplified exponentially, making their presence easy to detect by a colour formation step.

Enzyme Immuno–assay (EIA) and the Direct Fluorescent Antibody (DFA) techniques are no longer used as they have been found to be less sensitive and less specific. The PCR technique detects both “live” and “dead” Chlamydia (as do DFA and EIA), but experience indicates that treated infections will test as “negative” four weeks after treatment.

Specimen requirements
As with all laboratory testing, the accuracy of the result is affected by the quality of the specimen.

Chlamydia are Gram–negative, non–motile organisms that, due to their inability to synthetise ATP, exist as obligate intracellular pathogens of columnar, and not squamous epithelium. Appropriate cellular material must be collected in order to detect the organism.

Urine Collection
Urine specimens offer advantages in terms of ease of collection and patient comfort. PCR does not necessarily require cervical or urethral swabs, with the discomfort and inconvenience inherent in these procedures. It allows a prompt diagnosis of genitourinary chlamydia infection, in both males and females, using the first 20–30mL of the stream.

Patients should not have urinated for the previous two hours. Urine PCR replaces urethral swabs for diagnosis in all males and females in whom visualisation of the cervix is not indicated.

Cervical Samples
Remove mucus from exocervix with a large swab and discard. Insert another large swab into endocervical canal until tip is no longer visible. Rotate 3–5 seconds. Withdraw. Avoid contact with vaginal surfaces. If patient is also having a *ThinPrep* pap smear, then Chlamydia PCR can be done on the *ThinPrep* fluid. Please request "Chlamydia PCR testing" on request form.

Specimen Transport – Female Genital samples
Directly after sampling, vigorously agitate swab in Cobas Specimen Transport Medium for 15 seconds. Express liquid against side of the tube. Excess mucus should be removed by collecting it on the swab. Express any excess liquid from the mucus against the side of the tube. Remove swab and any excess mucus and discard.

Specimens should be stored at 4°C and transported to the laboratory as soon as possible.

Chloride

Specimen: Serum – Gel

Reference Range: 95–110 mmol/L

Chlorpromazine (Largactil)

Specimen: Serum – Plain clot
Do not use gel tube.
Collect pre dose (trough) specimen just before next dose.

Reference Range: Supplied with report

Cholesterol

Specimen: Serum – Gel

Total cholesterol is made up of three fractions:

Total = LDL + HDL + VLDL/IDL

Total and HDL cholesterols are measured directly in the laboratory

VLDL/IDL is estimated from the fasting triglyceride using the formula:

$$\text{VLDL} = \frac{\text{Triglyceride}}{2.2}$$

LDL is calculated by subtraction using the Friedewald formula:

$$\text{LDL} = \text{total} - \text{HDL} - \frac{\text{Triglyceride}}{2.2}$$

The formula is inaccurate and not used when the triglyceride is above 4.5.

Desirable range: the aim of lipid lowering therapy is to obtain a:

Total cholesterol of < 4.0

LDL cholesterol of < 2.5

HDL cholesterol of > 1.0

Triglycerides of < 2.0

See *Lipid Disorders*
Triglyceride

Cholinesterase

Specimen:

- Whole blood (Lithium Heparin) is preferred because it can be used for both red cell and plasma cholinesterase measurements when checking for insecticide poisoning and can be used in detecting scoline sensitivity
- Serum can be used, however, please specify serum cholinesterase is required
- Please include clinical details.

Indications:

1. Chronic exposure to organophosphate and carbamate anticholesterase sprays.

Organophosphates	Carbamate
Malathion	Carbaryl
Acephate	Methiocarb
Coumaphos	Methomyl
Chlopyrifos	Propoxur

These insecticide sprays are widely used by horticulturists.

Red cell and plasma cholinesterases should be measured before spraying commences to establish the individual's baseline reference range and at regular intervals thereafter.

If the baseline is unknown, estimations at 3–day intervals after removal from exposure will show recovery towards the baseline.

Because red cell cholinesterases are irreversibly inhibited by organophosphates (but not carbonates), levels remain low for the 4 month life of erythrocytes. The red cell level is the preferred test for monitoring low level chronic exposure.

2. Acute poisoning

Plasma cholinesterases fall sharply when acutely exposed to organophosphates or carbamates but recover to their previous levels within less than a week. Measure plasma (or serum) cholinesterase.

3. Scoline (suxamethonium) sensitivity

Measure serum or plasma cholinesterase and dibucaine number. Do not test within one week of scoline administration or two weeks of blood transfusion.

4. Other

Cholinesterase levels are reduced in chronic liver disease, renal disease, pregnancy/ oestrogens, acute illness.

Chronic Lymphocytic Leukaemia (CLL)

Typically CLL is an indolent disorder with a median survival of 10–14 years. In the early stages the patient does not require therapy but regular blood counts are performed to monitor the disease for increased activity.

Diagnosis is based on peripheral blood finding of lymphocytosis with cell marker analysis to demonstrate a monoclonal B cell population with light chain restriction. Bone marrow biopsy is not essential for diagnosis but is useful to determine bone marrow reserve and possibly outcome prediction.

The indications for treatment are:

- Bone marrow failure with anaemia, neutropenia or thombocytopenia
- Bulk disease with lymphadenopthy and/or hepatospenomegaly
- Constitutional symptoms such as fever, weight loss, night sweats
- Autoimmune complications, usually haemolytic anaemia.

Predictors of a poor outcome:

1. Clinical stage, RAI or Binet staging system
   - > 3 nodes with bone marrow failure: mean survival 2 years
   - > 3 nodes: mean survival 7–8 years
   - < 3 nodes with normal haemoglobin and platelets: approaches age–matched control survival.
2. Lymphocyte doubling time < 12 months
3. Abnormal karyotype on chromosome analyses
4. Marrow infiltration pattern, nodular being better than diffuse.

Treatment is not given on the basis of the high lymphocyte count alone. Initial therapy usually involves the alkylating agent chlorambucil with or without prednisone. Fludarabine is one of a new group of agents which is an alternative first line treatment for CLL.

Chronic Myeloid Leukaemia (CML)

CML is predominantly a disorder of middle life but the diagnosis is being made increasingly in younger patients. The disorder is characterised by a leucocytosis which is left shifted, typically showing the major increases in myelocytes and segmented neutrophils giving a bimodal differential white cell distribution. Blast cells are also present. The majority of patients are Philadelphia chromosome positive. The NAP score is low or absent.

CML is a state of bone marrow instability and after a variable time, median 3–4 years, the disease transforms into an acute leukaemia which is unresponsive to standard chemotherapy. The only potentially curative treatment is bone marrow transplantation. This may use a matched sibling donor or a matched unrelated donor transplant. Interferon is the treatment of choice in patients who cannot undergo bone marrow transplantation. Hydroxyurea is frequently used for cytoreduction.

Chyluria

Specimen: Random urine, ask for triglyceride

Chyluria is due to blockage of lymphatics, usually by filaria but sometimes by malignant disease. The urine is milky due to triglyceride–rich chylomicrons in the lymph.

CK / CKMB

See *Troponin*

Clonazepam (Rivotril)

Specimen: Plasma – Lithium heparin
Trough level is suggested, taken just before the next dose (within one hour).

Reference Range: Supplied with report

Clostridium difficile

Specimen: Faeces for *C.difficle* toxin

Reference Range: Not detected

A normal inhabitant of the bowel which can proliferate when other organisms are suppressed by antibiotics, particularly clindamycin or ampicillin. *C. difficile* produces toxins causing bloody diarrhoea and pseudomembranous colitis visible on endoscopy. Less severe forms of antibiotic–associated diarrhoea may also be associated with *C. difficile*.

Clostridium Tetanus Antibodies

Specimen: Serum – Gel

Reference Range: Supplied with report

Clozapine

Specimen: Whole blood – EDTA
Trough level at least 6 hours post dose. If peak level requested, collect at one hour.

Reference Range: Supplied with report

Coagulation Studies

Specimen: Plasma – Sodium citrate and EDTA
Sodium citrate tube must be filled to capacity.

Please request specific testing depending on clinical indications.

Anticoagulant Therapy

Heparin

The routine test for monitoring heparin therapy is the Activated Partial Thromboplastin Time (APTT).

Specimen: Plasma – Sodium citrate

Therapeutic range: 45–79 seconds

Ratio: 1.5–2.5

Notes:

- It is most important to obtain the correct volume of blood (i.e. exactly 4.5 mL added to the tube).
- The blood must not be collected from a limb being used for intravenous infusion.
- A periodic platelet count is recommended when monitoring heparin therapy Specimen required is 1 x 5 mL whole blood (EDTA).

Warfarin Therapy

The routine test for monitoring warfarin therapy is the International Normalised Ratio (INR).

Specimen: Plasma – Sodium citrate
The blood must be tested within six hours of collection and should be kept at 4–6 °C.

Therapeutic range: 2.0–4.5

Coagulation Screen

The following coagulation screen, in conjunction with a clinical history, provides the basis for the assessment of patients with suspected coagulation disorders.

The coagulation screen includes:

1. FBC
2. Prothrombin Time (PT)
3. Activated Partial Thromboplastin Time (APTT).
4. Thrombin Time (TT)
5. Fibrinogen.

Specimens: 1 x EDTA
1 x Sodium citrate

Reference Range:

PT	10–13 seconds
APTT	24–37 seconds
TT	10–15 seconds

Notes:

- It is most important to obtain the correct volume of blood (4.5 mL of blood added to tube) in the citrate tube.
- Traumatic venepuncture and/or long delay between collection and courier pick–up should be avoided, as should short collections. These may all affect the result.
- Skin bleeding time is not included as part of a routine screen.

D–Dimer

This screening test is elevated in patients with disseminated intravascular coagulation (DIC), pulmonary embolism (PE) and deep vein thrombosis (DVT).

Raised levels as an indication of reactive fibrinolysis have been reported in: Sickle Cell Disease, Liver Disease, Severe infection / sepsis, Pre–eclampsia and Malignancy.

Specimen: 1 x Sodium citrate

Factor Assays

Specimen: 2 x Sodium citrate

The specimens must be separated and frozen as soon as possible and transported on ice.

The Importance of the Bleeding History

A careful history is the most valuable part of an assessment. If the history of bleeding is convincing, a minor disorder may exist even if the laboratory tests are normal.

Ask specifically about:

- *Epistaxis* Especially if bilateral, recurrent, or requiring repeated cautery–unilateral epistaxis is often due to a local lesion.
- *Bruising* Specify size and whether spontaneous or traumatic; trunk and upper arm bruising are potentially more important than lower limb bruising–deep bruising and central induration is significant.
- *Lacerations* Particularly recurrent and prolonged bleeding
- *Surgery* Wisdom tooth extraction, tonsillectomy, other major surgery, blood transfusion requirement, prolonged postoperative hospital stay.
- *Menorrhagia* A bleeding disorder may potentiate anovulatory menhorragia.
- *Gum bleeding* When brushing teeth
- *Family history* Suggests inherited disorder
- *Drugs* Specifically ask about medication for headaches, arthralgias, particularly aspirin, NSAIDs.

Cocaine (Drug Screen)

Specimen: Random urine: (nil preservative)

Reference Range: Not detected

See *Drug Screen*

Coeliac Disease (Gluten Sensitive Enteropathy)

Coeliac disease, affecting at least 1:3000 (1:300 in some populations) is by far the commonest cause of intestinal malabsorption.

In severe cases it presents in infancy or childhood with failure to thrive and fatty diarrhoea.

Milder cases are more common and can easily be overlooked, particularly as many do not have intestinal symptoms.

Features of these milder cases can be one or more of:

- Anaemia
- Iron deficiency
- Folate deficiency
- Fatigue
- Diarrhoea, abdominal discomfort.

The disease is due to an intolerance to gliadin (gluten) found in wheat, barley and rye. Diagnostic antibodies are found in serum, and the mucosa of the small intestine shows flattened villi on biopsy. Withdrawal of gluten from the diet usually results in symptomatic improvement, normalisation of villi and disappearance of antibodies over the course of a year or so.

Dermatitis herpetiformis, which presents as a chronic pruritic papulovesicular skin rash, is a cutaneous manifestation of gluten sensitivity with the same diagnostic antibodies and improvement on gluten withdrawal.

When to test for Coeliac Disease

It is recommended that doctors test for the disease in patients with:

- GI symptoms (eg bloating, abdominal pain)
- Failure to thrive
- Type 1 DM
- Turner syndrome
- Down syndrome
- Family members who have coeliac disease
- Iron-deficiency anaemia
- Unexplained liver function test abnormalities and, in children, cryptogenic cirrhosis
- Osteoporosis
- Malabsorption syndromes.

Many disease states have been linked with coeliac disease and more commonly, gliadin antibodies. These include inflammatory arthritis, peripheral neuropathy, ataxia and infertility (including miscarriages). The evidence behind these associations is often weak, and in the case of gluten-sensitive ataxia, recent studies have essentially debunked the hypothesis. Infertility remains an area of uncertainty, though GESA suggest testing may be appropriate.

Testing recommendations

- Serum IgA TTG (order through TTG) and serum IgA.
- If IgA deficient, order IgG gliadin and consider referral to a gastroenterologist or clinical immunologist if clinically indicated.
- In high risk individuals (i.e. with associated conditions) order IgA TTG and if negative, consider ordering coeliac tissue typing.
- A negative DQ2 and/or DQ8 result significantly reduces the risk of having or developing coeliac disease (less than 1% in DQ2/8 negative patients)
- A positive DQ2 and/or DQ8 status confers an absolute risk of up to 4% of having or developing coeliac disease, and should not in itself be used to diagnose or label a person as being at high risk of having or developing coeliac disease.

Cold agglutinins (Cold Antibody Titre)

Specimen: Serum – Plain clot
Specimen to be kept at 37ºC until processed.

Reference Range: Not detected

Complement, C1q

Specimen: Serum – Gel

Reference Range: Supplied with report

Complement Fractions C3 and C4

Specimen: Serum – Gel

Reference Range: Supplied with report

The complement system is a cascading series of plasma proteins whose end–products cause bacterial lysis and remove the immune complexes found in post–streptococcal glomerulonephritis, SLE and other autoimmune disease. There is reduction of C3 in PSGN and of both C3 and C4 in SLE. They are increased in most other inflammatory states.

Complement, Total Haemolytic (CH50, CH100, Total Haemolytic Complement)

Specimen: Serum – Gel
Serum should be spun, separated and frozen within 20 minutes of collection.

Reference Range: Supplied with report

Congenital Adrenal Hyperplasia (CAH)

A group of inherited adrenal disorders due to enzyme defects, the commonest being 21–hydroxlylase deficiency by measuring 17–OH progesterone.

Severe CAH can present as hypokalaemia and dehydration in neonates; as virilisation in a female child; or as precocious puberty in males.

A milder and more common form of CAH can present as hirsutism in adult women. It can be detected by measuring serum 17–OH progesterone on a morning specimen collected during the follicular phase of the menstrual cycle.

See *17–Hydroxy Progesterone (17-OHP)*

Connective Tissue Diseases

A group of systemic autoimmune diseases characterised by the presence of fairly non–specific autoantibodies and associated with chronic inflammation of musculoskeletal structures. The main diseases are:

- Rheumatoid arthritis (1–2% of the population)
- SLE, Systemic Lupus Erythematosus (0.1% of the population)
- Sjogren's syndrome
- Diffuse scleroderma (0.002% of the population)
- Local scleroderma (CREST)
- Mixed connective tissue disease (MCTD)
- Dermato/poly/myositis.

The following tests may be useful : Anti Nuclear antibodies (ANA), Double stranded DNA (DS DNA), Rheumatoid factor, and Extractable Nuclear Antigens (ENA).

Diagnosis can be difficult or impossible in the early stages of these conditions which can evolve over months or years before enough features develop to establish the diagnosis.

Coomb's Test

Specimen: Whole blood – EDTA

Reference Range: Supplied with report

The Coomb's test detects antibodies directed against red cells.

The direct Coomb's test detects antibodies or complement which are coated on red cells as in autoimmune haemolytic anaemia, haemolytic disease of the newborn, incompatible transfusions, or drug–induced haemolysis, particularly that due to methyldopa or penicillin in large doses.

The indirect Coomb's test detects red cell antibodies in serum, as in maternal antibody screens in pregnancy or some autoimmune haemolytic anaemias. It is also used in cross–matching.

Copper

Specimen: Serum – Gel
or
Urine, 24-hour (nil preservative).
or
Liver biopsy placed in sterile container without formalin or saline
Occupational exposure – Urine.

Reference Range: Supplied with report

Wilson's disease is an autosomal recessive disease due to accumulation of copper in the body in toxic amounts. It presents usually at age 5–20 with unexplained liver disease, neurological or psychiatric symptoms, or Kayser–Fleischer corneal rings. There is an increased excretion of urine copper, but reduced serum copper. This is because ceruloplasmin, the protein which transports copper in serum, is reduced in Wilson's disease even though the total body load of copper, and its urinary excretion, are markedly elevated.

S. copper is reduced in protein–losing states and raised by oestrogens, pregnancy, or inflammatory states.

Cord Blood Testing

When haemolytic disease of the newborn is suspected, it is recommended that the cord blood is tested for Haemoglobin, Blood Group and Direct Coombs. If the direct Coombs is positive, further typing is performed and the coating antibody identified

Cortisol, serum

Specimen: Serum – Gel

Reference Range: a.m. 100–535 nmol/L
p.m. 80–480 nmol/L

There is marked diurnal variation, the peak at 09.00 hours being 50–100% higher than the trough at 23.00 hours.

Decreased levels

- Primary adrenal insufficiency (Addison's disease)
- Secondary deficiency follows adrenal suppression by steroid therapy
- Drugs–ketoconazole, phenytoin, metyrapone, steroids
- Decreased cortisol–binding proteins
- Hypopituitarism
- Exogenous steroids (variable).

Elevated levels

- Cushing's syndrome
  - pituitary adenoma or hyperplasia
  - adrenocortical tumour
  - ectopic ACTH from malignant tumour
- Stress, illness, depression, alcoholism
- Oral contraceptives, oestrogens, pregnancy

- Exogenous steroids (variable)
- Increased cortisol–binding proteins.

Cortisols are of limited value when monitoring replacement therapy.

See *Cushing's Syndrome*

Cortisol, urine

Specimen: 24 hour urine (nil preservative)

Reference Range: Supplied with report

Because only the unbound fraction of serum cortisol reaches the urine, this is a good screen test for Cushing's syndrome and a clearly normal result makes the diagnosis unlikely. In perhaps 10% of cases cortisol hypersecretion is intermittent.

Elevations up to 3x normal may be found in stress, depression or alcoholism.

Corynebacterium diphtheriae

A special medium is required to recover *C. diphtheriae* so the possibility of diphtheria must be specifically mentioned on the request form.

Tonsillar diphtheria is still occasionally seen in non–immunised persons. Skin infections with *C. diphtheriae* occur in the tropics and can be a source of infection.

Coxsackie Viruses

These are widely distributed enteroviruses associated with many different types of illness including minor febrile illnesses, the common cold, herpangina, pleurodynia, aseptic meningitis, myocarditis, post–viral fatigue syndrome, conjunctivitis, Type 1 diabetes and others. Infections are more common in summer and autumn. Virus can be recovered from throat swabs or rectal swabs and also conjunctival or vesicular swabs if lesions are present.

C–Reactive Protein (CRP)

Specimen: Serum – Gel

Reference Range: < 5 mg/L

CRP is the most useful of the acute phase reactants, rising sharply 4–8 hours after tissue damage by infection, inflammation or trauma. It returns to normal 2–3 days after disease activity has ceased. It can be regarded as a fast–changing ESR which, by contrast, rises and falls more slowly. It can be used as an indication of occult bacterial infection, suspected rheumatic fever, inflammatory bowel disease or other conditions where there is uncertainty whether symptoms are functional or due to organic disease. Chronic inflammatory diseases such as SLE can be monitored using serial CRPs and for this purpose it is a more useful test than the ESR.

Creatine Kinase (CK)

Specimen : Serum – Gel

Reference Range: Males 5–190 U/L
Females 5–165 U/L

Causes of an elevated CK – Possible causes:

Cardiac muscle
- Infarction
- Myopathy
- Myocarditis.

Skeletal muscle

Injury/Trauma
Crush; Surgery; IM injections; Ischaemia

Alcohol
Acute/Chronic alcohol excess

Infection
Influenza; Coxsackie A and B; Clostridia; Streptococcus pyogenes; Parasitic infestations

Endocrine
Hypothyroidism; Hyperthyroidism; Steroid myopathy

Metabolic
Hypokalaemia; Vitamin D deficiency; Carnitine deficiency; Carnitine Palmitoyl–tranferase deficiency; Hypoparathyroidism

Autoimmune
Polymyositis; Dermatomyositis

Exercise
Severe exertion; Marathon run; Convulsions; Paroxysmal myoglobinuria

Heat stroke
Malignant hyperpyrexia
Muscular dystrophy

Miscellaneous
- Macro–CK
- Malignancy
- Cerebrovascular disease
- Diabetic ketoacidosis.

Useful further tests
Troponin
Serum LD and AST
CK–Isoenzymes
Urinary myoglobin
Autoimmune immunology
DNA tests for muscular dystrophy

Developed by N. Walmsley 1995. Adapted with Permission.

Creatinine, serum

Specimen: Serum – Gel

Reference Ranges:

age	µmol/L
up to 7 days	20–60
8 days–4 years	20–40
5 years–11 years	30–70
Adult male	60–110
Adult female	40–80

Serum creatinine is widely used as a test of renal function both as a general screen, along with urine protein, for renal disease, and as a test for serial monitoring of renal function.

Creatinine reflects glomerular filtration rate (GFR), and it also reflects endogenous creatinine formation from skeletal muscle and to a much lesser extent, exogenous creatinine excretion from cooked meat.

GFR steadily reduces with increasing age but this is offset, though not completely, by diminishing muscle bulk and creatinine excretion.

The use of population–based reference range is even less satisfactory for creatinine than for other analytes because the individual creatinine range for a person remaining in good health is narrower than the traditional population range. When monitoring a potentially nephrotoxic process, reference should always be made to the individual's own range, as shown by creatinine results when disease–free, rather than to the population range.

Uses of serum creatinine

- To estimate GFR according to the MDRD formula (reference: Position Statement, The Australian Creatinine Consensus Working Group. Medical Journal of Australia, Vol. 183:3 pp138–142, 2005)
- For establishing an individual's baseline creatinine range
- Monitoring potentially nephrotoxic drugs, particularly in the elderly
  - NSAIDs
  - ACE inhibitors
  - Aminoglycosides
  - Diuretics
  - Lithium
  - Others
- Monitoring potential nephropathy e.g. in diabetics
- Monitoring established renal disease
- Monitoring renal transplant rejection
- Monitoring renal dialysis.

Creatinine, urine

Specimen: 24 hour urine, nil preservative

Reference Range: Supplied with report

24 hour urine creatinines are used when estimating creatinine clearance.

Wide variations in creatinine output for an individual are due to biological variation of ±20%. Sometimes compounded by incomplete (or over–complete) urine collects.

A creatinine concentration in a spot urine gives a way of compensating for urine concentration when expressed as the ratio, analyte/creatinine e.g. ACR (albumin/creatinine ratio) in diabetics.

Creatinine Clearance

Specimen: 24 hour urine (nil preservative) and serum gel
Must be accurately timed to 24 hours, plus serum obtained within collection period. Please report height and weight on the specimen.

Reference Range: 70–150 mL/min

CREST Syndrome

- Calcinosis
- Raynaud's phenomenon
- Esophageal hypomotility
- Sclerodactyly
- Telangiectases.

Also called limited scleroderma.
The ANA is usually positive showing the centromere pattern.

Creutzfeldt Jacob Disease

This is a prion disease where the clinical manifestations result from an accumulation of an altered prion protein molecule in the central nervous system. The diagnosis is suspected from the history and clinical examination. Confirmation of a case can be made by histology on brain biopsy or post mortem tissue supported by molecular biology techniques to look for expression of gene sequences.

Please contact the Director of Clinical Pathology to discuss further details on 02 6285 9895.

Crohn's Disease

Laboratory abnormalities can include:

- Iron deficiency
- Vitamin B12 deficiency
- Anaemia due to combinations of chronic disease and iron and B12 deficiency
- Raised ESR and CRP
- Reduced albumin
- Hypokalaemia.

Cryoglobulin and Cryofibrinogen

Specimen: Cryoglobulins – Serum (plain clot)
Cryofibrinogen – Plasma (Sodium Citrate) and plain clot.

Reference Range: Not detected

Specimen must be kept at 37°C prior to analysis

See *Cold Agglutinins (Cold Antibody Titre)*

Cryptosporidium, faeces

Cryptosporidium is now recognised as a cause of acute gastroenteritis, particularly in children. It is a notifiable disease. It is found in a variety of hosts and transmission from farm livestock or pets to humans can occur. Person to person transmission also occurs and has been responsible for outbreaks in child care facilities.

Diagnosis is by use of a special stain for oocysts in faeces and will be done on specific request. The typically watery diarrhoea usually settles without treatment within 10 days (range 1–20 days). In immunocompromised patients (especially those with HIV), it may cause a severe prolonged diarrhoeal illness. In this situation specialist advice should be sought on treatment.

Cryptococcal Antigen

Specimen: Serum – Gel or Cerebrospinal Fluid

Reference Range: Not detected

Crystal Identification

See *Histopathology*
Synovial Aspirate

Cushing's Syndrome

The main causes are:

- Excess ACTH (adrenocorticotrophic hormone) produced by the pituitary
- Excess ACTH produced by non–endocrine tumour particularly lung
- Adrenal tumours.

Basic screen test: 24-hour urine free cortisol

Follow–up test: Dexamethasone suppression test

Isolated serum cortisol is not recommended as a screen test though a level below 500 nmol/L in a specimen collected before 10.00 hours makes Cushing's unlikely.

Clinical features of Cushing's include obesity, diabetes, hypertension, plethora, muscle weakness, striae and osteoporosis.

Cyclosporin

Specimen: Whole blood – EDTA

Reference Range: Supplied with report

Cyst Fluids

Cyst fluids should be placed into a labelled specimen container and refrigerated at 4ºC. Storage overnight is satisfactory. It is preferable to send the whole specimen to allow for concentration of poorly cellular specimens and for preparation of a cell block. The cell block is then available for special staining including immunohistochemistry should this be necessary for the diagnosis.

If the volume of cyst aspirate is very small (i.e. several drops only), smears can be made directly from the fluid. These should be rapidly fixed with pump spray fixative. For greater detail on smear preparation please see section on Fine Needle Aspirate (FNA). Alternatively a small amount of normal saline can be added to the specimen container. Please note the volume of saline added on the request form.

Cystic Fibrosis PCR

Specimen: Whole blood – EDTA x 2

Two generations of family tree needed with details of any cases of cystic fibrosis. Need ethnic background, e.g. Caucasian etc.

Affects 1:3000 infants. Cystic fibrosis is carried on a recessive gene which can be identified in 70% of carriers by DNA testing. The underlying defects of exocrine gland function show up in respiratory tract, pancreas and sweat glands. Typically, presentation is in infancy or childhood with recurrent pulmonary infections and sometimes with malabsorption.

Cystinuria Screen

Specimen: Urine mid-stream

Reference Range: Detected or not detected

Cystinuria with an incidence of 1:10,000 is one of the commonest of the inborn errors of metabolism. Failure of the renal tubules to reabsorb cystine from urine results in excretion of a high concentration of poorly soluble cystine which can precipitate to form cystine stones 1–2% of all renal calculi.

Cytogenetics

Specimen: Amniotic Fluid
Bone marrow
Curettings from products of conception
Fetal tissue
Blood for karyotype – Whole blood, Lithium Heparin.

Collect specimens on a Monday – Thursday. Please inform the laboratory on 02 6285 9803 if collection is required out of these times.

Three major areas of testing are carried out:

1. **Prenatal diagnosis from:**
   - Amniotic Fluid
   - Chorionic Villi.

2. **Clinical diagnostic work for cases of:**
   - Infertility or Multiple Miscarriages
   - Stillbirth (Where Clinical Indicators are Present)
   - Babies With Multiple Malformations, With or Without Neurological Dysfunction
   - Children With an Unusual Appearance Who Are Developmentally Delayed or Mentally Retarded, Especially if There are Coexisting Congenital Defaults
   - In Some Children With Speech Delay, Behavioural Problems, Isolated Mental Retardation
   - Genital Abnormality, Hypogonadism.

3. **Cancer Cytogenetics:**
   - Leukaemia
   - Lymphomas
   - Solid Tumours.

Cytology

The Cytology Department at Capital Pathology is a fully accredited comprehensive cytology laboratory, which processes a full range of gynaecological, non–gynaecological and fine needle aspiration specimens. We participate successfully in the RCPA External Quality Assurance programme and our Performance measures are amongst the best in Australia.

See *Bronchial Specimens*
Cerebrospinal Fluid (CSF)
Cervical Cytology (includes Pap smear and HPV testing)
Cyst Fluids
Effusion Fluids (Pleural, Ascitic, Synovial)
Fine Needle Aspirate (FNA)
HPV testing
Nipple Secretions
Sputum Cytology
Urine Cytology

Cytomegalovirus (CMV) Antibodies

Specimen: Serum – Gel

Reference Range: Supplied with report

Interpretation

IgM antibodies, reported as +ve or –ve, are detectable for a period of about 6 months from the time of commencement of a CMV infection. False positives occur during some other viral infections, notably those due to EBV.

IgG antibodies, reported in units/L, become detectable soon after the commencement of infection and remain positive for life, usually at a level > 20 units/L. A rising titre in paired sera is the best evidence of current infection. Most adults are IgG CMV positive indicating previous infection.

The virus itself persists in latent form throughout life after recovery from the initial infection and can be reactivated in an immunocompromised patient.

Clinical infection can present in several ways:

- Subclinical infection is common, particularly in childhood.
- A mononucleosis–like syndrome is found in adolescents and young adults, spread by sexual and other intimate contact. CMV differs from EBV mononucleosis in its absence of heterophile antibodies. Exudative pharyngitis and cervical lymphadenopathy are rare. The illness can be severe with fevers and profound fatigue lasting several weeks and the virus can cause hepatitis. As in EBV infections, variant lymphocytes are a feature.
- Congenital infections range from inapparent to severe with congenital abnormalities or intrauterine death. Diagnosis is the identifying of the virus in urine collected during the first week of life.
- Immunocompromised patients can develop severe generalised disease.

D

D–Dimer

Specimen: Plasma – Sodium Citrate

Reference Range: < 0.2 mg/L

D–dimer is a fibrin break–down product and elevated levels can be found in any situation where there is thrombosis.

Clinically the test is used when DVT (deep vein thrombosis) with or without PE (pulmonary embolism) is suspected. A normal D–dimer level makes DVT/PE unlikely.

Elevated levels have a poor predictive value for DVT/PE as they are found in many clinical situations including chronic inflammatory disorders, malignancy, post–operative states and acute rheumatic conditions.

See *Coagulation Studies: Coagulation Screen*

Dehydroepiandrosterone and Dehydroepiandrosterone Sulphate (DHEA, DHEAS)

Specimen: Serum – Gel

Reference Range: Supplied with report

DHEA and its sulphate DHEAS are the most abundant androgenic steroids secreted by the adrenal cortex. DHEAS is more commonly measured than DHEA and for practical purposes the two estimations provide the same information.

Their uses are:

- In the investigation of virilism – levels are usually > 3x upper limit of normal
- In the investigation of hirsutism when there is a more than 2–fold increase in free testosterone
- To differentiate Cushing's disease (minor elevations of DHEAS) from adrenal neoplasms where there are large increases
- For monitoring steroid suppression therapy in congenital adrenal hyperplasia.

Dementia (Acute or Chronic Organic Reaction)

The cardinal features are alteration in level of consciousness, confusion and focal or global cognitive impairment. It is important to exclude treatable causes, and so consideration can be given to the following lists as a guide.

Causes

- Cerebral lesion–infarct, haemorrhage, raised intracranial pressure, trauma, abscess, tumour which could be primary or secondary
- Sepsis – consider septicaemia , bacterial endocarditis, subphrenic abscess, meningitis, urinary tract infection, encephalitis
- Specific infections – HIV, neurosyphilis, Creutzfeldt-Jakob disease
- Metabolic – diabetes, acidosis, uraemia, decompensated liver disease, hypothyroidism (myxoedma madness), Cushing's Disease, hypercalcaemia (psychic moans), hypoxia
- Drug reactions
- Hospitalisation in the elderly
- Acute or first presentation of psychosis / psychoaffective disorder
- Poisoning – mushrooms, prescribed medications in overdose, illicit drugs, lead, ethanol
- Primary dementias and neurological degenerative disorders – Alzheimer's disease, multi-infarct demetia, Huntington's chorea, storage diseases.

Tests to consider

- FBC and ESR
- CRP
- U&E, LFT, calcium, magnesium
- Vitamin B12 and folate
- Syphilis serology
- Thyroid function tests
- Lumbar puncture and CSF examination
- Blood cultures
- HIV serology
- Serum cortisol, or 24-hour urine cortisol
- Plasma glucose
- Blood gases
- MSU
- CXR, ECG, cerebral CT.

Dengue Antibodies

Specimen: Serum – Gel

Reference Range: Supplied with report

Dengue, a painful febrile illness ("break-bone fever") caused by a mosquito-borne arbovirus, is found all over the Pacific including Fiji. Typically it occurs in outbreaks but can be sporadic.

Diagnosis is by measuring IgM and IgG antibodies preferably in paired sera. Although both antibodies may date back to an earlier infection, the ratio of IgM to IgG, or rising titres in paired sera, will help decide whether infection is recent.

Deoxypyridioline Cross Links (DPD)

Specimen: Urine – Collect the second morning void into urine container (nil preservative).

Reference Range: Supplied with report
Useful bone turnover marker.

Desipramine (Pertofiren)

Specimen: Plasma – Lithium heparin

Reference Range: Supplied with report

See *Antidepressant Drugs, tricyclic*

Dexamethasone Suppression Test

By arrangement with collecting rooms or main laboratory

Specimen: Serum – Gel

Protocol: **Day 1**, collect baseline cortisol specimen at 08.00-09.00 hours.
Give 1 mg dexamethasone orally at 23.00 hours

Day 2, collect cortisol specimen at 08.00–09.00 hours

Reference Range: Cortisol < 100 nmol/L at 09.00 hours post–suppression

Normal suppression, especially to < 50 nmol/L makes Cushing's syndrome unlikely.

Failure to suppress is less helpful and may occur with a range of conditions including:

- Cushing's syndrome
- Endogenous depression
- Chronic alcoholism
- Drugs–phenytoin, barbiturates, oestrogens
- Significant stress or illness.

Diabetes insipidus

The essential feature is inadequate ADH (antidiuretic hormone) from the hypothalamus/posterior pituitary causing excessive loss of dilute urine from the kidney; or a kidney which is unresponsive to ADH. The usual problem is to separate compulsive water or simple urinary frequency from true diabetes insipidus in a patient complaining of polydipsia and polyuria.

- Other causes of polyuria need to be eliminated.
- diabetes mellitus
- hypercalcaemia
- renal disease
- medication: lithium, diuretics.

A 24–hour urine should be collected to check whether urine volume is genuinely excessive e.g. > 4 L. in severe disease, volume can be as high as 18 L.

The final screen test is urine osmolality after 8–12 hours of water deprivation overnight. A urine specimen is collected on waking and 1/2–1 hour later before drinking anything, the osmolality in these two specimens indicating ability to concentrate urine.

The person with diabetes insipidus will excrete a large volume of dilute urine. Water restriction should only be performed under medical supervision, due to the risk of serious dehydration.

Diazepam

Specimen: Plasma – Lithium heparin tube
Trough level is taken before next dose (within 1 hour).

Reference Range: Supplied with report

Dibucaine Number

Specimen: Serum – Gel

Reference Range: Supplied with report

The dibucaine number detects the qualitative difference in cholinesterase enzymes in scoline–sensitive persons.

See *Cholinesterase*

DIC (Disseminated Intravascular Coagulation)

This is an uncommon condition in which there is a generalised consumption of plasma clotting factors and platelets resulting in fibrin deposition within the microcirculation.

Secondary haemorrhagic events are due to the consumption of normal clotting factors and secondary fibrinolysis.

Clinical setting for DIC:

- Major trauma
- Septicaemia (usually with acidosis)

- Obstetric crises (placental abruption, eclampsia, retained dead fetus)
- Malignancy (acute promyelocytic leukaemia).

Tests for DIC include FBC (with blood film for fragments), platelet count (as part of FBC), prothrombin time, APTT, fibrinogen assay and D–dimers.

Digoxin

Specimen: Serum – Gel
Specimen should be collected at least 6 hours post dose.

Therapeutic Range: 0.6–1.3 nmol/L

Dilantin

Specimen: Serum – Gel
Tough level is taken just before next dose (less than one hour).
Peak level is collected between 4–7 hours post dose.

Reference Range: 40–80 umol/L therapeutic range adults

Disaccharidases

See *Histopathology*

Disopyramide (Rythmodan)

Specimen: Plasma – Lithium heparin

Reference Range: Supplied with report

DNA Antibodies (Anti–double Stranded DNA)

Specimen: Serum – Gel

Reference Range: Supplied with report, qualitative test

Interpretation: < 1:20 or 1:40 equivocal
> 1:80 supports diagnosis of SLE

Anti–ds DNA is positive at 1:80 or higher in 60–80% of SLE. Low titres may be seen in rheumatoid arthritis, autoimmune hepatitis and in other immunological disorders.

Dothiepin (Prothiaden)

Specimen: Plasma – Lithium Heparin
Suggest take a trough level just before next dose (within one hour).

Reference Range: Supplied with report

See *Antidepressant Drugs, tricyclic*

Down's Syndrome Screening

See *Prenatal Testing*

Doxepin (Sinequan)

Specimen: Plasma – Lithium Heparin
Suggest take a trough level just before next dose (within one hour).

Reference Range: Supplied with report

See *Antidepressant drugs, tricyclic*

Drug Screen, Screen for Drugs of Abuse

Specimen: Urine – Random

Reference Range: Detected or Not Detected

Tests for drugs of abuse
Qualitative drug screening may be undertaken to determine whether a person has taken a medication or drug. Testing is useful to ascertain:

- Compliance with prescribed drugs e.g. methadone
- Use of non–prescribed drugs
- Accidental or deliberate abuse of illegal drugs.

Factors affecting a valid specimen
Urine creatinine concentration is assayed to detect diluted specimens. This can be caused by surreptitious addition of tap water or excessive fluid intake prior to collection. A urine creatinine level < 2.0mmol/L indicates a dilute specimen. A repeat specimen should be considered.

The temperature of the specimen indicates if it is freshly passed. If the specimen is fresh, the temperature should fall in the range of 33–38˚C. If the temperature is not in this range, specimen substitution should be suspected.

Adulteration can affect the final results, and pH, smell and visual checks are all performed to check for these possibilities.

Factors affecting detection time:
- Usage pattern
- Drug and dose of drug used
- Urine concentration
- Assay method and cut off value used.

Chain-of-custody
Urine samples are collected under supervision into specially designed beakers with temperature strips affixed. The supervising collector is specially trained in the requirements surrounding urinary drug screening and he/she records the temperature of the specimen and divides the specimen into three tubes. Each tube is labelled and signed by the client to verify identity. The tubes are then individually packaged with tamper evident tape and placed in secure tamper-proof satchels for transport to the laboratory.

An audit trail is maintained such that every person involved in the collection, transport and checking process is required to sign either the chain-of-custody form or the transport form accompanying the specimens. The integrity of the samples and transport satchel is noted on the forms throughout the process. These forms are stored in a secure facility for future reference if required

Interpretation of Results

The presence of each drug or metabolite is tested for at or above a predefined cut-off level. These levels are dictated by International Standards for urine drug testing and defined in the Australian/New Zealand Standard AS/NZS 4308:2008. The “cut-off” levels are established because the aim of workplace testing is usually to identify significant residues of the targeted drug, not minute traces. For a result to be “non-negative”, the amount of the drug detected must be at or above the “cut-off” level. If a drug is detected but the level is below the “cut-off” the result will be negative. The “cut-off” levels for some classes differ for screening and confirmation. This is due to the non-specific nature of the screening assay versus the highly specific nature of the GC/MS confirmation.

A confirmed positive result reveals the presence of a drug in the specimen at or above the “cut-off” level. It gives no information about how or when the drug was taken. It also does not provide an indication of impairment. A positive result may relate to previous drug use with no current physical effects. Positive results are reported as ‘Detected’, while a negative result is reported as ‘Not Detected’.

Drugs Detectable

The general screen detects groups of drugs including: opiates; methadone; amphetamines; cocaine; benzodiazepines; and cannabis. Further confirmatory testing can be undertaken on request.

Please note Capital Pathology collection centres have accredited staff who have completed the ASNZ4308:2008 training.

D–Sialated Transferrin Assay

Specimen: Collect nasal / fluid drips into sterile container

Reference Range: Supplied with report

Used for identification of CSF in a discharge fluid.

E

Ear Swabs and Infections

Otitis media

It is not possible to sample the middle ear routinely and the flora of the external meatus bears no relation to that behind the drum. Antibacterial treatment is therefore empirical, based on studies which show the role of *H. influenzae, Strep. pneumoniae, Moraxella* (previously *Branhamella*) *catarrhalis*, and *Strep. pyogenes*. Amoxycillin, cotrimoxazole and amoxycillin–clavulanate provide effective treatment. Cefaclor has insufficient activity against *Strep. pneumoniae* for it to be recommended in this situation.

Otitis externa

Ear swabs are taken from just inside the external meatus. The commonest pathogens are *Staph. aureus* and *pseudomonas aeruginosa. H. influenzae* and *S. pneumoniae* are also frequently isolated.

ECGs

These are performed at Capital Pathology Collection Centres. With a request for "ECG Trace only" you will be provided with the ECG for your own interpretation. With a request for "ECG Trace and report" you will be provided with the ECG and a report from a cardiologist.

Please indicate on the request form when an ECG is urgent.

Holter Monitor services are also provided by Capital Pathology, with a report and interpretation from a cardiologist.

Please contact your most convenient Collection Centre to arrange an appointment.

Effusion Fluids (Pleural, Ascitic, Synovial)

Effusion fluids should be placed into a labelled specimen container and refrigerated at 4ºC. Storage overnight is satisfactory. It is preferable to send the whole specimen with a minimum of 100 mL, to allow for concentration of poorly cellular specimens and for preparation of a cell block. The cell block is then available for special staining including immunohistochemistry should this be necessary for the diagnosis.

eGFR (Glomerular Filtration Rate)

GFR can be calculated in a number of ways. The MDRD formula has now become available to calculate estimated GFR (eGFR) based on serum creatinine, age and sex. eGFR is routinely reported on patients aged 18 years and over. The aim of reporting eGFR is to detect kidney disease at a time when there may be benefit from therapy to prevent complications. There are a number of limitations that apply to using the formula. The formula has not been fully validated in certain populations (Aboriginal, Asian and Torres Strait Islander), and is not appropriate for pregnancy, obesity, dialysis, rapidly changing renal function or with some diets (vegetarian).

Full details of the application of eGFR can be found in the position statement from Medical Journal Of Australia 2005 Vol 183:3 pp138–142.

Electrophoresis of Serum Proteins

Specimen: Serum – Gel x 2

Reference Range: Supplied with report

Serum protein electrophoresis (EPP) is an essential test when interpreting a raised total globulin or immunoglobulin to determine whether a paraprotein is responsible for the increase.

A. Monoclonal bands, paraproteins, M–bands

A paraprotein, which appears as a sharply defined abnormal band on serum EPP, consists of a single population of identical immunoglobulin molecules formed by a clone of neoplastic plasma cells. They can be benign (MGUS) or malignant (myeloma, macroglobulinaemia or lymphoma).

Occasionally an M–band is due to an extra–medullary plasmacytoma.

Benign paraproteins (MGUS – Monoclonal Gammopathy of Uncertain Significance)

These paraproteins, composed of IgG, IgA, or IgM, appear with increasing frequency in the elderly – 8% of patients over age 70. They can be transient, particularly in younger individuals. Characteristics are a relatively low concentration (total IgG < 20g/L, IgA or IgM < 10g/L), normal blood picture, no depression of normal immunoglobulins, no urine Bence Jones Protein. Bone marrow examination is not usually indicated but if done will show < 10% plasma cells with no atypical cells.

MGUS can be regarded as a “carcinoma in situ” of the immune system and like any in situ lesion it has the capacity to progress to malignancy, quickly or slowly, or it can remain largely static for the life of the individual, hence the need for monitoring at least annually. Evolution to multiple myeloma (or macroglobulinaemia) occurs in 25% of patients.

Malignant paraproteins

These consist either of IgG or IgA (multiple myeloma) or IgM (Waldenstrom's macroglobulinaemia or lymphoma) and show the invasive characteristics of malignancy with infiltration and destruction of bone marrow.

Characteristics of a malignant, as compared with a benign, paraprotein are:

- Anaemia, thrombocytopenia, neutropenia
- Immune paresis, i.e. reduction of normal IgG, IgA and IgM
- High and rising concentration of paraprotein
- Bence Jones protein (free light chains) typically appears in the urine in myeloma
- Myeloma causes diffuse osteoporosis or lytic lesions, either of which can cause back pain, compression fractures or bone pain in other parts of the body such as ribs or pelvis
- Hypercalcaemia, raised ALP
- Renal impairment
- Hyperviscosity syndrome, particularly with macroglobulinaemia.

Bone marrow examination and skeletal survey establish the diagnosis.

Where there is a strong clinical suspicion of myeloma (pancytopenia, osteolytic lesions, bone pain, immune paresis) but no serum M–band, urine EPP may show Bence Jones myeloma in which the band (of free light chains) is found only in urine.

Polyclonal increases

These are reflected in serum proteins as an increase in total globulins and in the EPP as an increased density in the gamma zone.

They are caused by any chronic inflammatory or liver disorder:

- Chronic hepatitis, viral, alcoholic or autoimmune
- Connective tissue disease
- Chronic inflammatory bowel or pulmonary disease
- Parasitic infestations.

Electrophoresis of urine

Specimen: Random urine

Endomysial Antibodies (EMA)

Specimen: Serum – Gel

Reference Range: Supplied with report

EMA is no longer recommended for coeliac disease diagnosis, as TTG is the preferred test.

See *Coeliac Disease*

Eosinophils

Specimen: Whole blood – EDTA

Reference Range: Adults < 0.6 x 10^9/L

Eosinophilia

Eosinophil counts are part of a routine blood count. The common causes of eosinophilia are drug effect, allergy or parasitic infestation of the gut.

The differential diagnosis includes:

- Allergic disorders
- Parasitic colonisation – hookworm, filaria, hydatids, toxocara
- Drug administration
- Skin disease, e.g. eczema, psoriasis, pemphigus
- Collagen disorders, especially polyarteritis nodosa
- Infections, e.g. TB, scarlet fever
- Malignant disease, e.g. lymphoma, Hodgkins disease, ovarian cancer
- Pulmonary eosinophilia
- Hypereosinophilic syndrome (a rare myeloproliferative disorder).

Epilim (Valproate)

Specimen: Serum – Gel
Suggest trough level collected just before next dose. If peak requested collect between 0.5–1.0 hours for Syrup, 1–3 hours for capsules and 2–6 hours for coated tablets.

Reference Range: Therapeutic 350–700 umol/L

Epstein-Barr Virus Antibodies

Specimen: Serum for heterophile antibodies, liver enzymes, EBV antibodies
Whole blood – EDTA for blood count.

Epstein–Barr Virus is the causative agent in infectious mononucleosis which is characterised by lymphadenopathy, pharyngitis, fever, variant lymphocytosis in the blood film and transient heterophile antibodies in serum. Fatigue can be profound and continue for months.

EBV antibodies
Where EBV infection is suspected but heterophile antibodies remain negative–as happens in 10% of young adults, and 50% of children – the measurement of EBV antibodies will provide a definitive answer.

VCA (Viral Capsid Antigen) IgM antibodies appear at 4–7 days after symptoms develop and persist for 2–4 months, occasionally up to 1 year. Their presence indicates current or recent infection.

VCA IgG antibodies appear at the same time as IgM but persist for life.

EBNA (Anti EB Nuclear Antigen) antibodies appear 3–6 weeks after onset and also persist for life. 80% of the population are EBNA +ve, indicating past infection. A minority of people infected with EBV never develop EBNA antibodies but will have a positive VCA IgG test to indicate their past exposure.

Erythropoietin (EPO)

Specimen: Serum – Gel

Reference Range: Supplied with report

Erythropoietin (EPO), a hormone produced by the kidney in response to low renal p02, has an important role in the regulation of erythropoiesis.

Measurement of EPO can be used when differentiating primary polycythaemia (low EPO) from secondary polycythaemia (high EPO).

Reduced EPO formation is a feature of the anaemia of chronic renal disease and EPO can be used to treat the anaemia.

Erythrocyte Sedimentation Rate (ESR)

Specimen: Whole blood – EDTA

Reference Range:

Age	mm/hr	
	female	male
Child 1–18	< 16	< 16
Adult < 50yrs	< 21	< 16
Adult > 50yrs	< 36	< 31
Pregnancy	< 100	

Levels are higher in pregnancy due to hyperfibrinogenaemia.

The ESR is an antique test that still has a place as a nonspecific indicator of inflammatory disease and abnormal protein states.

In an acute illness, the ESR may take a week or more to start to rise and stay elevated for some weeks after its resolution. The CRP, by contrast, rises and falls more quickly and is in some ways a better marker for acute inflammation.

See *C – Reactive Protein (CRP)*

Essential Thrombocythaemia

Essential thrombocythaemia is a myeloproliferative disorder characterised by a sustained increase in the platelet count, particularly above 600 x 109/L.

Clinical features include splenomegaly, an increase in thrombotic risk and bleeding of varying severity. Bone marrow biopsy and elevated NAP score are useful in diagnosis. Treatment is aimed at reducing the platelet count. Therapy at counts which are only modestly increased is controversial.

Ethosuximide (Zarontin)

Specimen: Plasma – Lithium heparin
Trough level is taken before next oral dose (within one hour).
Peak level 2–4 hours post dose.

Reference Range: Supplied with report

Extractable Nuclear Antigens (ENA)

Specimen: Serum – Gel

Reference Range: Supplied with report, qualitative test

Some nuclear antigens can be extracted into solution and patients' serum tested for presence of antibodies against those antigens. ENA screen is used as a follow–up test on positive ANAs or where one of the connective tissue diseases other than rheumatoid arthritis is suspected.

Antigen	Associated disease (with approximate % frequency of antigen)
Sm	SLE (20%)–antigen is 99% specific when present
RNP	MCTD Autoimmune hepatitis SLE (30%) RA
Ro (SS–A)	Sjogren's (90%) usually associated with La (SS–B) SLE (40%) Polymyositis (5%) RA (5%) Neonatal lupus in pregnancy
La (SS–B)	Sjogren's (80%) usually associated with Ro (SS–A) SLE (10%) Diffuse scleroderma Neonatal lupus in pregnancy
Scl 70	Diffuse scleroderma (50%) CREST (10%)
Jo1	Polymyositis (30%)– > 95% specific when present

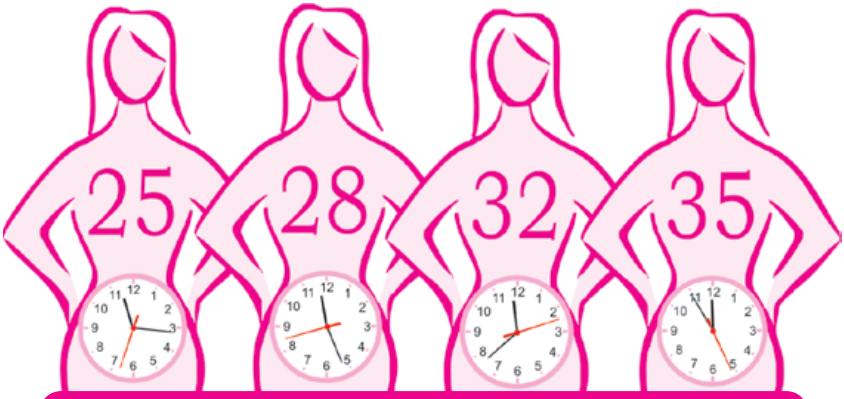
Eye Swabs

If the eye is moist, use a dry swab; if the eye is dry, moisten the swab in transport medium. After pulling the lower lid down roll the swab across the inner part of the lower lid. If there is pus in the corner of the eye, get some of this onto the swab as well.

In neonates, conjunctivitis can be due to gonococci or *Chlamydia trachomatis* transmitted from mother to baby during birth. When testing for Chlamydia remove any purulent exudate before collecting conjunctival epithelial cells by rubbing the small Chlamydia swab over the everted palpebral conjunctiva.

Bacterial conjunctivitis outside the neonatal period is most commonly caused by *Staph. aureus, H. influenzae* or *Strep. pneumoniae* and usually resolves spontaneously or in response to topical eye drops or ointment. Occasionally unusual bacteria are found. A negative bacterial culture usually indicates a viral or allergic conjunctivitis.

A few blood tests (FSH, LH, E2, PROG, QHCG) at the right time, can create life!



Act while your patient's biological clock is still ticking.....

At Canberra Fertility Centre, we care and strive for all patients to take home a baby. We individualise each patient's treatment to ensure they have the best possible chance of success based on their age and fertility status.

This so often can be as simple as Ovulation Tracking, a non-invasive low cost option involving just a few blood tests and pelvic ultrasound scans which pinpoints the time of ovulation to within a few hours.

Ovulation tracking also gives a good indication as to the quality of ovulation, so whether your patient is partnered or single Ovulation Tracking can give your patients great peace of mind.

Tel: (02) 6282 5458

Fax: (02) 6281 2087

Canberra Fertility Centre

IVF & Assisted Conception

EST. 1986

Level 2, Peter Yorke Building, 173 Strickland Crescent Deakin ACT 2600

www.cfc.net.au

Canberra Fertility Centre

IVF & Assisted Conception

Medical Director
Dr Martyn Stafford-Bell

- Infertility investigation
- Natural fertility enhancement
- Male fertility/infertility management
- Assisted reproduction
- IVF/ICSI
  IVF (InVitro Fertilisation)
  ICSI (intracytoplasmic sperm injection)
- Embryo cryopreservation
- Oocyte donation
- Ovulation monitoring
- Hormone evaluation
- Semen evaluation
- Donor sperm insemination
- Infertility counselling
- Surrogacy
- Endocrine evaluation
- Washed sperm insemination
- Sperm banking for pre-vasectomy, chemotherapy or radiotherapy patients and donors

Canberra Specialists

Dr Robert Armellin

A/Prof Steve Robson

Dr Bronwyn Devine

Dr David O'Rourke

Level 2, Peter Yorke Building,
173 Strickland Crescent Deakin ACT 2600

Tel: (02) 6282 5458

Fax: (02) 6281 2087

www.cfc.net.au

F

Factor Assays (Coagulation Factor Assays)

Specimen: Plasma – Sodium Citrate (2 x 4.5 mL)
Sodium Citrate tubes must be filled to capacity.
Spin, separate and freeze plasma.

Reference Range: Supplied with report

See *Coagulation Studies: Factor Assays*

Factor V Leiden (R506Q)

Specimen: Whole blood – EDTA

The most common of the inherited causes of hypercoagulability, first recognised in 1993, is a mutation of the Factor V gene known as Factor V Leiden. Normal Factor V, which promotes normal clotting, is inhibited physiologically by Activated Protein C. Factor V Leiden is abnormally resistant to this inhibitory action and the presence of Factor V Leiden is detected by the APCr test.

Factor V Leiden occurs in 3– 7% of Caucasians and is a factor in 20–40% of cases of venous thromboembolism (VTE). Risk of VTE is increased 5–10 times in heterozygotes compared with normal people and 5–100 times in homozygotes. Addition of oral contraceptives increased the risk a further 3–5 times in heterozygous individuals.

Faeces, for Clostridium Difficile Toxin

Specimen: Faeces
Note any antibiotic therapy.

Reference Range: Not detected

Faeces, for culture and microscopy

Faeces Examination for Microbiology
Specimens should be fresh and submitted contained in the brown top faeces jar. Optimum sensitivity in stool examination is provided by the permanent fixed smear which is routinely performed at Capital Pathology. The standard faeces series is, according to best clinical practice and current HIC tables, two faeces specimens within a seven day period. Microscopy and culture is performed on specimen one and microscopy alone is performed on specimen two.

If a specimen of faeces is unobtainable, a rectal swab may be used (in transport medium). This not adequate for Clostridium difficile toxin detection or for parasites.

Please specify if non–routine examinations are required, (e.g. Rotavirus, Clostridium difficile, Viral culture).

Patient Information leaflets are available from the Client Services Department on 02 6285 9802 or at any Collection Centre.

Faeces, for Occult Blood

Specimen: Special diet required – please contact Collection Centre for information leaflets. Collect samples on three separate days.

Indications
When done properly, a positive result (even one) increases likelihood of gastrointestinal blood loss, especially from lower bowel.

However, because of low sensitivity for detecting small, often intermittent bleeding, a negative result does not exclude GI blood loss as from a tumour. Further evaluation (e.g. colonoscopy) is still therefore indicated in at-risk patients with unexplained iron deficiency.

Types of test/dietary advice

Nonspecific guaiac tests
These, the most commonly used tests, detect the peroxidase activity of haem whether of human or animal origin.

They can give false positives with high meat diets, peroxidase–containing foods such as horseradish or turnip, or sometimes with iron supplements. Gastric irritant such as NSAIDs can give positives because of the minor bleeding they induce.

High dose vitamin C or E supplements can give false negatives.

Specific immunochemical tests
Most of these detect intact human Hb but others are based on human haem, porphyrin or even albumin. Although these tests avoid dietary false positives, there is still the large problem of positives due to minor benign bleeding.

Faeces, for pH and Reducing Substances

Specimen: Faeces
Require same day specimen.

Reference Range: Supplied with report

Faeces, for Rotavirus

Specimen: Faeces

Reference Range: Not detected

Ferritin

Specimen: Serum – Gel

Reference Range:		
	Adult female < 50 years	15–200 ug/L
	Adult female > 50 years	30–300 ug/L
	Adult males	30–300 ug/L

Fibrinogen

Specimen: Plasma – Sodium Citrate

Reference Range: 2.0–4.0 g/L

Fibrinogen is reduced in:

- Afibrinogenaemia (total absence of fibrinogen)
- Hypofibrinogenaemia (decreased synthesis of fibrinogen)
- Dysfibrinogenaemia (structural abnormality of the fibrinogen molecule).

Fibrinogen is increased in inflammatory conditions.

It is measured as part of the basic haemostasis screen.

Filariaria Antibodies

Specimen: Serum – Gel

Reference Range: Supplied with report

Filariasis occurs in Africa, Latin America, Asia and the Pacific Islands where the most common cause is *Wuchereria bancrofti*. Diagnosis is by finding microfilaria in peripheral blood. An eosinophilia may be the only evidence of filarial infestation. Lymphatic filariasis occurs when host inflammation and fibrosis lead to lymphatic occlusion and in patients heavily and repeatedly infected, elephantiasis may develop. Bacterial cellulitis contributes to tissue damage. The syndrome of tropical eosinophilia is an extreme reaction to filariasis.

Treatment with diethyl carbamazine or ivermectin clears microfilaria from the blood. Dying microfilaria can stimulate a severe allergic reaction requiring treatment.

Fine Needle Aspirate (FNA)

FNA is an easy, relatively painless and inexpensive procedure, which should be considered in the diagnostic work–up of any palpable, non–pulsatile mass. The complication rate is low, with local haematoma formation and bruising the most commonly occurring complications.

FNA has high sensitivity (i.e. low false negative rate) and high specificity (i.e. low false positive rate). The specificity of FNA approaches that of frozen section analysis and in experienced hands, has a false positive rate of less than 0.5%.

The most common site for FNA is the breast, but thyroid nodules, head and neck lumps, enlarged lymph nodes and soft tissue masses can all be aspirated.

The procedure will be performed by one of Capital Pathology's experienced cytopathologists. Alternatively, specimens obtained by the referring doctor can be sent to the laboratory via the courier for processing.

Appointments
Please phone the Cytology Department to arrange a patient appointment on 02 6285 9867. A patient information sheet is available from the Client Services Department on 02 6285 9802, and this also has a map to direct the patient to the laboratory.

Procedure
FNA is performed without anaesthesia using a 23 or 25 gauge needle. Negative pressure is sometimes applied by retracting the plunger of an attached syringe while moving the needle backwards and forwards within the lesion.

Cytological assessment is optimised by preparing both wet-fixed and air-dried smears. Wet-fixed slides should be fixed as rapidly as possible using spray fixative or by immersion in 95% alcohol for 20 minutes. The slides then can be air-dried and placed in a slide mailer. When making air dried smears, complete and rapid drying is necessary before transportation. A hair dryer on low setting can be used. Slide smearing techniques can be discussed with the pathologists. It is necessary to write on the frosted end of the slide whether that particular slide has been wet-fixed (WF) or air-dried (AD), as the laboratory staining for each of these techniques is different.

The needle should be washed out using RPMI. The washings are submitted for preparation of a cell block. Two or more aspirations will generally result in a greater yield of cellular material.

A separate pass transferred into RPMI is advisable when investigating enlarged lymph nodes or unusual masses. This allows for lymphocyte marker studies by flow cytometry or for immunohistochemical staining,used in the investigation or tumours of uncertain lineage.

Smear preparation

For non–gynaecological and fine needle aspiration specimens (see diagrams)

The simplest way to make a smear is the “pull apart method”. This is particularly useful for bloody specimens and cyst fluids:

A small drop of fluid is placed on a clean glass near the label end. A second slide is placed parallel over the first with the label end of the second slide at the opposite end. Let the fluid start to spread between the two slides and then gently pull them apart The specimen usually distributes relatively evenly between the two slides.

The second method is useful for thick fluids or aspirates. Much of the specimen remains on the test slide, leaving the spreading slide relatively clean. All slides should be sent to the laboratory.

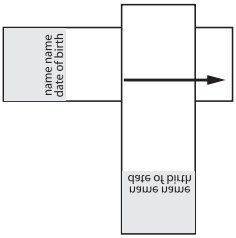
Fix rapidly, by spraying with pump fixative or dropping into 95% alcohol, or by rapidly air drying. Both wet–fixed and air–dried slides are useful, so it is preferable to use both methods if the amount of material is sufficient. However it is important to label the slides as to which method has been used.

Please ensure that the slides are labelled with pencil on the frosted end, with the patient’s full name and date of birth.

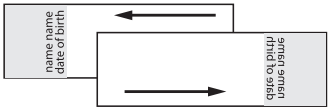
F

Slide preparation diagrams

Right angle
for thick material



Pull apart
for bloody or cystic material



To discuss any matter related to FNA with a pathologist, please phone 02 6285 9867. Results are usually available the following day.

An FNA Collection Procedure Sheet is available from the Client Services on 02 6285 9802 on request.

Specimen Type	Comment	Test Required			
		Cytology of solid lesion	Cytology of fluids	For suspected lymphoma	Microbiology/ culture
Direct Smears **Air dried (Romanowsky stain)** **Spray fixed (Pap stain for nuclei)**	**Made at the time of FNA – BOTH methods of preparation preferred for a single episode.** Hair dryer helpful. Alcohol spray fixative.				
RMPI-FCS **For suspected lymphomas** Available from laboratory.	A separate pass into the RPMI is recommended as it gives more material **Flow cytometry** helpful in classifying NHL Cell blocks helpful especially if primary tumour unknown or to prove a metastasis from a known tumour. Immunohistochemistry for tumour characterisation possible.	Cell blocks – if primary tumour unknown or to prove metastasis from unknown tumour. Immunohistochemistry for tumour characterisation possible.		**Flow cytometry** helpful in classifying NHL	
Yellow top jar					Contact laboratory if special tests required

Flow Cytometry

Specimen: Whole blood – EDTA x 2

This may also be performed on fresh, unfixed lymphoid or other tissue removed surgically or by fine needle aspiration. Blood or bone marrow aspirate may be submitted to flow cytometry.

This test is used for the classification of lymphomas and leukaemias and to establish a diagnosis of reactive or neoplastic lymphoid proliferations.

The report will include quantitation of cell lines identified.

See *Lymphocyte Markers*

Flucytosine (Ancotil)

Specimen: Plasma – Lithium Heparin
Sample just before next dose and 2 hours after oral dose (or 30 minutes after IV dose).

Reference Range: Supplied with report

Fluid Examination

Body fluid
Specimens should be transported in a yellow top jar or as a swab in transport medium. Depending on the clinical circumstances individual requests should specify which tests are required e.g. microbiology, cytology or biochemistry (protein and glucose).

Cerebrospinal fluid
See *Cerebrospinal Fluid (CSF)*

Peritoneal dialysis
Send entire bag of peritoneal dialysate. Usually one bag per 24 hours is required.

Pus
Where sufficient pus is available, aspirate with a sterile syringe fitted to a broad gauge needle. Inoculate some of the pus into a blood culture bottle and leave the remainder in the syringe. Remove air from the syringe and carefully remove the needle and discard appropriately. Cap syringe and send to the laboratory.

Synovial fluid
For full synovial fluid analysis the specimen should be apportioned as follows:

1 EDTA for cell count. Collect 2 mL, mix immediately and thoroughly.
1 sterile specimen container for crystals, gram stain, culture, and urate level.

Folate, red cell

Specimen: Whole blood – EDTA

Reference Range: > 1000 nmol/L

Red cell folate is a better indicator of tissue stores than serum folate, which is more labile.

Interrelationship between B12 and folate deficiencies:

1° deficiency	Serum B12	Serum folate	Red cell folate
B12	↓↓	n ↑	n ↓
Folate	n ↓	↓↓	↓

Follicle Stimulating Hormone (FSH)

Specimen: Serum – Gel

Reference Ranges:

Adult female	FSH IU/L
Follicular and luteal phases	2– 23
Mid–cycle ovulatory peak	11– 30
Post–menopausal	21– 175
Adult male	**FSH IU/L**
	1.0– 14

Applications

Identifying the menopause – the FSH rises, FSH rises relatively more than LH.

Ovarian or testicular failure – both FSH and LH rise to menopausal levels. Gonadal failure can be primary as in agenesis, dysgenesis, Klinefelter's or Turner's syndromes. Or it can be secondary as in premature menopause, oophorectomy or testicular damage.

Identifying day of ovulation–in fertility control.

Hypopituitarism – LH and FSH are not usually undetectable in hypopituitarism but low or inappropriately normal values in patients with low oestradiol (women) or low testosterone (men) are suggestive. Mid-range or high values can help exclude hypopituitarism.

Polycystic Ovary Syndrome–LH is often raised relative to FSH with an LH/FSH ratio > 2.

Fructosamine

Specimen: Serum – Gel

Fructosamine is useful in the monitoring of diabetes mellitus and the assessment of overall control of blood glucose over the one to three weeks prior to collecting the specimen. Increased values are suggestive of significant persistent hyperglycaemia. Levels are lower in the presence of hypoalbuminaemia. It is useful together with the assay of haemoglobin A1C (glycated haemoglobin) which reflects diabetic control over a longer period.

See *HbA1c*

FTA (Fluorescent Treponemal Antibody)

Specimen: Serum – Gel

Reference Range: Not detected

A positive FTA indicates past or present treponemal infection. It does not distinguish between syphilis and yaws. The FTA stays positive life–long.

See *Syphilis*

Fungal Investigation

Skin lesions – take a scraping of the advancing edge.

Nails – nails should be cut back. Collect clippings or scrapings, including necrotic debris from beneath the nail.

Hair – include hair roots.

Collect specimen into a yellow top jar. The more material submitted the greater the odds of positive findings.

Microscopy is reported within 24 hours. Cultures are maintained and examined on a regular basis for 2–4 weeks.

G

Galactorrhoea

Most patients are women but occasionally galactorrhoea is found in men.

Causes are:

- ***Hyperprolactinaemia***
  This is commonly, but not invariably, associated with galactorrhoea. There may or may not be a pituitary tumour. Elevations of prolactin can be minimal and intermittent.
- ***Hypothyroidism and hyperthyroidism***
- ***Drugs***
  phenothiazines, benzodiazepines, tricyclics, methyldopa, butyrophenones, metoclopramide, H2 receptor antagonists, digoxin, spironolactone, post–oral contraceptive.

Galactose

Specimen:	Random urine Specimen needs to be frozen.
Reference Range:	Supplied with report

Gallstones

These consist mainly of cholesterol except in chronic haemolysis when pigment (bilirubin) stones may be formed. Analysis of a stone is of interest only when haemolysis is suspected or when an unidentified object ?gallstone, is found in faeces.

Gamma Glutamyl Transpeptidase (GGT)

Specimen:	Serum – Gel	
Reference Range:	Adult female	5–35 U/L
	Adult male	5–50 U/L

GGT is found almost entirely in the liver. It is elevated particularly in cholestatic disorders, by alcohol, and also as an effect of some drugs, notably anticonvulsants.

GGT is the enzyme most reliably raised by excessive alcohol, typically in the range 60–200 but occasionally > 1000. 30% of heavy drinkers have normal levels. After a weekend binge, levels rise up to 100% over 3 days. If the cause of an elevated GGT is uncertain, total abstinence from alcohol with weekly GGT estimations over a 4–week period may provide the answer.

See *Liver Function Test / Interpretation*

Gardnerella Vaginalis

Formerly known as Haemophilus vaginalis and Corynebacterium vaginalis.

A common bacterial isolate from the vagina. It may be present in up to 30% of healthy women. When found in association with “non–specific vaginosis”, treatment with metronidazole or tinidazole will eliminate odour and discharge though there is a tendency to recurrence requiring repeat treatment.

See *Vaginal Swabs for Discharge*

Gastrin

Specimen: Serum – Gel
Patient should be fasting.
Spin, separate and freeze immediately.

Reference Range: Supplied with report

Gastrin, released from the gastric mucosa, is a potent stimulant of gastric acid secretion.

Gastrin measurements are used in the diagnosis of pancreatic gastrinomas associated with severe peptic ulceration in the Zollinger–Ellison syndrome in which there is marked hypersecretion of gastric acid.

Vagotomy, non–fasting state, renal failure, or the gastric acid hyposecretion due to pernicious anaemia and chronic atrophic gastritis, are associated with an increase in gastrin levels.

Gentamicin

Specimen: Serum – Gel

If Gentamicin is given as a single daily dose a measurement of plasma concentration should be made between 6–14 hours after the end of the infusion. For intermittent dosing regimes, peak or trough levels may be measured.

Gentamicin dose adjustment is required to avoid the effects of gentamicin toxicity.

Monitoring of renal function – renal function should be checked at regular intervals during gentamicin treatment.

Specialist advice may be required for monitoring and assessment of gentamicin levels.

Giardia

Specimen: 1–2 random faeces samples

Giardia lamblia is a protozoan infesting the upper small bowel where it can cause acute or chronic diarrhoea and sometimes fat and vitamin malabsorption.

It is commonly taught that three stool specimens are required to detect intestinal parasites such as Giardia but several studies have shown that > 85% of cases are detected on the first specimen. When evaluating chronic or relapsing diarrhoea it may be necessary to send multiple specimens for testing.

The two commonly–used tests for Giardia:

- Detection of Giardia antigen in faeces using an Elisa method.
- Microscopy for cysts in faecal concentrate. Occasionally trophozoites are seen in acute infections.

The antigen method is the more sensitive.

Treatment with tinidazole or metronidazole is usually effective but may need to be repeated.

Gilbert's Syndrome

This common, harmless, inherited condition, in which the liver has reduced ability to conjugate bilirubin, was first described in 1901 by the Frenchman Gilbert whose name is usually, though not always, pronounced in the English fashion.

It affects somewhere between 2 and 10% of the male population depending whether the cut–off is taken as 25 or 20 umol/L. The male/female ratio is about 4:1.

Because a genetic test is not available, diagnosis is based on exclusion.

Features are:

- Elevation of bilirubin is the only laboratory abnormality and the level seldom exceeds 80 ug/L
- Liver enzymes are normal
- The bilirubin is unconjugated
- There is no evidence of haemolysis as measured by reticulocytes and haptoglobins
- The bilirubin level fluctuates; the condition is often first detected in the 2nd–3rd decade
- Fasting, dehydration, fever, sea–sickness, intercurrent illnesses, can all cause the bilirubin to rise, sometimes to the point where it becomes visible as jaundice – at this point the patient may note malaise, blaming it on the Gilbert's rather than the precipitating factors.

Gliadin (gluten) Antibodies (AGA)

Specimen:	Serum – Gel
Reference Range:	Supplied with report
See	*Coeliac Disease*

Glomerular Basement Membrane Antibodies

Specimen:	Serum – Gel
Reference Range:	Not detected

Glucose

Specimen: Plasma (Fluoride Oxalate) is ideal, or serum if cells can be separated within 2 hours

Reference Range:

	Age	mmol/L
Fasting glucose	All	3.4– 6.0
Random glucose	Day 1	1.7– 5.6
	Day 1– 2 yr	2.5– 5.6
	> 2yr	3.4– 7.7

If the specimen is collected in a plain tube the glucose level will tend to be low by an unpredictable amount because red cells consume glucose, causing the serum level to fall. Fluoride prevents this glycolysis.

Diagnostic Criteria For Diabetes

Single fasting and symptoms > 7.0 mmol/L or

2 hours post prandial or casual post prandial and symptoms > 11.1 mmol/L

If no symptoms or equivocal symptoms:

At least one additional glucose measurement (preferably fasting) on a different day with a value in the diabetic range is necessary to confirm the diagnosis.

See *Glucose Tolerance Test*
Hypoglycaemia

Glucose Tolerance Test (GTT)

Two hour 75g oral GTT

Procedure

The oral carbohydrate intake should be at least 150 g on each of the three days immediately preceding the test. Carbohydrate restriction to less than 125 g daily may impair glucose tolerance.

All glucose tolerance tests should be scheduled for the morning. Glucose tolerance decreases in the afternoon and evening.

The patient must fast for 12 hours prior to the test. Water and usual medications only are permitted throughout this period and during the test.

75 g of glucose (available in liquid form from the laboratory) should be ingested within 5–10 minutes. There is a dose adjustment for children/ adolescents of 1.75 g of glucose per kilogram of bodyweight to a maximum of 75 g. A GTT may not be necessary in children – please contact the pathologist for advice if unsure.

1 x 5 mL fluoride oxalate whole blood is to be collected before ingestion of the glucose load and at one hour and two hours post glucose load.

This procedure is performed by appointment at a Collection Centre.
Patient Information leaflets are available on our web site, from Stores Department on 02 6285 9813 or your nearest Collection Centre.

Oral glucose tolerance test interpretation

(75 gram load, adult, non pregnant)

1. Diabetes Mellitus
   Fasting > 7.0 mmol/L and
   2 hour glucose > 11.1 mmol/L
   If only one value is in diabetic range, then diagnosis should be confirmed by repeat on another day, unless there is unequivocal hyperglycaemia with acute metabolic decompensation or obvious symptoms. The diagnosis of diabetes must be definite.
2. Impaired Glucose Tolerance
   Fasting < 7.0 mmol/L and
   2 hour glucose 7.8–11.0 mmol/L
3. Impaired Fasting Glucose Tolerance
   Fasting 6.1–6.9 mmol/L and
   2 hour glucose normal

Gestational diabetes mellitus (GDM)

GDM is a form of glucose intolerance that develops during and disappears after parturition.

It is associated with increased fetal risk which is reduced if the GDM is treated successfully with diet or, in the few where hyperglycaemia persists, with insulin. A mother with GDM is at increased risk for developing diabetes in later life.

Screening test for GDM

Screening at 26–28 weeks is currently recommended for all pregnant women, though clearly at–risk patients, e.g. previous GDM, can be evaluated earlier, even before conception.

The test is usually done in the morning. No booking is required and the patient does not need to fast before the tests. A suspension of 50 g of glucose is given and a single blood collected one hour later. If the one hour glucose is equal to or above 7.8 mmol/L, the test is positive and a 2–hour 75 g GTT is ordered as the definitive test.

Diagnostic criteria for GDM (2–hour 75 g OGTT)

GDM is present if:

Fasting glucose > 5.5 mmol/l
or 2–hour glucose > 8.0mmol/

Specialized GTTs

Protocols are available for extended GTTs (3,4,5 hour GTT) to investigate hypoglycaemia.

GTTs with insulin levels can be performed to investigate insulin resistance. Kidson Protocol (HOMA) index GTTs can be arranged on request.

Reference:

1. Colman PG, Thomas DW, Zimmet PZ, et al. New classification and criteria for diagnosis of diabetes mellitus. MJA 1999; 170:375–378.
2. Hoffman L, Nolan C, Wilson JD, et al. Gestational diabetes mellitus–management guidelines. The Australian Diabetes in Pregnancy Society. MJA 1998; 169:93–97.`

Glycosuria

Glucose appears in the urine when the renal threshold has been exceeded; this typically being at a blood glucose level around 11 mmol/L.

With advancing age, or renal impairment, the renal threshold rises so that some diabetics have glucose–free urine even with blood glucose levels in the 12–15 mmol/L range.

Glycosuria is found in:

- Diabetes mellitus – once the renal threshold is exceeded.
- Renal glycosuria – in which the individual has a lowered renal threshold. The GTT is entirely normal but urine specimens collected during the test show glucose. The patient who is proven to have this uncommon condition can be reassured that it is of no clinical significance and is unrelated to diabetes.
- In pregnancy – renal glycosuria is common in healthy non–diabetic women during pregnancy, beginning during the first trimester and disappearing within a week of delivery. A feature of this glycosuria is its variability from day to day, throughout the course of the pregnancy and even during the course of a day.

Gold, heavy metal

Specimen:	Urine – 24-hour urine in acid washed bottle Plasma – EDTA tube. Plasma should be spun and separated.
Reference Range:	Supplied with report

Gonorrhoeae

Please see Neisseria gonorrhoeae.

Gram Stain

The Gram stain, described as the single most useful procedure in diagnostic microbiology, was perfected by the Danish microbiologist Christian Gram in 1884. When a microbiological specimen reaches the laboratory there are typically two initial procedures:

- Gram stain of the specimen smeared on a slide to determine numbers of bacteria, whether cocci or bacilli and whether gram–negative or gram–positive. In a case of meningitis, for example, the gram stain gives a presumptive diagnosis and dictates initial antibiotic treatment.
- The specimen is cultured on a microbiological plate and the colonies gram–stained next day.

Some examples:

Gram–positive organisms

cocci	Staphylococcus	
	Streptococcus	
bacilli	Corynebacterium	

Gram–negative organisms

cocci	Neisseri	
bacilli	enteric bacteria:	Escherichia, Salmonella, Shigella, Enterobacter, Klebsiella, Serratia, Proteus, Citrobacter, Yersinia

Growth Hormone

Specimen: Serum – Gel

Reference Range: The normal values vary with age and sex

Suppression or stimulation tests are used in preference to random measurements

Elevated levels

- Pituitary gigantism
- Acromegaly
- Stress, exercise.

Low levels

- Pituitary dwarfism
- Hypopituitarism
- Glucose infusion – used in suppression tests for gigantism or acromegaly.

Gynaecomastia

Transient gynaecomastia is normal in the newborn and in adolescence and minor degrees of persistent gynaecomastia are common in the elderly male as testosterone levels decline with an increase in the oestrogen/testosterone ratio.

The causes of more troublesome and progressive breast enlargement include:

- Drugs oestrogens spironolactone
- Digoxin cimetidine
- tricyclics diazepam
- methyldopa antiandrogens
- Hypogonadism – check LH, FSH, testosterone
- Liver disease
- Thyrotoxicosis
- Feminising tumours
- Klinefelter's syndrome.

H

Haemochromatosis

Specimen: Whole blood – EDTA

Reference Range: Supplied with report

Haemochromatosis is a condition of iron overload which is usually subclinical but which in some patients causes systemic disease due to parenchymal tissue damage.

In its common hereditary form, the diagnosis of haemochromatosis is being made with increasing frequency because of the ready availability of tests for ferritin, iron and iron saturation and recently, the HFE gene.

Iron overload can also be acquired due to repeated transfusions for refractory anaemia or to inappropriate parenteral iron over a long period of time.

Hereditary haemochromatosis (HH)
In HH the iron overload is due to excessive absorption which steadily increases the total body mass of iron throughout life. Women are protected during child–bearing years by menstrual loss of iron.

HH is carried on a recessive gene with the remarkably high prevalence of 1:10 in the general population for the heterozygous (carrier) state and 1:400 for the homozygous (affected) state.

In 1996 the HFE gene, also known as the cys282 tyr mutation, was identified as a marker for at least 90% of HH.

Clinically HH is characterised by:

- Lethargy, skin pigmentation, reduced sweating
- Hepatic damage: elevated enzymes, cirrhosis, hepatocellular carcinoma
- Endocrine damage: diabetes, hypogonadism, loss of libido, amenorrhoea, hypopituitarism, hypothyroidism
- Arthropathy
- Myocardial damage: congestive cardiac failure.

Diagnosis
HH is suspected when iron saturation is consistently above 45–55% and ferritin above 400 ug/L.

The HFE gene should be tested for and if present the test will show whether the patient is a hetero–or homozygote.

The molecular testing currently looks for three gene mutations:
C282Y, H63D and S65C.

Haemoglobin Electrophoresis (Hb EPG)

Specimen: Whole blood – EDTA, and Serum – Gel

Hb EPG detects abnormal and unstable haemoglobins and gives an indication whether HbA2 or HbF are increased.

Haemoglobinopathy Screen

The screen consists of: Hb electrophoresisHbA2 quantitation
HbF quantitation
HbH body stain
Hb stability test.

Indications

- Suspected thalassaemia or haemoglobinopathy – hypochromic microcytic anaemia in the absence of iron deficiency or chronic disease, particularly in persons of Asian or Mediterranean descent.
- Unexplained haemolytic anaemia.
- The patient's partner has been diagnosed with a haemoglobinopathy or thalassaemia and the couple are in the child–bearing age group.

See *Haemoglobins, normal and abnormal*

Haemoglobins, Normal and Abnormal

The haemoglobin (Hb) molecule consists of two pairs of polypeptide chains with a haem attached to each. The porphyrias are due to disorders of haem synthesis. The haemoglobinopathies are due to disorders in the globin chain pairs which are labelled á â ä ã etc. In the haemoglobinopathies, the amino acid sequence in a globin chain can be abnormal, or there can be a quantitative abnormality in globin chain production.

The main haemoglobins are:

HbA á $_2$â$_2$	The normal adult Hb comprising 97% of the total. Also called HbA1, HbA1C, of interest to diabetologists, is the non–enzymatically glycated fraction of HbA1.
HbF á $_2$ã$_2$	Fetal Hb comprises 70–90% of the total at birth falling to 1% by age 2. It is increased in beta thalassaemia and hereditary persistence of HbF (HPFH).
HbA$_2$ á $_2$ä$_2$	The minor component of Hb comprising 1.5–3.2% of the total. It is increased in beta cell thalassaemia.
HbH beta$_2$ beta$_2$	Is increased in the α thalassaemias in which there is deletion of 1–4 of the ã chains. In the 3–chain deletion HbH disease, HbH comprises 5–30% of total Hb.
HbS	The cause of sickle cell anaemia (HbSS) found in South Africans and occasionally in those of Mediterranean or Middle Eastern origin. The heterozygous stat HbAS, is asymptomatic.

See *Haemoglobinopathy Screen*
Thalassaemias
Sickle Cell Test

Haemolysis

The features of haemolysis are:

- Anaemia
- Polychromasia–reticulocytosis
- Haptoglobins–reduced or absent
- Hyperbilirubinaemia.

The blood film is used to distinguish between spherocytic and non–spherocytic haemolytic anaemia:

Spherocytes present	Spherocytes absent
Hereditary spherocytosis	Hereditary elliptocytosis
Autoimmune haemolysis	Haemoglobinopathies
(Coomb's positive)	Enzymopathies
Drug–induced haemolysis	(e.g. G–6–PD deficiency)
Delayed transfusion reaction	PNH
ABO incompatibility in the newborn	

Haemophilia

The haemophilias are due either to Factor VIII deficiency (classical haemophilia) or Factor IX deficiency (Christmas disease). Spontaneous mutations account for 30% of new cases in the absence of a family history. Severe haemophilia presents early in infancy but mild haemophilia can present later in life following surgery or trauma.

Diagnosis is from:

- Clinical bleeding history
- Family history
- Abnormal APTT (correcting with mixing test)
- Reduced level of FVIII or FIX
- DNA genotyping.

See *Coagulation Studies*

Haemosiderin in Urine

A granular storage form of iron which can be seen histologically. Physiologically it is the normal storage form of iron in bone marrow. Patholgically it is found in many parenchymal tissues in iron–overload. S. ferritin levels reflect tissue haemosiderin load.

In chronic haemoglobinuria, haemosiderin may be detected in a fresh random urine.

Hairy Cell Leukaemia

This is a rare form of B cell leukaemia (1–2%) with a 5:1 male predominance. It is characterised by pancytopaenia, circulating "hairy cells" and splenomegaly (80%). Hairy cells may be infrequent in the peripheral blood, and the characteristic bone marrow fibrosis usually leads to a dry tap at bone marrow aspiration. Treatment with chemotherapy is highly effective, with a response and remission observed in the majority of patients.

Haptoglobins

Specimen: Serum – Gel

Reference Range: Supplied with report

Haptoglobin estimation is one of the screening tests for haemolysis. Hb released by the haemolytic process combines with haptoglobin to form a complex which is removed by the liver. It is also an acute phase reactant, migrating with the alpha 2 band on protein electrophoresis.

It is reduced by:

- Haemolysis – haptoglobins are absent in moderate or severe haemolysis
- Oestrogens, oral contraceptives, pregnancy
- Acute or chronic hepatocellular liver disease
- Specific genotype
- Lower in first year of life.

It is elevated by:

- Biliary obstruction
- Many acute or chronic inflammatory disorders–following recovery from an infective episode the level falls to normal within 10–14 days
- Specific genotype.

HbA1c

Specimen: Whole blood – EDTA

Reference Ranges: 4.3–6.4% reference population

Clinical Evaluation

- 6.1–7.0% HbA1c indicates very good control.
- 7.1–8.0% HbA1c indicates adequate control.
- 8.1–9.0% HbA1c indicates suboptimal control.

{I Reference: ADA Position Statement. Diabetic Care 20 (Supp 1), 55–511, 1997 and RCPA–AACB Chemical Pathology QA Program, 2001}

Suggest continue monitoring diabetic control with HbA1c measurements at three monthly intervals.

Heavy Metal Testing

Specimen: Preferred speciman is 24-hour plain urine
Lead testing requires Whole blood – EDTA.
All other heavy metals can be done on whole blood – Trace element tube.
It is recommended to specify individual heavy metals required.
A screen includes aluminium, arsenic, cadmium, cobalt, lead, and mercury.

Reference Range: Supplied with report

Helicobacter pylori

Diagnosis

Histology of gastric biopsies has long been the mainstay of diagnosis of *H. pylori* related disease. Serological diagnosis remains controversial, and while levels of IgG do fall after successful eradication treatment, post–treatment monitoring can take six months or longer to show a conclusive drop. The urea breath test is accepted as a simple, reliable and safe diagnostic alternative.

Patient Preparation

This procedure is performed by appointment at a Collection Centre.

The patient should have nil by mouth for six hours.

The following drugs may effect the urea breath test:

Drugs to be stopped at least FOUR weeks prior to testing:

- All antibiotics (e.g. Amoxycillin, Tetracyclines, Metronidazole)
- Bismuth–containing compounds (e.g. Denol).

Drugs to be stopped at least ONE week prior to testing:

- Proton–pump inhibitors (e.g. Omerprazole, Lanzaprazole, Pantoprazole).

Patient information leaflets are available at Collection Centres or from the Client Services Department on 02 6285 9802 or from www.capitalpath.com.au.

Hepatitis

Hepatitis is characterised by elevations of ALT and AST and a histological picture on biopsy which may be diagnostic. When ALT and AST are above 1000 U/L with ALP and GGT below 300 U/L, the cause will usually be viral infection. In chronic hepatitis, changes in ALT are used as an indication of disease activity.

Main causes of hepatitis include:

- Viral infection
  - Hepatitis A virus (HAV)
  - Hepatitis B virus (HBV)
  - Hepatitis C virus (HCV)
  - EB virus (infectious mononucleosis)

    Other viruses
    - HIV
    - Hepatitis D, E
    - CMV (cytomegalovirus)
    - Coxsackie virus
    - Sundry other viruses causing flu–like illnesses circulating in the community
- Autoimmune hepatitis
- Alcoholic hepatitis
- Drug associated hepatitis
- Other.

Hepatitis A Antibody (HAV)

Specimen:	Serum – Gel
Tests:	Anti–HAV IgM
	Anti–HAV IgM and IgG (total antibody)

There is no test for HAV antigen.

Interpretation

The IgM antibody appears at about the time clinical symptoms develop and persists for 6–12 months. This anti–HAV IgM is the marker for current or recent infection. The IgG appears not long after the IgM but persists for life and is thus the marker for HAV immune status.

Hepatitis B (HBV)

Specimen:	Serum – Gel	
Tests:	HBs Ag	(HBV surface antigen)
	HBe Ag	(HBV e antigen)
	Anti–HBs	(antibody to HBV surface antigen)
	Anti–Hbe	(antibody to HB e antigen)
	Anti–HBc IgM	(IgM antibody to HBV core antigen)
	Anti–HBc	(IgM and IgG antibody to HBV core)
	HBV–DNA	(HBV–DNA detected by PCR)

Indications and interpretation

1. HBs antigen
If positive indicates one of:
– Acute HBV infection (enzymes elevated) or
– Chronic HBV infection (enzymes fluctuate) or
– HBV carrier (enzymes not elevated)

2. HBe antigen
This should be checked whenever HBs antigen is positive. If Hbe antigen (Ag) is present, it indicates greater infectivity of the HBV infection towards sexual partner or fetus and greater likelihood of developing chronic hepatitis. Hbe Ag is found only in association with Hbs Ag, never on its own.

3. Anti–HBs
Indicates immunity to HBV whether naturally acquired or following immunisation. Anti–HBs can be measured quantitatively–levels above 10 IU/L indicate immunity.

4. Anti–HBc IgM
Is measured when current HBV infection is strongly suspected but HBs Ag and anti–HBs are both negative. Anti–HBc IgM rises early in HBV infection and persists for about 6 months. It fills the one month “window” between disappearance of HBs Ag and the appearance of anti–HBs. Positive anti–HBc does not confer immunity.

5. Anti–HBc total
Indicates past infection but unlike anti–HBs it does not indicate immunity and can coexist with HBs Ag in carriers or chronic hepatitis. Anti–HBc remains negative after HBV vaccination because the vaccine contains surface but not core antigen.

6. Anti–HBe
In chronic HBV infection, anti–BHe indicates low infectivity.

7. Anti–HBV– DNA
If positive indicated HBV viraemia which is an indication for considering interferon treatment in chronic infections.

Hepatitis C (HCV)

Specimen:	Serum – Gel
Tests:	Anti–HCV (total antibody to HCV)
	HCV–RNA)

Indications and Interpretation

1. Anti–HCV

Used when hepatitis C is a possibility, e.g. IV drug users, raised liver enzymes of unknown cause. Anti–HCV starts to rise 1–6 months after the initial infection and persists for life. Presence of anti–HCV indicates either past infection or continuing chronic infection.

2. HCV–RNA

This is a follow–on test when anti–HCV is positive. It is also used when clinical suspicion of HCV infection is high but anti–HCV is negative because infection is recent or the patient immunosuppressed.

It indicates current active HCV infection and is an indication for considering interferon and ribavirin therapy.

Clinical course and epidemiology

HCV was identified in 1989 having previously been included in the non–A non–B hepatitis category. A random population sample in the United States shows 1.8% positivity.

Transmission in blood products has been largely eliminated, leaving contaminated needles in drug users as the main source of new infections. It is the cause of 5–15% of acute non–A, non–B hepatitis.

Mother–to–fetus transmission occurs but is infrequent, < 5%. Sexual transmission is believed to be uncommon.

More than 50% of HCV infections become chronic with potential to develop cirrhosis and hepatocellular carcinoma.

Treatment with a combination of interferon and ribavirin clears the virus in about 50% of patients.

There is no vaccine yet.

Follow–up on HCV +ve patients

A positive HCV Ab test should be followed by HCV RNA test and ALT.

If HCV RNA is positive, the patient should be referred for consideration of antiviral therapy.

An ALT > 60 units indicates active disease but a level < 60 does not exclude activity.

The patient who is HCV Ab +ve but HCV RNA –ve is presumed to be free of active hepatitis provided the ALT is < 60. The ALT should be monitored annually and the HCV RNA for at least 2 years to confirm it is staying negative.

Hepatitis D (HDV–Delta)

Specimen:	Serum – Gel
Tests:	Anti–Delta IgG
	HDV RNA

HDV infections are found only as a co–infection or superinfection in HBV positive patients. HDV is associated with more severe acute disease and a greater risk of developing chronic hepatitis and its complications.

HDV infections once established, will persist for as long as HBV carriage continues, but if HBV is eliminated HDV will go with it.

Anti–delta antibodies indicate either current active infection or past infection with immunity.

Positive HDV RNA indicates current active infection.

Hepatitis E (HEV)

Specimen:	Serum – Gel
Tests:	Anti–HEV IgG

Hepatitis E resembles hepatitis A in its faecal–oral mode of transmission. It is endemic in India, SE Asia, China, Mexico and parts of Africa.

Presence of HEV antibodies does not distinguish current from past infection.

Identifying the virus in a small specimen of faeces may identify current infection.

Hepatitis, Autoimmune

Specimen:	Serum – Gel
Tests:	Immunoglobulins
	Antimitochondrial antibodies
	Liver/kidney microsomal antibodies
	ANA
	Smooth muscle antibodies
	Anti–ds DNA

Autoimmune hepatitis, formerly known as chronic active hepatitis and before that lupoid hepatitis, affects predominantly young women. The hepatitis can be of all grades of severity ranging from minor enzyme elevations in a well person to a clinical laboratory picture like that of severe viral hepatitis. Diagnosis is by excluding viral causes; by finding specific autoantibodies; by markedly elevated IgG; and by liver biopsy.

Smooth muscle antibodies (SMA) in high concentration (> 80) define autoimmune hepatitis.

Antimitochondrial antibodies (AMA) and total IgM may be elevated but these are typically the markers of the other autoimmune liver disease, primary biliary cirrhosis. Liver/kidney microsomal antibodies (LKM) are uncommon but help in disease classification. Titres tend to be lower in viral hepatitis than in autoimmune hepatitis.

Hereditary Spherocytosis

Specimen: Whole blood – EDTA

A relatively common haemolytic disorder found in all races and typically inherited as an autosomal dominant. In the 25% of individuals who have unaffected parents, the disease is assumed to be due to autosomal recessive inheritance or a spontaneous mutation.

More severe forms present in the first 10 years of life but milder forms may be discovered incidentally either on a blood count or because of a minimally elevated bilirubin resembling Gilbert's syndrome.

Laboratory tests typically show the features of a haemolytic disorder – increased reticulocytes, reduced haptoglobins, hyperbilirubinaemia, anaemia.

The more specific findings are:

- Spherocytes on the blood film
- Increased osmotic fragility of red cells.

Herpes Simplex Virus (HSV)

PCR (preferred)

Specimen: Fluid or vesicle scrape
Collected in a sterile container, a viral swab, or plain swab.

Immunofluorescence and culture is being superceded by PCR.

There are two types of herpes simplex, HSV1 and HSV2.

HSV1 commonly gives rise to recurrent oral infections ("cold sores") and HSV2 to recurrent genital infections but either can be found at either site.

Herpes Type 6 (Roseola infantum)

Specimen: Serum – Gel

Reference Range: Supplied with report

HIAA (5–Hydroxy Indole Acetic Acid)

Specimen: 24-hour urine (HCL preservative)

Reference Range: Supplied with report

Avoid the following drugs and foods for 3 days before collecting the urine:

- Phenothiazines
- Methyldopa
- Naproxen
- Cough mixture
- Acetaminophen
- Bananas
- Plums
- Pineapple
- Fruit juices
- Kiwifruit
- Tomatoes
- Eggplant
- Avocado
- Walnuts
- Pecans.

In carcinoid syndrome the HIAA is usually elevated. Carcinoid syndrome, due to release of serotonin from a carcinoid tumour, is characterised by cutaneous flushing, diarrhoea, valvular heart disease, and less often, by wheezing, paroxysmal

hypotension and telangiectasia. HIAA is the breakdown product of serotonin and is usually measured in response to a history of flushing though this is more commonly due to menopause, alcohol (particularly in combination with a sulphonylurea or disulfiram). Phaeochromocytoma is infrequently a cause of flushing.

Histopathology

Specimens for routine histopathology should be placed promptly in formalin fixative (10% buffered formalin) after surgical removal.

Tissue specimen containers with fixative added are suitable for most biopsies and small excision specimens. Other containers are available for larger specimens. Ideally, the container should hold a volume of fixative at least 10 times that of the specimen.

Tissue for the following investigations **should not be placed in formalin:**

- Frozen section
- Liver iron estimation
- Microbiological investigation
- Flow cytometry
- Disaccharidase assay on small bowel biopsy
- Immunofluorescence
- Chromosome studies
- Crystal identification
- Testicular biopsies for infertility.

Crystal Identification in Tissue Specimens

In cases of suspected gout where the detection and identification of crystals is required in tissue specimens, special collection procedures should be followed.

The tissue specimen should be placed into a specially marked “specimen container” containing absolute alcohol. Where routine histopathology is required in addition to crystal detection, the specimen should be divided. Place one portion in formalin (as normal), the other into alcohol.

Appropriate specimen collection jars containing alcohol are available from the Histology Department by phoning 02 6285 9855.

Disaccharidase Assays

These tests are performed on small bowel biopsies of patients with pain or diarrhoea where sugar intolerance is suspected. Wrap the biopsy loosely in parafilm and then in foil, store in a small screw cap vial and immediately freeze. Phone Couriers on 02 6285 9877 for rapid pick up of specimen.

Electron Microscopy

With the continuing advancements in immunohistochemistry, electron microscopy is less commonly required as an adjunct to light microscopy.

Applications of this technique include the further identification of undifferentiated malignancies, interpretation of renal biopsies and identification of some microbial organisms.

Tissue for electron microscopy should be finely sliced and placed in glutaraldehyde. Please phone the Histology Department on 02 6285 9855 for further details.

Frozen Sections
The histopathologists are available by arrangement for frozen section diagnosis. For frozen section bookings, please phone the Histology Department on 02 6285 9857. It is preferred that at least 24 hours notice is given to facilitate arrangements for a pathologist and technologist to be on site for the operation.

Cryostat facilities are available at Calvary John James Hospital, Calvary Hospital and The National Capital Private Hospital to provide an on–site frozen section diagnosis. There is also a cryostat at the laboratory.

Hormone Receptor Assays
Tumour hormone receptor analysis (usually breast carcinoma) is now performed on formalin fixed tissue by the immunoperoxidase method, and no special collection procedure is necessary.

Liver Iron Content
Liver biopsies for iron estimation require special collection procedures and should not be placed in fixative.

Please phone the Histology Department on 02 6285 9855 for further information.

Lymph Node Biopsy
Lymph nodes can be surgically excised or sampled via fine needle aspiration for microbiological examination, suspected primary or metastatic neoplasia, lymphoproliferative diseases and other causes of lymphadenopathy. Lymph node biopsy may also be useful for staging carcinoma and lymphoma, particularly if lymphoproliferative diseases are suspected. For optimal results, the node should be removed with minimal trauma and submitted fresh to the laboratory in a clean dry specimen container that is kept cool, (but not frozen).

In the laboratory the tissue may be examined using frozen section, smears, imprints, and flow cytometry while fixed tissue may be examined using light microscopy and immunohistochemistry.

Microbiological Investigation of Tissue Specimens

Tissue specimens for culture should be obtained using full aseptic technique and transferred to an appropriate container without contamination. Transport should be in a “specimen container” with the addition of a small amount of sterile saline to prevent drying out of the tissue. Specimens must not be transported in formalin.

Microsatellite Instability for Hereditary Colorectal Cancer
An immunoperoxidase antibody staining method is used to examine for the gene products. This is performed on routinely processed tissue.

Skin Biopsy for Inflammatory Skin Diseases
A detailed clinical history, including medications, clinical appearance and distribution of the skin lesions can be very helpful in arriving at an accurate diagnosis. Fresh tissue from skin biopsies can be examined by immunofluorescence and, if indicated, microbiology.

For the investigation of skin lesions such as bullous rashes and other dermatoses two punch or incisional biopsies are recommended. One should be placed in buffered 10% formalin for light microscopy and the other placed in transport medium for immunofluorescence (*see skin biopsy*).

Please phone the Histology Department on 02 6285 9855 for appropriate transport medium.

Testicular Biopsy for Infertility
Formalin is not an appropriate fixative for preservation and interpretation of testicular tissue. Bouin's fixative is used for this purpose.

Please phone the Histology Department on 02 6285 9867 for Bouin's fixative and for further instructions.

HIV (Human Immunodeficiency Virus)

Specimen: Serum – Gel

HIV Antigen / Antibody
At Capital Pathology routine initial serology provides testing for the p24 antigen and antibodies to HIV-1 and HIV-2. The antigen may be present early in infection. The antibodies can be seen with recent or past exposure. Negative results do not exclude infection. Repeat serology may be required, for instance if serum has been taken only a short time after exposure. Reactivity on the initial diagnostic test will result in further confirmatory tests being required, which are referred to a specialized reference laboratory. Supplementary HIV tests may include HIV viral load testing, HIV by Western Blot and HIV by PCR. Although the results from the routine initial tests are readily available, results from supplementary testing may take longer. Medicare benefit rebate is now available for HIV serology testing.

HIV PCR (Viral Load)–Quantitative

Specimen: Whole blood – EDTA x 2

Reference Range: Supplied with report

HLA–B27 Antigen (Human Leucocyte Antigen B27)

Specimen: Whole blood – EDTA
Keep specimen at room temperature.
Collect Monday–Friday (Friday am only).

- HLA B27 has a frequency of 7–10% in the general population but a much higher frequency in the seronegative spondyloarthropathies which are characterised by:
- Sacroileitis and spondylitis, causing low back pain and loss of lumbar mobility
- Tendonitis, including heel pain and other pain at muscle and tendinous insertion
- Oligoarthritis
- Urethritis
- Iritis and conjunctivitis.

As indicated by the term seronegative, all of these disorders are negative for rheumatoid factor.

The principal members of the group with the % positivity for HLA B27 are:

- Ankylosing spondylitis (95%– 98%)
- Reiter's disease (90%)
- Arthropathies associated with ulcerative colitis and Crohn's disease (70%)
- Post–enteric and post–venereal reactive arthritis (70%)
- Psoriatic arthritis (30%).

HLA Tissue Typing

Contact laboratory prior to collection.

Specimen: Whole blood ACD, EDTA, plain clot tube
The number and type of gel tubes for collection depends on the indication and history.
Specimen collection is available at all the Collection Centres.
Red Cross Sydney provides further information on specific requirements.
Can be collected Mondays to Thursdays, however a booking may need to be made with Red Cross by the doctor or Collection Centre prior to specimen collection.
Red Cross 02 9234 2322.
For further clinical information please contact the Director of Clinical Pathology on 02 6285 9895.

Homocysteine

Specimen: Plasma (Lithium heparin-gel tube)
Spin as soon as possible. Patient should be fasting.

Reference Range: Supplied with report

Previously, clinical interest in homocysteine in blood was confined to the rare inborn error of metabolism homocystinuria in which plasma homocysteine is markedly elevated to above 100 umol/L.

More recently an association has been described between mild to moderate hyperhomocysteinaemia (15–100 umol/L) and cardiovascular disease and many now regard it as an independent risk factor for coronary, cerebral and peripheral arterial disease.

Increasing intake of folic acid and to a lesser extent vitamin B6 and perhaps B12 may lower homocysteine levels. A daily supplement of 400 ug folic acid will in most patients increase serum folate above 15 nmol/L and drop plasma homocysteine to a low plateau. As yet there is no proof of a favourable effect on cardiovascular outcomes.

Homocysteine elevation above 20 umol/L is also a risk factor for venous thrombosis (2–3 x increase).

Hookworm

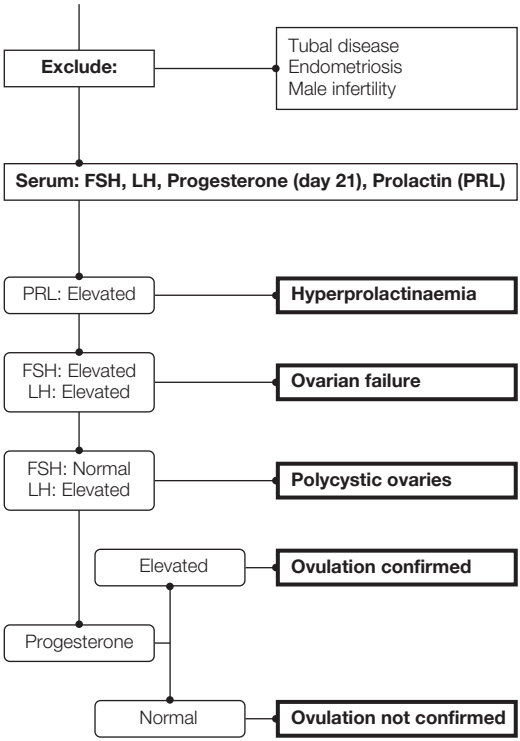
Hookworm refers to intestinal infestation with *Ancylostoma duodenale* (found in the Mediterranean basin, the Middle East, India and China) or *Necator americanus* (found in the Americas, sub–Saharan Africa and SE Asia). Heavy infestations are an important cause of iron–deficiency anaemia.

Diagnosis is by identifying eggs in faeces.

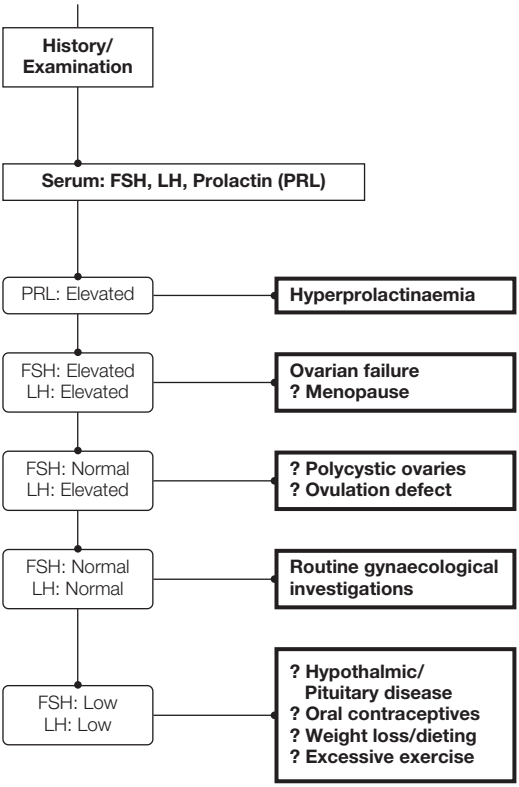
Treatment is with mebendazole 100 mg, 12–hourly for 3 days.

Hormone Assessment (Female)

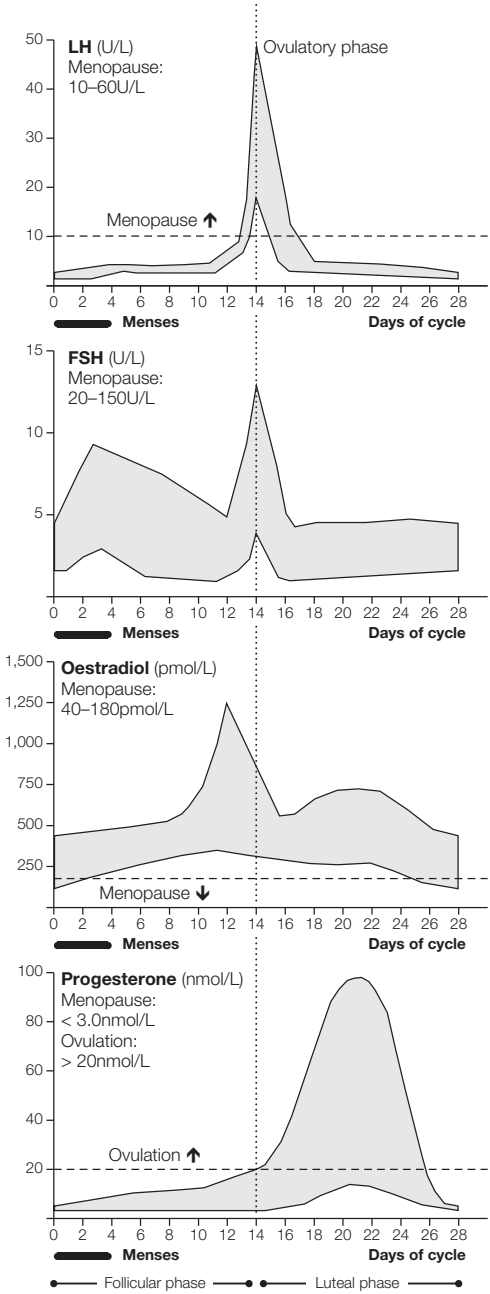
Suggested scheme for the work-up of Female Infertility



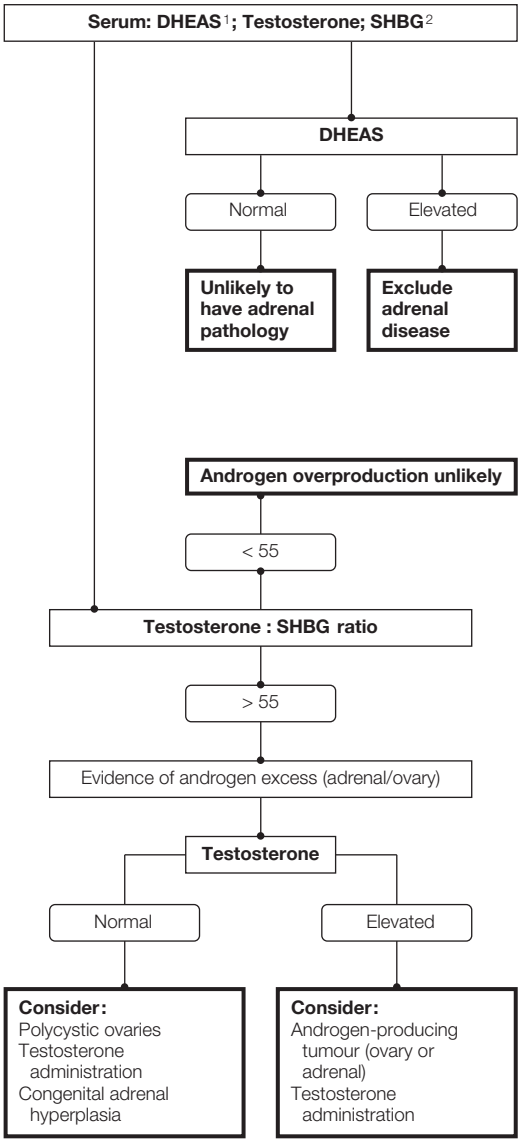
Evaluation of irregular periods



Hormonal changes during menstrual cycle



Evaluation of ?Androgen excess and/or Hirsutism (Female)



1. DHEAS: dehydroepiandrosterone sulfate
2. SHBG: sex hormone binding globulin

Human Papillomavirus (HPV)

Capital Pathology offers DNA testing for high–risk strains of Human papillomavirus (HPV) using the Cobas 4800 HPV DNA assay.

HPV infection is now considered a necessary but insufficient cause of cervical cancer. Co–factors are required to transform the cells into truly neoplastic cells. There are, however, many different sub–types of HPV and only some of these have been shown to be associated with the development of high–grade cervical lesions. The HPV test we offer identifies only **high risk** subtypes.

The HPV test is designed to be used in conjunction with the Pap smear result to assist doctors in making decisions on the management and follow–up of selected patients.

The basis of the Test?

- The test used is the Cobas 4800 HPV DNA assay. This is more specific and sensitive than previous tests.
- The test specifically identifies HPV 16 and HPV 18, the HPV genotypes most strongly associated with cervical neoplasia. A further 12 high risk genotypes (31,33,35,39,45,51,52,56,58,59,66 and 68) are tested and reported as a group but not individually identified. Infection with these genotypes confers a lower risk of developing cervical cancer than infection with HPV 16 or HPV 18.
- The result is reported as detected or not detected.

When is it indicated?

The clinical usefulness of this test is still being evaluated in many trials. However, the test may be of benefit in the following situations:

Management of smears showing persistent low grade non–specific changes

HPV testing may clarify management options in smears with mild atypia or uncertain changes. A positive HPV test indicates that a high grade intraepithelial lesion may be present. A negative HPV test may allow follow–up with further smears. At present there is no Medicare rebate for HPV testing in this circumstance.

Management of low grade (CIN 1) lesions

Neither cytology nor histological biopsy can predict which low–grade lesions will progress. Many CIN1 lesions will harbour a high risk HPV subtype, but many of these will regress. HPV testing in this circumstance will not predict which lesions will regress, persist or progress. HPV testing in this situation is not indicated.

Follow–up after treatment

Persistence of high risk HPV after treatment carries a risk of progression. Testing for HPV after treatment is therefore an effective way of monitoring cure. This is the only situation where there is a Medicare rebate for HPV testing. This is the “test of cure” following treatment of a high grade squamous intraepithelial lesion (see below).

Management of equivocal histology

A negative HPV test and a normal colposcopy give a negative predictive value of 98%. Testing for HPV therefore can assist management decisions when a negative or equivocal result is reported on a colposcopically directed biopsy.

How is it collected?
The HPV test can be collected:

- Co-collection of the conventional smear and the *ThinPrep* pap test – the conventional Pap smear is performed and the instrument is then rinsed in the *ThinPrep* vial. The HPV test can be performed from the sediment remaining in the vial after the *ThinPrep* test has been completed. There is no need to take a separate HPV sample.
- As a separate specimen using a cervex sampler and transferring the material into the *ThinPrep* vial.
- At colposcopy – the HPV test is collected prior to the application of acetic acid or any other solution.

Cost

The HPV test has no associated Medicare rebate except for follow up of high grade lesions after treatment. Currently the cost of HPV testing is $70 if The Medicare criteria are not met.

Management of women with treated high grade squamous intraepithelial lesions:
There is now a Medicare rebate for HPV testing in the following circumstance alone.

When a woman has received excisional or ablative treatment for a high grade squamous intraepithelial lesion (HSIL) of the cervix, or who within the last 2 years has had a positive HPV test after exisional or ablative treatment for high grade squamous intraepithelial lesions of the cervix, or who is already undergoing annual cytology review for follow up of a previously treated HSIL, then this test can be ordered to a maximum of two requests per 24 month period. When there have been two consecutive negative Pap smears and HPV tests, then the woman can return to the normal screening interval. Therefore if ordering HPV testing for these circumstances, please clearly state the reason for the HPV test on the pathology request form. We will then be able to bill for the correct item which will be rebatable from Medicare. If a patient does not fulfil the criteria as per the Medicare Schedule, then a non rebatable account will be issued to the patient for the cost of the HPV DNA assay.

See *Cervical Cytology*

Hydatid Serology

Specimen:	Serum – Gel
Reference Range:	Supplied with report

Hydroxyproline

Specimen:	Fast overnight. In the morning, void all the urine and discard. Do not have any breakfast (water only). After 2 hours collect 10 mL urine in specimen container. Keep cool. May be refrigerated for up to one week, otherwise freeze.
Reference Range:	Supplied with report

5–Hydroxytryptamine (5HT) (Serotonin)

Specimen:	Serum – Gel tube Spin, separate and freeze serum or 24–hour urine (HCL preservative). Patient should follow special diet. Please contact your nearest Collection Centre or the Client Services Department on 02 6285 9802 for information sheet.
Reference Range:	Supplied with report
See	*Carcinoid Syndrome*

17–Hydroxy Progesterone (17–OHP)

Specimen:	Serum – Gel
Reference Range:	Supplied with report

17–OHP has replaced urinary pregnanetriol as the best screening test for the most common form of congenital adrenal hyperplasia (CAH).

17–OHP is a precursor of cortisol, androgens and oestrogens. In CAH, a block in the cortisol pathway leads to overproduction of 17–OHP and androgens.

See *Congenital Adrenal Hyperplasia*

Hypertension–Endocrine Causes

These include:

- Conn's syndrome
- Cushing's syndrome
- Phaeochromocytoma
- Renal artery stenosis
- Acromegaly
- Hyperthyroidism.

Hypogammaglobulinaemia

Can be found in:

- Myeloma
- Chronic lymphocytic leukaemia
- Lymphoma
- Non–selective protein loss
- Transiently with a recent viral infection
- Congenital deficiency.

See *Immunoglobulins IgA, IgG, IgM*

Hypoglycaemia

The glucose threshold at which symptoms of hypoglycaemia develop varies widely between individuals but in general a glucose level < 3.0–3.5 mmol/L can be described as hypoglycaemic except in neonates where the lower limit of "normal" is about 2.5 mmol/L.

Hypoglycaemia in diabetics treated with insulin or sulphonylureas
Symptoms are common, diverse and often misinterpreted or overlooked. The transient nature of the hypoglycaemia means that laboratory collects usually miss the trough but any value below 4.0 is suspicious. Symptoms and signs include irritability, tremulousness, faintness, sweating, pallor, hunger, weight gain, headaches (headache on waking may be due to nocturnal hypoglycaemia), personality change, deterioration in level of consciousness, coma.

Reactive (functional) hypoglycaemia
This common but elusive syndrome with its symptoms of irritability, tremulousness and hunger ("faint with hunger") is not easily diagnosed by the laboratory. It is seen in children, young adults, and to a lesser extent older people. The symptoms are felt some 2–5 hours after food, not while fasting.

The extended GTT has been the traditional test but is a burden to the patient and has too many false positives and false negatives.

A marginally more useful test is the measured blood glucose during an attack but the logistics of this are often not easy.

The best test is a trial diet which, if successful, combines diagnosis with therapy. Sweet refined carbohydrates are changed to unrefined carbohydrates with one or two snacks added between meals.

Fasting hypoglycaemias
These are much less common than the non–fasting varieties and much more likely to have an identifiable organic basis.

The first test is to measure a fasting glucose. If low it should be followed by a fasting insulin and repeat glucose.

- Raised or inappropriately normal fasting insulin
  – Insulinoma
  – Exogenous insulin or sulphonylureas.

- Low fasting insulin
  – Advanced liver disease
  – malignancy
  – insulin autoantibodies
  – alcohol
  – pituitary/adrenal insufficiency.

Neonatal hypoglycaemia

The at–risk infants are:

- Low birth weight
- Infants of diabetic mothers.

In both, blood glucose levels should be monitored, beginning an hour after birth, and treatment given if levels are below 2.0–2.5 mmol/L. Glucose levels can rise and fall quickly during the neonatal period. Clinical signs are an inadequate indication of hypoglycaemia.

Hypopituitarism

Usually due to a tumour of the pituitary or hypothalamus but pituitary infarction or haemorrhage can occur in a normal pituitary, classically as Sheehan's syndrome after obstetric haemorrhage, but also in other situations, including bleeding disorders, cardiac by–pass surgery and some imaging procedures.

Clinical hypopituitarism may present as amenorrhoea, hypogonadism, hypothyroidism, adrenal insufficiency, hyponatraemia.

Useful tests include:

Thyroid T4 low but TSH usually normal. The TSH measured in this situation can coexist with clinical hypothyroidism so is assumed to be biologically inert.

Gonads FSH and LH may be low, particularly in post–menopausal women who otherwise have high levels. Testosterone may be low in men.

Hyposplenism

After splenectomy, red cells show Howell–Jolly bodies, targeting and spherocytosis and there is thrombocytosis and leukocytosis which may or may not persist.

A hyposplenic blood picture can also be seen in other conditions associated with splenic atrophy:

- Coeliac disease
- Inflammatory bowel disease
- Autoimmune disease, particularly with haemolysis
- Sickle cell disease.

After splenectomy a patient may have increased susceptibility to infection which can be overwhelming in children. Pneumococci are the usual cause but other organisms such as Haemophilus or meningoccus may be responsible. Pneumococcal and meningococcal vaccines can be given every 5–7 years or penicillin prophylaxis in susceptible patients.

I

Idiopathic Immune Thrombocytopenic Purpura (ITP)

A relatively common autoimmune disorder characterised by an isolated thrombocytopenia in the absence of drugs, chemicals or any haematological or non–haematological disorders.

Acute immune thrombocytopenia is distinguished from acute viral thrombocytopenia by the clinical course with the latter usually recovering within a few days without the requirement for specific therapy. Chronic immune thrombocytopenia is diagnosed when the thrombocytopenia persists for more than 6 months. It may have a viral trigger, occur as part of an autoimmune disorder (e.g. SLE), arise in association with a lymphoproliferative disease or as an idiopathic finding.

ITP is usually a diagnosis of exclusion. Appropriate investigations may include a coagulation screen with bleeding time (to evaluate a possible associated platelet dysfunction), autoimmune investigations (ANA, anti–DNA rheumatoid factor, anticardiolipin antibodies, platelet antibodies), lupus anticoagulant screen, protein electrophoresis and immunoglobulins, HIV.

Treatment is either with prednisone or intravenous immunoglobulin, particularly when the platelets fall below $20x10^9$/L.

See *Thrombocytopenia*

IgA Deficiency

A selective reduction in IgA is a common congenital immune deficiency syndrome occurring in 1:700 people. Most individuals are asymptomatic. IgA deficiency is associated with autoimmune disorders including coeliac disease, SLE, autoimmune haemolytic anaemia, rheumatoid arthritis and thyroiditis.

Anaphylaxis may occur as a result of IgG antibodies to IgA when the individual is exposed to blood transfusion or gammaglobulin therapy.

See *Immunoglobulins IgA, IgG, IgM*

IgE total

Specimen: Serum – Gel

Reference Range: Supplied with report

IgE antibodies mediate Type I allergic reactions – allergic rhinitis, asthma, atopic dermatitis, anaphylaxis.

Total IgE is elevated in patients with multiple allergies and parasitic infestations. Very high levels, above 1000 IU/L, are found with atopic dermatitis, fungal sinusitis and allergic pulmonary aspergillosis.

See *RAST*

Imipramine

Specimen: Plasma – Lithium heparin

Reference Range: Supplied with report

See *Antidepressant Drugs, tricyclic*

Immunoglobulin G Subclasses

The four IgG subclasses are numbered 1 to 4. In some patients, recurrent respiratory tract infections are associated with Ig subclass deficiency even though the total IgG is within the usual range.

Immunoglobulins, IgA, IgG, IgM

Specimen: Serum – Gel

Reference Range: Supplied with report

Quantitation of immunoglobulins, particularly paraproteins, is method–dependent so when monitoring a patient with myeloma or an apparently benign paraprotein, the immunoglobulins will ideally be measured by the same laboratory using the same method. In practice, laboratories change methods at times so when a result shows an unexpected rise or fall, the laboratory should be consulted.

Immunoglobulins are antibodies and constitute the gamma fraction of serum globulins.

Abnormalities of immunoglobulins include:

- Monoclonal increases, benign or malignant
- Polyclonal increases as seen in chronic inflammatory disorders
- Polyclonal decreases: immune paresis.

Influenza

Specimens: Swab – One dry Nasopharangeal or viral throat swab for Influenza viral PCR.
Serology – Serum – Gel

The accompanying request form is to request Influenza PCR .

If serology is required – serum (gel tube) 5–10mls of blood.

The diagnosis of suspected influenza in a patient with respiratory symptoms can be made by direct viral antigen detection (preferred) or by indirect antibody detection.

The preferred test is influenza A and B viral detection by polymerase chain reaction (PCR) which is highly sensitive and specific. There is a rapid immunochromatographic test for influenza viral detection, however it has limited sensitivity and specificity and should be used only as a guide. Serology for influenza specific antibodies is available, and a positive diagnosis is defined by a single high titre or by a rise in titre by at least four fold in repeat serum specimens 10–14 days apart. Serology is less specific and may take longer for results.

If Avian influenza is suspected on the basis of the history and clinical findings, contact the local area public health unit first for advice on how to proceed. Depending on the presentation the patient may need to be triaged via the public health unit, with specimens and tests referred to a nominated laboratory with access to PCR tests for Avian influenza.

INR (International Normalised Ratio)

Specimen: Plasma – Sodium Citrate

The INR is the test used to monitor warfarin anticoagulation therapy which is being increasingly used across the range of thromboembolic disease. Warfarin is a vitamin K antagonist and acts by reducing levels of Factors II (prothrombin), VII, IX, and X. The INR is a standardised prothrombin ratio calibrated so that INR results from one laboratory are directly comparable with those from another.

Recommended therapeutic range: clinical state

1.5–2.0	Consider in cases where risk of cerebral haemorrhage is high, i.e. over 75yrs, for some indications such as atrial fibrillation (AF). Long–term prevention of CVT/PE after > 3 months after last event.
2.0–3.0	Prevention of DVT, therapy for DVT or PE, preventing systemic embolism (AF, valvular heart disease, after AMI, tissue heart valves–1st three months).
2.5–3.5	Bileaflet mechanical heart valve (aortic).
3.0–4.5	Mechanical prosthetic heart valve (high risk), secondary prophylaxis in antiphospholipid syndrome.
Reference:	ACT Regional Guidelines for the Use of Warfarin, Sept 2003

Insulin

Specimen: Serum – Gel
Patient should be fasting.

Reference Range: Fasting insulin < 12mU/L makes insulin resistance unlikely

Insulin measurements may be requested in conjunction with a Glucose Tolerance Test.

Insulin-like Growth Hormone Factor–1 (IGF–1)

Specimen: Serum – Gel

Reference Range: Supplied with report

The action of Human Growth Hormone (HGH) from the pituitary is mediated through IGF–1 which is formed in the liver. As well as its effects on growth, it has insulin–like actions. IGF–1 synthesis is suppressed by malnutrition as well as HGH deficiency.

IGF–1 and HGH are measured in suspected acromegaly and in pituitary dwarfism.

Intrinsic Factor Antibodies

Specimen: Serum – Gel

Reference Range: Not detected

See *Vitamin B12*
Pernicious Anaemia

Iron

Specimen: Serum – Gel

Reference Range: Child 4.5–27 umol/L
Adult 9–27 umol/L

Serum iron levels show a marked diurnal variation with morning levels higher by 30%.

They are also rapidly depressed to low levels by acute or chronic infections or any other chronic disorder.

Because of these large fluctuations, serum iron is of little diagnostic use on its own but when combined with iron–binding capacity/transferrin and ferritin, total body iron deficiency or excess can usually be defined.

Serum iron is elevated by:

- Oral or parenteral iron supplements
- Haemochromatosis
- Alcohol
- Hepatitis
- Oestrogens/oral contraceptives
- Iron poisoning – children absorb oral iron easily and in acute poisoning can present with levels in the 100–500 umol/L range.

Iron deficiency

Useful tests

Ferritin	The most sensitive and specific test. If low it is almost pathognomonic of iron deficiency, latent or overt, but a normal result does not exclude iron deficiency as coexisting inflammatory disease can elevate ferritin independently.
MCV & MCH	Reduced, but not until ferritin is < 10
Hb	Reduced, but anaemia develops later than the other changes
Blood film	Oval cells, hypochromia, microcytosis
Iron	Reduced, but many other conditions cause a fall in serum iron
IBC	Increased (may be normal if inflammation coexists)
% saturation	Reduced

Aetiology of iron deficiency

The treatment of iron deficiency must always be accompanied by a search for the cause. Deficiency is so common in women of child–bearing age that it is easy to attribute all deficiencies in women to menstrual loss while forgetting this in not possible after the menopause – nor in males of any age. Deficiencies fall into three broad groups:

1. Chronic blood loss

Female genital tract:

- Menstrual loss
- Pregnancies
- Tumours of female genital tract with abnormal bleeding.

Gastrointestinal loss:

- Oesophagus, stomach, duodenum–peptic ulcer, NSAIDs, hiatus hernia, varices
- Large bowel – tumours (iron deficiency is a common presentation), ulcerative colitis, haemorrhoids
- Small bowel – hookworm, congenital vascular malformations, Meckel's diverticulum.

Other:

- Epistaxis, haematuria, haemoptysis, telangiectasia.

2. Dietary insufficiency

Mainly in infants, adolescents and the elderly.

3. Malabsorption
Coeliac disease particularly.

Iron deficiency in athletes
Iron deficiency is more common in athletes for three reasons:

- They may be on relatively low iron diets
- They frequently suffer occult gastrointestinal blood loss
- They may suffer haemoglobinuria with urinary iron loss.

For these reasons many athletes take oral iron supplements when their ferritins are towards the lower end of the reference range.

Islet Cell Antibodies (ICA)

Specimen: Serum – Gel

Reference Range: Supplied with report

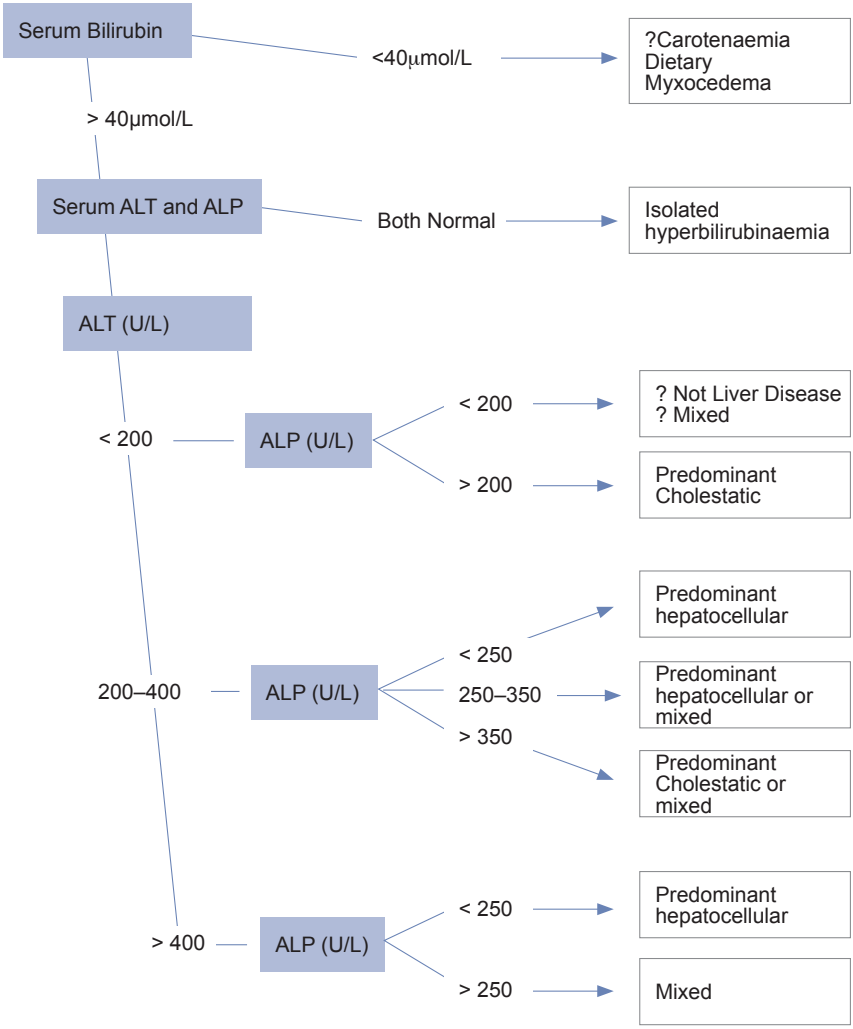
J

Japanese Encephalitis Antibodies

Specimen: Serum – Gel

Reference Range: Supplied with report

Jaundice



Joint Aspirate

See *Synovial Aspirate*

K

Ketones

Specimen: Random urine

This test, which is part of routine urinalysis, is usually done by a dipstick method. Mild positives occur with starvation or illness. Moderate or strong positives in Type I diabetics indicate poor control requiring additional insulin and rehydration – urgently if there is significant acidosis.

"Ketostix" detects acetoacetate which accounts for only a small percentage of "ketone bodies", the greater portion being beta–hydroxybutyrate.

Klebsiella pneumoniae

An aerobic gram–negative enteric bacillus. Found on mucosal surfaces and in the faeces of about 5% of healthy people. It occasionally causes pneumonia as well as biliary and urinary tract infections.

Kleihauer Stain for Fetal Haemoglobin

Specimen: Maternal whole blood – EDTA

Reference Range: Supplied with report

Klinefelter Syndrome

XXY is the usual karyotype. Testosterone levels are typically reduced and FSH and LH increased. Other pituitary tests are usually normal.

Clinically, patients are often unusually tall with long legs. Testes are small, pubic hair scant and there may be gynaecomastia.

L

Lactate Dehydrogenase (LD) (LDH)

Specimen: Serum – Gel

Reference Range: Adults 100–250 U/L

LDH is an enzyme of low specificity found in liver, myocardium, skeletal muscle and red cells. It is also found in some malignancies, notably lymphomas and particularly non–Hodgkin's lymphoma.

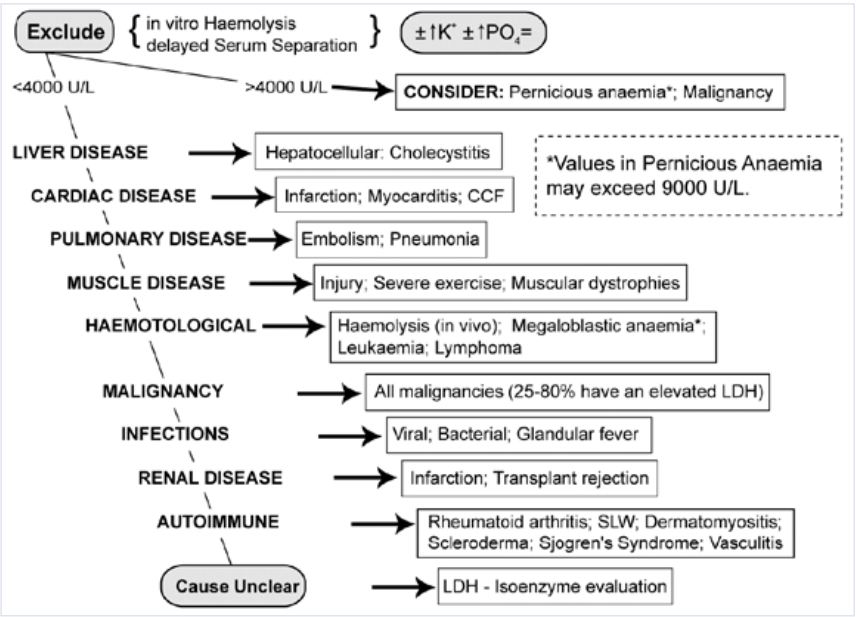
Elevations can be due to:

- Lymphomas, up to 2–3x the upper limit of the reference range – the level gives an estimate of tumour bulk
- Other tumours, especially germ cell
- Myocardial infarction, increase commences 12–24 hours after infarction, disappearing after 7–12 days
- Megaloblastic anaemias due to B12 or folate deficiency
- Haemolytic anaemia
- Artefactual haemolysis due to faulty specimen or collection
- Liver disease, particularly hepatitis
- Skeletal muscle damage.

Of the LD isoenzymes, LDI is derived from myocardium or erythrocytes, LD5 from liver.

Lactate Dehydrogenase (LDH) Interpretations

Elevated LDH



Lead, blood

Specimen: Whole blood – EDTA

Reference Range: Supplied with report

Domestic exposure occurs when old lead–based paints are being sanded or burnt off. Children are at risk when they ingest paint fragments or dust, as are pets when they lick their fur.

High levels should be repeated in 3–4 weeks.

Lead poisoning interferes with porphyrin metabolism causing elevation of urinary coproporphyrin, PBG and ALA. The porphyrin changes are a better indicator of toxicity than the blood lead on its own but are not used for routine industrial testing. Red cells may show basophilic stippling.

Lead, urine

Specimen: 24-hour urine (nil preservative)

Reference Range: Supplied with report

Urine leads are used for monitoring chelation therapy in lead poisoning after the diagnosis has been established using blood lead levels.

Legionella Antibodies

Specimen: Serum – Gel
Sputum for culture.

There are more than 30 species of Legionella, the cause of Legionnaire's disease.

The organism is found in hot water systems or the humidified air of air–conditioning systems, causing both sporadic cases and institutional outbreaks of atypical pneumonia.

Culture is the diagnostic method of choice. Legionella culture must be asked for specifically as it requires the use of specially prepared media. Specimens contaminated with sodium e.g. saline–induced sputum and bronchial washings, are not suitable for Legionella culture.

Most healthy adults have an antibody titre of < 1:128. Rising antibodies in paired sera provide good evidence of active infection. Paired sera should ideally be collected two weeks apart.

Leptospirosis

Specimen: Serum – Gel

Reference Range: Supplied with report

Since vaccination of cattle was introduced several years ago, the incidence of leptospirosis has dropped to the point where it is now rare. Host animals include cattle, pigs, horses and rodents. Human infection typically follows exposure to blood of infected animals or water contaminated with their urine.

For diagnosis, an antibody screen test is performed followed by (on sera that are positive) specific agglutination testing against the common strains of leptospira.

After infection, titres rise sharply into the thousands in those who have been infected in the past. The highest titre may not indicate the cause of the latest illness.

Consultation with the pathologist is suggested when acute leptospirosis is a possibility so that blood culture in special medium can be arranged.

Leuco–erythroblastic Blood Picture

Immature myeloid cells (myelocytes, metamyelocytes, band neutrophils), and immature red cells (nucleated red cells), are present due to invasion or disturbance of the normal marrow.

Causes include:

- Metastatic malignancy
- Primary haematological malignancy
- Myelofibrosis
- Acute haemolysis
- Thalassaemia major, especially after splenectomy
- Chronic lymphocytic leukaemia
- Chronic myeloid leukaemia.

Light Chains in Urine

See *Bence Jones Protein (BJP)*

Lignocaine

Specimen: Plasma – Lithium heparin
Blood should be collected at least every 12 hours in patients with evidence of cardiac or hepatic insufficiency.

Reference Range: Supplied with report

Lipaemia

After a fat–containing meal, and in the primary or secondary hypertriglyceridaemias, the serum is turbid or creamy due to fat particles. A severe degree of lipaemia can cause technical interference with some tests to the point where they become invalid.

See *Triglyceride*

Lipase

Specimen: Serum – Gel

Reference Range: < 61 U/L

Increase in lipase levels indicates pancreatitis, and this rise may be seen before amylase.

Lipid Disorders

A lipid screen consists of Cholesterol and Triglycerides. If HDL is required, please order specifically.

When classifying a lipid disorder for the purposes of treatment, two questions need to be asked:

1. Is it a primary lipid disorder or secondary to some other metabolic abnormality?
2. Which lipid fractions are elevated?
   Cholesterol only
   Cholesterol and triglyceride
   Triglyceride only.

Secondary disorders

These are easily identified and when treated may entirely correct the lipid abnormalities.

The common causes are:

- Obesity
- Alcohol
- Diabetes
- Hypothyroidism
- Nephrotic syndrome
- Liver disease
- Drugs: oestrogens, oral contraceptives, beta blockers, corticosteroids, thiazides, isotretinoin, antivirals, valproate.

Primary disorders

If no secondary causes are identified the disorder is presumed to be primary. There is a range of familial disorders which are presumptively identified by the family history or by physical signs such as xanthomata, corneal arcus.

The commonest familial disorders are familial hypercholesterolaemia and familial combined hyperlipidaemia. There are many others.

However, the commonest primary dyslipidaemia by a wide margin is polygenic hypercholesterolaemia, which is not familial. It is identified mainly by exclusion – no significant family history, no abnormal physical signs. Presumed to result from a combination of genetic and environmental factors, it provides a convenient diagnostic refuge.

Which lipid fractions are elevated?

Defining the abnormal fractions is important in both diagnosis and treatment.

The feature of familial hypercholesterolaemia is very high serum cholesterol.

The lipid elevation of familial combined hyperlipidaemia may be cholesterol only (1/3), triglyceride only (1/3), or both (1/3).

Causes of secondary dyslipidaemias do not reliably produce a constant picture. For example, though hypothyroidism typically causes elevation of cholesterol only, it can at times raise the triglyceride also.

Treatment

Dietary modification is common to the treatment of all dyslipidaemias.

Drug treatment, if required, will depend on whether the abnormality is cholesterol only (statins being the agents of choice), triglyceride only (fibrates usually first choice) or both.

PBS Qualifying Criteria for Lipid Lowering Therapy

Patient Category	Lipid Level for PBS Subsidy
Patients with existing coronary disease	Cholesterol > 4mmol/L
Other patients at high risk with one or more of the following: • Diabetes mellitus • Familial hypercholesterlaemia • Family history of coronary heart disease (first degree relative less than 60 years of age) • Hypertension • Peripheral vascular disease	Cholesterol > 6.5 mmol/L or Cholesterol > 5.5 mmol/L and HDL< 1mmol/L
Patients with HDL < 1 mmol/L	Cholesterol > 6.5 mmol/L
Patients not eligible under the above: • Men 35–75 years • Post menopausal women up to 75 years	Cholesterol > 7.5 mmol/L or Triglyceride > 4 mmol/L
Other patients not included in the above	Cholesterol > 9 mmol/ L or Triglyceride > 8 mmol/L

Lipoproteins

Specimen: Serum – Gel

In serum, lipids are carried as lipoprotein particles which are grouped in four fractions:

Lipoprotein Fraction	Cholesterol	Triglyceride
Chylomicrons	small	++++
LDL	++++	small
VLDL/IDL	++	++++
HDL	++	small

Diagnosis of the hyperlipoproteinaemias requires a fasting specimen to eliminate the contribution of dietary triglyceride in both chylomicrons and the VLDL fraction.

Hyperlipoproteinaemias can be familial or secondary (see separate entries under *Triglyceride* and *Cholesterol* for aetiology of the secondary disorders) or, commonly, a mixture of both. Occasionally the diagnosis is obvious, e.g. the clear–cut familial hypercholesterolaemias causing severe coronary artery disease in early adult life – but most of the common hypercholesterolaemias and hypertriglyceridaemias are part of a large amorphous population, deeply affected by diet and lifestyle and mixed inextricably with the "normal" population, whoever they are.

Typing of hyperlipoproteinaemias

The WHO classification is that of Fredericksen who recognised six types; I, IIa, IIb, III, IV, V. The classification has limitations. Familial hyperlipoproteinaemias can present as different types in different family members. A single aetiology (e.g. hypothyroidism) can be associated with several types. Treatment can convert one type to another. Nevertheless the terms are still used. Types II and IV, and to a lesser extent V, are commonest.

	IIa	IIb	IV	V
Total cholesterol	+++	+++	+	+
Triglyceride	n	++	+++	+++
LDL cholesterol	++	++	n	n
VLDL cholesterol	n	+	+++	++
Chylomicrons	–		–	++
Turbid serum	–	++	++	++

Note that the classification does not use HDL cholesterol.

See *Apolipoproteins*
Lipoprotein Electrophoresis
Cholesterol
Triglyceride
Lipid Disorders

Lipoprotein (a)

Specimen: Serum – Gel

Reference Range: Supplied with report

Lipoprotein (a), not to be confused with apolipoprotein A which is the protein component of HDL, is an independent risk factor for coronary artery disease (CAD). It can be of particular interest in patients whose CAD is not explained by other risk factors.

Levels are largely genetically determined but tend to rise after the menopause or with renal impairment. Fasting, exercise or statins have little effect. Levels may be lowered by niacin and perhaps by fibrates. Acute illness can have an effect and lipoprotein (a) should not be measured within 3 months of a myocardial infarction.

Lipoprotein Electrophoresis

Specimen: Serum – Gel

Four fractions are identified:

Alpha–I	Contains HDL
Pre–beta	Contains VLDL, elevated in Type IV, IIb or III
Beta	Contains LDL, elevated in Type IIa or IIb
Chylomicrons	Found in non–fasting serum or Type V

Lipid EPG is a specialist investigation. For most clinical purposes a lipid abnormality is defined by quantitation of cholesterol fractions and triglyceride on a fasting specimen.

Listeria monocytogenes

Specimen: Blood cultures, CSF, for acute infection
Stool for carrier state.

Listeria, an aerobic gram–positive bacillus, is widely distributed in soil, water and many animals. It is occasionally found in the human gastrointestinal tract. Spread to humans is mainly via contaminated food. Food borne outbreaks due to contaminated coleslaw, milk, soft cheese and mussels have been reported. Although most infections are mild and harmless, septicaemic infection in pregnant women, presenting as a flu–like illness, may lead to serious fetal infection leading to stillbirth or neonatal meningitis. Immunocompromised persons and the elderly are also at risk of invasive disease.

Treatment is penicillin (or amoxycillin) with or without gentamicin. Erythromycin or cotrimoxazole are less desirable alternatives. Treatment should be for a least 14 days. *L. monocytogenes* is resistant to all cephalosporins.

Lithium

Specimen: Serum – Gel

Desirable therapeutic range: 0.5–1.0 mmol/L

The specimen should be taken 12 hours after the last dose. Steady state levels are achieved 2–5 days after a dose change, or longer in older patients with renal impairment. When lithium is stopped, the level falls with a half–life of 1–3 days.

Because of the high incidence of toxic effects, monitoring is regarded as essential every 3 months, and more often if clinically indicated. Baseline TSH, T4 and creatinine levels should be measured before commencing therapy and 6 to 12 monthly thereafter.

Manifestations of toxicity

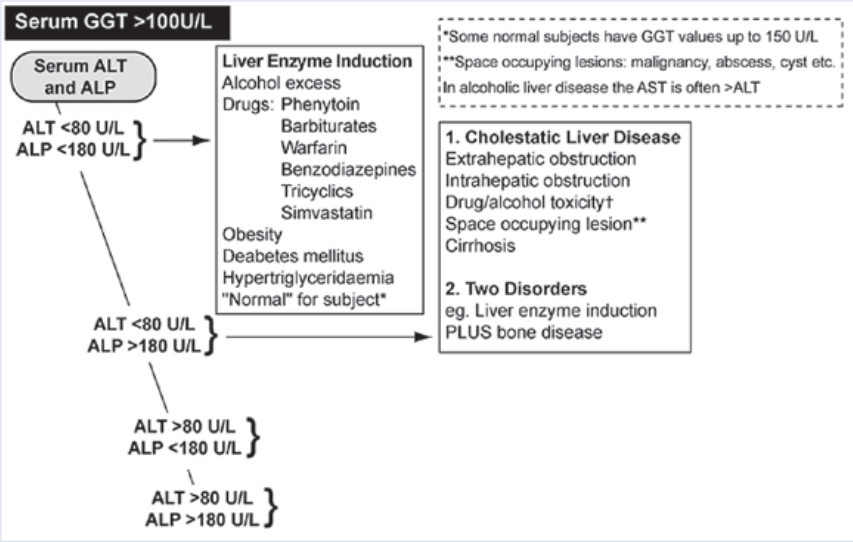
- A wide range of CNS and GI effects
- Hypothyroidism – relatively common
- Nephrogenic diabetes insipidus
- Serum potassium maybe elevated
- Renal damage with raised s. creatinine may occur when serum levels have been consistently in the toxic range over a long period
- High serum calcium
- Cardiac toxicity: AV block, T wave changes, premature ventricular contractions.

Drugs that elevate lithium:

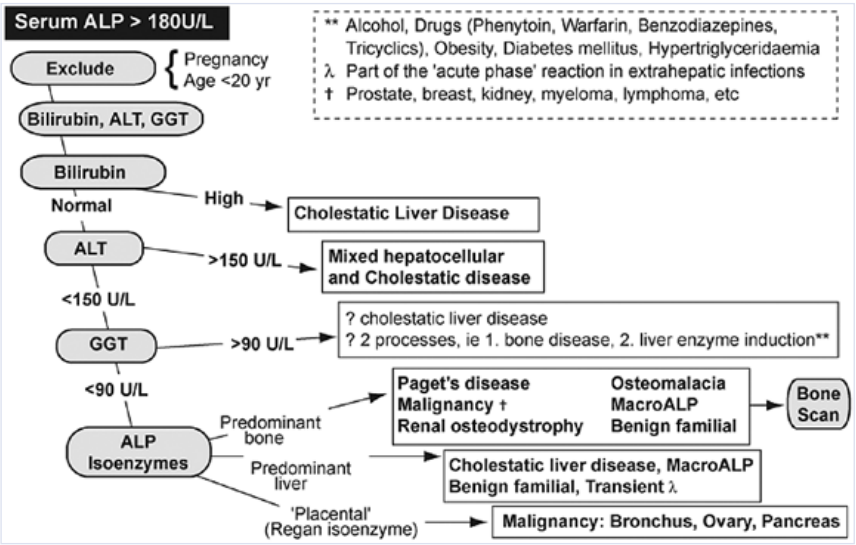
- Diuretics
- NSAIDs
- Metronidazole
- ACE inhibitors.

Liver Function Test / Interpretation

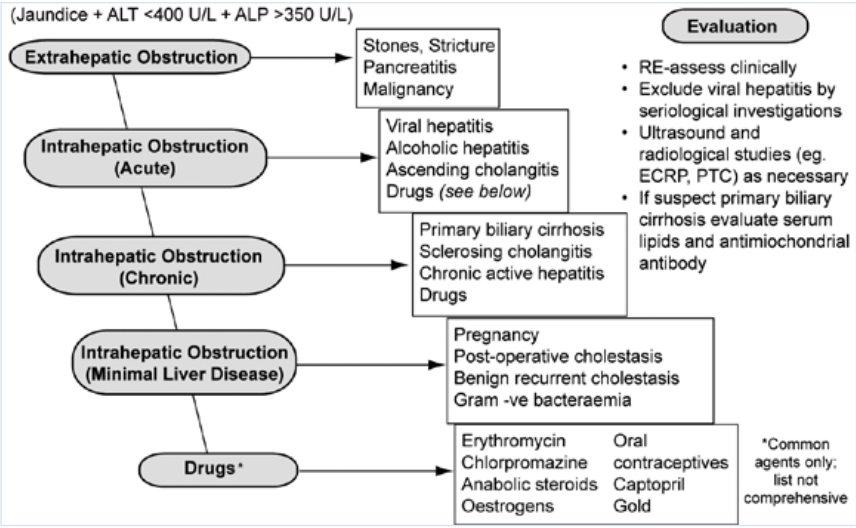
Elevated GGT



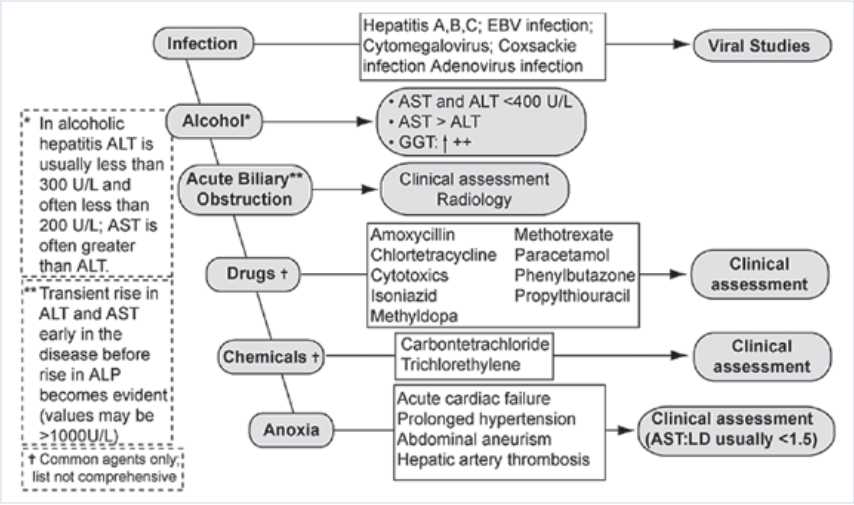
Elevated ALP



Predominant Cholestatic Pathology



Predominant Hepatocellular Pathology



Lorazepam (Ativan)

Specimen: Plasma – Lithium heparin

Reference Range: Supplied with report

Lupus Anticoagulant / Inhibitor

Specimen: Plasma – Sodium Citrate x3

Reference Range: Not detected

The lupus anticoagulant is an acquired coagulation inhibitor, which is associated with SLE and thromboembolic disorders. It is associated, but not synonymous with cardiolipin antibodies. In the context of thrombotic disease or recurrent abortions, a positive lupus anticoagulant detected on two occasions at least three months apart, with or without anticardiolipin antibodies, defines an antiphospholipid antibody syndrome. Anticardiolipin antibodies are sometimes found transiently in healthy people.

The APTT is prolonged in patients with LAC and fails to correct in the APTT 1+1 test.

The tests performed routinely as the LAC screen are KCT (Kaolin Clotting time), APTT, PR, DRVVT (Dilute Russell's Viper Venom Time) and DTTA (Dilute Tissue Thromboplastin Assay).

Luteinising Hormone (LH)

Specimen	Serum – Gel	
Refernce range:	Adult Female	
	basal	2–14 IU/L
	mid-cycle	14–110 IU/L
	post menopausal	15–97 IU/L

Lymphocyte markers

Specimen Whole Blood EDTA x 2

Lymphocytes play a central role in the immune response. The two main types of cell, identified by cell marker studies are:

- T cells, primarily responsible for cell–mediated immunity.
- B cells, primarily responsible for humoral immunity (immunoglobulin production).

Lymphocytes are also classified by their CD (cluster designation) number.
For example, CD4 cells are T cells used as a marker for immune system damage in HIV infections.

Lymphocytosis
An absolute lymphocytosis (> $4.0x10^9$/L) is typically found in two situations:

- Viral infections, particularly infectious mononucleosis, acute viral hepatitis and CMV infections.
- CLL (Chronic Lymphatic Leukaemia), in which the proliferating lymphocytes are monoclonal B cells with light chain restriction. CLL should always be suspected when there is an unexplained progressive lymphocytosis at or beyond middle age.

Lymphopenia
Causes include:

- Viral illnesses
- Pancytopenia due to any cause
- Advanced Hodgkin's disease
- Congestive heart failure
- Steroid therapy
- AIDS.

Variant lymphocytes
These cells, also called reactive lymphocytes, transformed lymphocytes or atypical lymphocytes, are small lymphocytes which have undergone antigenic stimulation (e.g. by a virus) that has transformed them into larger cells readily recognised in a blood film.

The commonest causes of variant lymphocytes are:

- Infectious mononucleosis (EBV)
- Acute viral hepatitis A, B or C
- CMV infections
- Toxoplasmosis.

Less common causes are:

- Other viral infections
- Immune disorders
- Chronic non–viral infections.

See *Histopathology*

M

Macrocytosis

Macrocytes are red cells with an MCV (Mean Cell Volume) above 98. They can be divided into three groups.

1. **The megaloblastic macrocytic anaemias**

These are distinguished by the presence of megaloblasts in the bone marrow as precursors of the peripheral blood macrocytes. Megaloblastic anaemias are due to B12 or folate deficiency. The macrocytes are oval in shape rather than round. With well–established deficiency there are hypersegmented neutrophils, neutropenia and thrombocytopenia.

2. **Macrocytosis due to reticulocytosis**

Reticulocytes are somewhat larger than older red cells. They are an important feature of haemolytic and post–haemorrhagic anaemias.

3. **Other**

In this large group of normoblastic processes, the macrocytes are round. The list includes:

- Alcoholism
- Liver disease
- Hypothyroidism
- Malignant infiltration
- Cytotoxic drugs (especially hydroxyurea).
- Myeloma
- Aplastic anaemia
- Myelodysplastic syndromes
- Chronic obstructive airways disease

Macroglobulinaemia

IgM paraproteins are present in three conditions:

1. **Benign IgM paraproteinaemia**
   The commonest cause. The blood picture is normal, the patient asymptomatic and IgM levels are static and usually < 10g/L.
2. **Waldenstrom's macroglobulinaemia**
   A malignant process in which the IgM band is heavy, the blood picture is abnormal and the marrow infiltrated with abnormal plasmacytoid lymphocytes.
3. **Malignant lymphoma or CLL (Chronic Lymphatic Leukaemia)**
   These should always be searched for clinically, and on the blood film, when an IgM paraprotein is found.

See *Immunoglobulins, IgA, IgG, IgM*

Magnesium (Mg)

Specimen: Serum – Gel

Reference Range: 0.65–1.05 mmol/L

Decreased by:

- Excessive loss of fluid and electrolytes, diarrhoea etc
- Inadequate intake: inadequate parenteral nutrition, low levels in diet and water, malabsorption
- Drugs: diuretics, aminoglycosides, digoxin, cytotoxics, laxative abuse
- Alcoholism
- Endocrine: hyperthyroidism, hyperparathyroidism, diabetic ketoacidosis, SIADH
- Hypokalaemia
- Redistribution of Mg into cells, e.g. alkalosis, acidosis, severe illness.

Increased by:

- Renal insufficiency
- Dehydration
- Addison's disease
- Haemolysis.

Severe magnesium deficiency affects neuromuscular function and the cardiac conduction system. Clinically this can be manifested as tetany, convulsions, cardiac arrest. Epidemiologically, magnesium deficiency in water supplies has been linked to cardiac dysrhythmias and myocardial infarction.

Causes of Hypomagnesaemia

- Alcoholism
- Renal loss
  - Hypercalcaemia
  - Renal tubular acidosis
  - Osmotic diuresis
- Drugs
  - Loop Diuretics
  - Gentamicin
  - Cisplatin
- Gastrointestinal disorders
  - Vomiting
  - Diarrhoea
  - Malabsorption
- Endocrine
  - Diabetes mellitus (probably increased renal loss)
  - Hyperaldosteronism
  - Hypoparathyroidism
  - SIADH
- Miscellaneous
  - Acute pancreatitis (sequestration)
  - Insulin administration (redistribution)
  - Hungry bone syndrome:
    post–parathyroidectomy
    post–thyroidectomy.

Note
If hypomagnesaemia is associated with a urinary excretion rate > 0.5 mmol/day then renal wastage is indicated. Levels < 0.5 mmol/day suggest an extrarenal cause.

Developed by N. Walmsley 1995

Malabsorption

The aetiology of malabsorption can be considered under four headings:

- Mucosal defects
  - Coeliac disease, the commonest cause
  - Crohn's disease
  - Severe lactose intolerance
- Pancreatic disease
  - Cystic fibrosis
  - Chronic pancreatitis
- Post–infectious malabsorption
  - Giardia
  - Tropical diarrhoea
- Previous gastrointestinal surgery on stomach or small bowel.

Malabsorption particularly involves iron, folic acid, vitamin B12, vitamin K, vitamin D and fats.

Basic screen tests
- Blood count, looking for macro–or microcytosis
- Iron studies
- B12 and folate
- Stool for Giardia and other pathogens
- Coagulation screen
- Gliadin and TTG antibodies for coeliac disease.

Follow–up tests
- Jejunal biopsy is definitive in the diagnosis of coeliac disease.

Malaria

Specimen: Whole blood – EDTA

Malaria is endemic in:

- The Pacific west of Fiji. This includes Papua New Guinea, the Solomon Islands, Indonesia. It does not include Fiji, Samoa, Tonga, New Caledonia, Niue, Tahiti or Hawaii.
- SE Asia and India
- Africa and S. America.

The diagnosis needs to be considered in any visitor to these areas with unexplained fever, sweats, headache, malaise, anaemia, abnormal liver enzymes.

Diagnosis is established by detecting parasites in thick and thin films. Serial blood specimens may be required. Leucopenia is usual but there can be leucocytosis. Variant lymphocytes are occasionally seen. Malaria can be accompanied by anaemia which may be caused by a haemolysis, marrow depression or hypersplenism.

P. falciparum causes severe, sometimes lethal, illness.

P. vivax is less severe but can relapse.

P. ovale and *P. malariae* are the two less commonly found subtypes.

Treatment should be discussed with those who have experience in treating malaria.

Malassezia furfur

A fungus causing tinea versicolor (pityriasis versicolor) which show as brown scaly patches on white skin or pale patches on dark skin. Microscopy of skin scrapings shows grape–like clusters of cells. The organism is not readily cultivated. Diagnosis is based on clinical features, direct microscopy and absence of fungal growth on culture.

Treatment is with a topical imidazole, ciclopiroxolamine or terbinafine. Oral terbinafine is not effective.

Mantoux Test

Method
The standard Mantoux test consists of an intradermal injection of 10 units of tuberculin (PPD–purified protein derivative) contained in 0.1 mL of solution. The injection is made into the anterior aspect of the forearm and read three days later (2–4 days are in the outer limits if 3 days is impracticable).

When reading the test, the arm is palpated to define an area of induration and the diameter of this induration, if present, is measured in mm transverse to the long axis of the forearm.

An area of surround erythema (redness), or erythema alone, is ignored.

If vesiculation (blistering) or necrosis (darkening or ulceration) of the indurated skin is present, these must be described as an important part of the result.

Interpreting results
A diameter of induration > 10 mm is described as positive and is consistent with current or past TB infection.

A strong positive in a person with active TB disease may be > 15 mm with vesiculation and necrosis.

Previous BCG vaccination is associated with induration up to 15 mm though usually < 5 mm.

In immunocompromised persons, the tuberculin reaction may be reduced or suppressed, even in the presence of proven infection.

False positive Mantoux results
These are usually due to operator error, particularly reading errors where the area of erythema rather than induration has been read.

False negative Mantoux results
Although a negative Mantoux usually indicates that a person has never been exposed to TB, there are important exceptions:

- The person had TB in the past but the immune response has faded
- Overwhelming infection or illness of any type, including overwhelming TB
- Patients with immune suppression – HIV, immunosuppressive drugs, sarcoidosis, renal failure, malignancy, malnutrition
- Acute viral infections, recent live virus vaccinations
- Neonates.

Measles Antibodies (morbilli, English measles, rubeola)

Specimen: Serum – Gel

Reference Range: Supplied with report

The test can be for immune status (IgG antibody) or acute illness (IgM).

IgM antibodies appear about two days after the rash and peak about 2 weeks later. IgM antibodies disappear after the acute illnesses. IgG antibodies persist for life.

Meloidosis Antibodies

Specimen: Serum – Gel

Reference Range: Supplied with report

MEN (Multiple Endocrine Neoplasia)

These are familial clusters of endocrine adenomas or hyperplasias:

- **Type I** Parathyroid adenoma (95%)
  Pancreatic islet cell adenoma or hyperplasia (80%)
  Pituitary adenoma (50%)
- **Type II** Medullary thyroid carcinoma (95%)
  Phaeochromocytoma (50%)
  Parathyroid adenoma/hyperplasia (40%).

Meningitis

Please see Neisseria meningitidis.

Mercury

Specimen: 24–hour urine (nil preservative) or random urine
Whole blood – Trace element tube.

Reference Range: Supplied with report

Most of the public interest in mercury toxicity centres on amalgam in teeth but studies have not shown harmful effects. After an exhaustive investigation, the U.S. Public Health Services concluded there is no serious health risk. Urine mercury levels in people with filled teeth are less than 5% of the stated acceptable limit.

Organic mercury compounds have been used as fungicides with widespread poisoning in Iraq in 1971 when bread was accidentally made from seed wheat preserved with methyl mercury. Mercury–contaminated waste in sea–water is passed up the food chain to reach its most concentrated levels in large predatory fish such as shark, tuna and swordfish. The Miranda Bay disaster in Japan in 1955 was due to industrial waste discharge being concentrated in edible fish.

Mercury as calomel in children's teething powders and laxatives was found to be the cause of acrodynia (pink disease) as late as the 1940s.

Mercury poisoning causes renal damage and neurotoxicity.

Methadone Screening

Specimen: Random urine (nil preservative)

Reference Range: Not detected

See *Drug Screen*

Methyltetrahydrofolate Reductase Gene (MTFHR gene)

Specimen: Whole blood – EDTA

See *Thrombophilia*

Microalbumin

See *Proteins, urine*
Albumin Excretion Rate (AER)

Microsomal Antibodies

Specimen: Serum – Gel

Reference Range: Not detected

See *Thyroid Antibodies*

Molecular Genetics – Genetic Studies

Increasing numbers of inherited diseases are now identifiable using genetic studies.

Tests are available or are being developed for:

- Fragile X syndrome
- Myotonic dystrophy
- Prader Willi syndrome and Angelman syndrome
- Huntington disease
- Spinocerebellar ataxia
- Familial adenomatous polyposis
- Multiple Endocrine Neoplasia Type 2
- Spinocerebellar muscular atrophy
- Spinal muscular atrophy
- Duchenne muscular dystrophy
- Cystic fibrosis
- Breast cancer testing
- Adrenoleukodystrophy
- Hereditary haemochromatosis.

Morphine Screening (drug screen)

Specimen: Random urine (nil preservative)

Reference Range: Not detected

See *Drug Screen*

Mumps Antibodies

Specimen: Serum – Gel

Reference Range: Supplied with report

Interpretations:	IgM	IgG
Current or recent infection	+	+
Previously infected, now immune	–	+
Never infected	–	–

The diagnosis of mumps is usually based on clinical grounds and does not necessitate a blood test.

Serum amylase levels rise sharply.

Myasthenia Gravis

Specimen: Serum – Gel

This is an autoimmune disorder in which neuromuscular transmission is reduced to a varying extent by acetylcholine receptor autoantibodies (anti–ACLR) which are detectable in about 80% of cases.

Diagnostically the Tensilon (edrophonium) test is used, an injection of anticholinesterase which restores neuromuscular function.

See *Acetylcholine Receptor Antibody*

Mycobacterium avium (MAC)

Specimen: Sputum

Mycobacterium avium complex (MAC) infection is seen predominantly, but not exclusively, in patients with HIV infection. In that group it usually presents with dramatic fever spikes, rigors, malaise, often hepatosplenomegaly, anaemia and cholestatic liver function tests.

Otherwise normal children may also present occasionally with cervical adenopathy due to MAC. It is sometimes recovered from sputum specimens sent to TB culture. Although it can cause true respiratory tract disease it is often just a coloniser in an abnormal respiratory tract. Evidence for its being a pathogen includes repeated isolation, a positive smear result and CXR changes. Treatment regimens include ethambutol and clarithromycin with or without rifabutin. Decisions on whether to treat and with what, require specialist advice.

Mycoplasma pneumoniae

Specimen: Paired sera – Gel

Mycoplasma is a bacterium not demonstrable with usual stains or growth media. It is the commonest cause of atypical pneumonia, particularly in children and young adults, and can spread as a low–grade epidemic. It responds well to erythromycin or tetracycline but not to penicillin.

Diagnosis is by exclusion, by response to therapy, and by demonstrating a rising titre in paired sera, the first collected as soon as possible in the illness, the second 2 weeks later. Not infrequently the antibody titre has already plateaued by the time the first specimen is collected.

See *Pneumonia*

Myelodysplastic Syndromes (MDS)

Also known as evolving leukaemia or preleukaemia.

These conditions are primary bone marrow disorders which usually behave as slowly evolving bone marrow failure syndrome. Most often they present insidiously and in an older population. A variety of peripheral blood findings may be noted including pancytopenia, refractory anaemia, macrocytosis with anaemia, neutropenia and thrombocytopenia. Diagnosis is by excluding vitamin deficiency (B12, folate) and demonstrating dysplastic features in the bone marrow. Chromosome analysis and cell

marker studies may also be performed on the bone marrow. Management is usually by supportive care but in selected patients more aggressive therapy may be indicated.

Myelofibrosis

Myelofibrosis is a chronic myeloproliferative disorder characterised by bone marrow fibrosis, extramedullary haemopoiesis, splenomegaly and a leucoerythroblastic blood film. The condition may present as myelofibrosis at the outset or be the end stage of another myeloproliferative disorder such as polycythaemia vera or essential thrombocytopenia. Survival varies with a median of four years. Therapy includes blood transfusions, cytoreductive therapy (usually hydroxyurea) for thrombocytosis, and splenectomy in selected cases.

See *Myeloproliferative Disorders*

Myeloproliferative Disorders (MPD)

These conditions are characterised by excessive production of erythroid, myeloid and megakaryocytic cell lines (panmyelosis). In many situations only one cell line may predominate, e.g. platelet proliferation causing essential thrombocythaemia. In polycythaemia rubra vera (primary polycythaemia), erythroid hyperplasia is the most prominent feature with variable increases in myeloid cells (neutrophils) and platelets. There may also be excessive proliferation of reticulin and fibroblasts within the bone marrow giving rise to myelofibrosis. Acute myeloid leukaemia can develop as a terminal event, usually untreatable, in any of the MPDs.

Myoglobin

Specimen: 10 mL random urine

Reference Range: Not detected

Myoglobinuria occurs after skeletal muscle trauma or myocardial infarction (MI). After major trauma, myoglobin imparts a coffee colour to urine but small amounts, as in MI, are detectable only by chemical tests.

See *Troponin*

Mysoline (Primidone)

Specimen: Plasma – Lithium heparin

Reference Range: Supplied with report

Mysoline is metabolised to phenobarbitone which is responsible for much of the drug's anticonvulsant activity. Mysoline drug levels are monitored as phenobarbitone.

N

Needlestick Injuries

Investigations
Bloods from the exposed workers and the source specimen should be tested for markers of infection and immunity.

- HIV antibody/antigen
- HBs antigen
- HBs antibody
- HCV antibody.

The event must be fully documented and the exposed worker counselled and kept informed.

Prevention
All health care workers should be immunised against HBV.

Venepuncturists and nurses giving injections should be trained in safe practice.

Needles and other blood–contaminated sharps must be disposed of into puncture-proof containers.

Neisseria

These gram–negative diplococci are typically found inside neutrophils in direct smears from lesions. The two main pathogens are *N. gonorrhoeae* and *N. meningitidis*.

N. catarrhalis, a member of the normal flora of the throat, was known as *Branhamella catarrhalis* and is now known as *Moraxella catarrhalis*. It is a recognised respiratory tract pathogen.

A. Neisseria gonorrhoeae

Specimen: Bacterial swabs must be placed in transport medium immediately after collection. Store room temperature, not in the fridge, and transport to the laboratory as soon as possible–they will survive 24 hours in transport medium. In general, Neisseria are sensitive to drying, sunlight, heat, cold and many disinfectants. PCR is available and can be done off fist void urine and *ThinPrep*. Please indicate specific testing required on request form.

The main infections are:

Male STD	Urethritis and sometimes epididymitis. The organism can also sometimes be recovered from the rectum and throat.
Female STD	Endocervicitis is the initial infection with variable spread upwards to fallopian tubes and downwards to urethra and perianal skin. As with males, gonococci also grow in the throat and rectum.
Neonatal conjunctivitis	Acquired during birth from an infected mother.
Gonococcal arthritis	A suppurative arthritis. Synovial fluid aspirate establishes the diagnosis.

Treatment
As with any STD, treatment of partners is essential. Concomitant infection with Chlamydia is common and empirical treatment for this is recommended e.g. Ig azithromycin stat.

B. Neisseria meningitidis
The causative organism of meningococcal meningitis and septicaemia which can occur sporadically or in epidemics. Travellers to countries where epidemics occur, e.g. Nepal, should be vaccinated according to recommendations current at the time of travel. There are three main sub–groups: A, B and C.

Suspected meningococcal meningitis is a medical emergency and in the community situation, parenteral antibiotics should be given by the diagnosing doctor before sending the patient to hospital.

Neutrophil Alkaline Phosphatase (NAP score)

Specimen: Whole blood – Lithium Heparin

Reference Range: Supplied with report

Films must be made within a few hours of collecting the specimen. Although the test is of low specificity it is of some value in the following conditions:

Elevated NAP
- Polycythaemia vera (NAP up to 300)
- Myelofibrosis
- Infections
- Other inflammatory conditions
- Pregnancy.

Reduced NAP
- Chronic myeloid leukaemia
- PNH.

Neutrophil alkaline phosphatase activity is present in the specific granules of all myeloid cells with highest activity in the youngest neutrophils.

Neutrophils (Polymorphs)

Neutrophil counts are expressed and interpreted in terms of the absolute count rather than % of total white cell count.

Neutropenia
The adult reference range is 2.0– 8.0x10^9/L.

Mild neutropenia, 1.0– 2.0x10^9/L, is a common finding, often suspected to be caused by an immune–medicated mechanism (immune neutropenia) when drugs are not implicated.

Below 0.5x109/L there is an increase in spontaneous infection with a dramatically increased risk below 0.2x10^9/L (=agranulocytosis).

Common causes of neutropenia
- Viral infection, sometimes with an associated thrombocytopenia
- Some bacterial infections, e.g. typhoid
- Overwhelming sepsis, often with toxic granulation of neutrophils and a shift to the left
- B12 or folate deficiency
- Aplastic anaemias
- Malignancy – haematological malignancy or metastatic deposits in bone marrow

- Immune – mediated – post viral, Felty's syndrome, SLE
- Hypertension
- Thyrotoxicosis
- Chronic benign neutropenia, including cyclical neutropenia.

Drugs those more commonly implicated are:

- Phenylbutazone, indomethacin
- Chlorpromazine, promazine, other phenothiazines
- Carbamazepine
- Propylthiouracil, carbimazole
- Sulphonamides, cotrimoxazole
- Dapsone
- Thiazides
- Captopril.

Cyclical neutropenia

Characterised by periodic fluctuations in the neutrophil count with the nadir occurring usually at three weekly intervals. In the most severe cases the neutropenia may be extremely low, < 0.2x109/L, with infective complications, malaise and fever. Hospital admission may be required, as occasionally these may be life–threatening events. In some patients, due to the severity of the neutropenic episodes and the infective complications, growth factor support with granulocyte colony stimulating factor (G–CSF) is required.

Neutrophilia

A neutrophil count > 8. $0x10^9$/L (in non-pregnant adults)

Minor degrees of neutrophilia are a common response to almost any disease process. In more serious disorders, neutrophil precursors may be present, the sequence from cell division to maturity being myeloblast, myelocyte, metamyelocyte, band neutrophil, segmented neutrophil. An increase in band neutrophils, meta–myelocytes and eventually myelocytes, i.e. a "shift to the left", should prompt a search for serious disease.

Causes of neutrophilia

- Bacterial infection – there may be a shift to the left and with more severe infections there may be toxic granulation and eventually neutropenia as the marrow is overwhelmed. At the other end of the scale is the leukaemoid reaction, a massive outpouring of cells in response to infection with counts up to 80x109/L and a striking shift to the left.
- Tissue injury – due to infarction, burns, surgery, trauma and other processes causing necrosis
- Other inflammatory disorders – connective tissue disorders, etc
- Malignancy
- Drugs – steroids, epinephrine, heparin
- Other – haemorrhage, stress, metabolic disorders.

Nipple Secretions for Cytology

These specimens should be collected by applying the slide directly to the nipple and smearing secretion onto the slide.

If the secretion is scant the slide can be lightly touched against the nipple several times.

A mixture of air–dried and wet–fixed slides is appropriate. Slides can be fixed using a spray fixative or by immersion in 95% alcohol for 20 minutes immediately after collection. Air–dried slides should be dried rapidly (rapidly waving the slide backwards and forwards through the air ensures rapid drying). The slides must be marked as to whether they are air–dried or wet–fixed, as laboratory preparation and staining differ for each.

Labelling the slides on the frosted end of the slide with pencil, indicating the patient's family name, first name and date of birth.

O

Oestradiol (E2, Oestrogen)

Specimen: Serum – Gel

Reference Range: Supplied with report

- Oral contraceptives are associated with very low levels
- Pregnancy is associated with very high levels.

The assay is specific for oestradiol. Other oestrogens such as ethinyl oestradiol or conjugated equine oestrogens (Premarin) are not detected.

Oestradiol is the major endogenous oestrogen is ovulating women. It rises to a pre–ovulatory peak mid–cycle, falls, and then returns to a luteal phase plateau before falling again prior to menstruation.

Indications for estimating oestradiol are strictly limited:

- During artificial induction of ovulation
- Investigation of feminising states including precocious puberty, gynaecomastia
- Investigation of possible pituitary–gonadal deficiency states in women.

It is not used during the early investigation of infertility, for diagnosing menopause, for monitoring HRT (hormone replacement therapy), or for investigating irregular menstruation.

Oestriol (E3)

Specimen: Serum – Gel

Reference Range: Supplied with report

Unconjugated serum oestriol is used when testing for Down's syndrome during the first or second trimester.

See *Pre-Natal Testing*

Opiates Screening (Drug Screen)

Specimen: Random urine (nil preservative)

Reference Range: Not detected

See *Drug Screen*

Osmolality

Specimen: Serum – Gel
Urine, early morning, or random, or timed collect, or 24–hour urine.

Reference Range: Serum 275–295 mosmol/kg water
Urine 50–1500 mosmol/kg water

Urine osmolality, which is under the control of pituitary ADH, varies over a wide range to ensure that alterations in fluid intake have little effect on the tightly controlled serum osmolality. Urine osmolality is typically highest on rising in the morning because of the absence of fluid intake during sleep. An osmolality > 600 (especially > 800) makes diabetes insipidus unlikely.

An approximate serum osmolality is given by the formula:

Osmolality = 2 x Na+ + urea + glucose

The normal physiological response to hyponatraemia is the secretion of dilute urine with osmolality < 100. A higher osmolality suggests SIADH.

Osteocalcin (Serum Bone GLA protein)

Specimen: Serum – Gel
Spin, separate and freeze serum immediately.
Suggest collection of specimen at your nearest Collection Centre.

Reference Range: Supplied with report

P

Paget's Disease of Bone

Asymptomatic Paget's is a common cause of elevated ALP (150–500 units or higher) in the elderly and the ALP level is an indication of disease activity and response to treatment. 24–hour urine N–telopeptide is another indicator used in monitoring treatment.

A bone scan is a sensitive method for identifying foci of Paget's disease.

See *Alkaline Phosphatase (ALP)*

Pancytopenia

A reduction in all three cellular elements in blood i.e. anaemia, leucopenia (neutropenia) and thrombocytopenia. An unexplained pancytopenia is always an indication for a bone marrow biopsy.

Causes include:

- Haematological disorders requiring bone marrow examination – aplastic anaemia, leukaemias, myeloma, myelodysplastic disorders, metastatic tumour
- Drug effects
- B12 or folate deficiency
- Hypersplenism
- SLE, PNH
- Overwhelming infection, HIV.

PAP Smear

The quality of the Pap smear specimen is of critical importance if false negative cytology reports are to be minimised.

There are two main reasons for the occurrence of false negative Pap smear reports. One is laboratory error and the other is sampling error. At Capital Pathology we have put in place numerous quality assurance measures to minimise laboratory false negatives. Sampling errors can be reduced by taking an optimal sample. This also helps the laboratory in the interpretation of the smear.

See *Cervical Cytology*

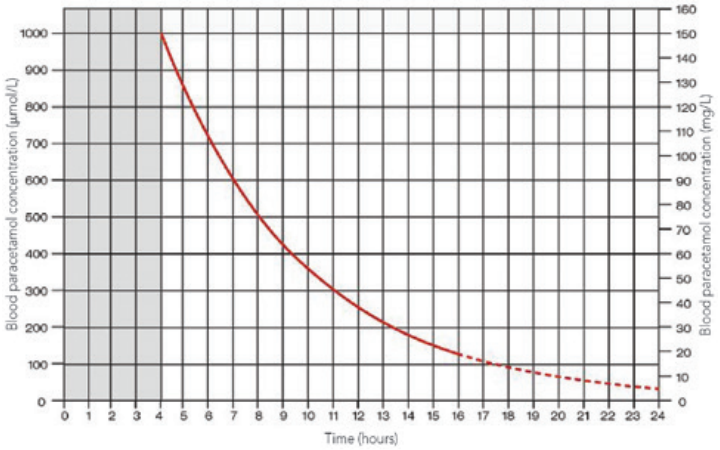
Paracetamol (Acetaminophen)

Specimen: Serum – Plain clot or Lithium heparin
Do not use gel tube.

Serum paracetamol levels are used to assess the need for *N*-acetylcysteine administration in all patients with deliberate paracetamol self-poisoning, regardless of the stated dose.The recommendations for the management of paracetamol poisoning in Australia and New Zealand are derived from consensus guidelines, that include the treatment nomogram shown.

The nomogram uses a single line to simplify decision making, and it uses the treatment threshold with the most clinical data to support its efficacy and safety.

New treatment nomogram for paracetamol poisoning



Daly FFS, Fountain JS et al. Guidelines for the management of paracetamol poisoning in Australia and New Zealand — explanation and elaboration. Med J Aust 2008; 188(5): 296-302. © Copyright 2008. *The Medical Journal of Australia* – reproduced with permission.

Parathyroid Hormone (PTH)

Specimen: Serum – Gel
Spin, separate and refrigerate serum.
Freeze if to be kept overnight.

Reference Range: 1.0–6.0 pmol/L

Elevations of PTH with elevated calcium:

- Primary hyperparathyroidism due to solitary adenoma (85%) or hyperplasia (15%) is the commonest cause.
- Lithium therapy
- Familial benign hypocalciuric hypercalcaemia is a rare autosomal dominant disorder in which excessive renal calcium reabsorption is associated with relatively small elevations in PTH. Parathyroid surgery is contraindicated.

Elevations of PTH without elevated calcium:

- Renal failure–2° hyperparathyroidism
- Vitamin D deficiency.

Paroxysmal Nocturnal Haemoglobinuria (PNH)

Specimen : Whole blood – EDTA x 2

Reference Range: Supplied with report

An absence of CD59 activity on the cell surface of red cells characterises Paroxysmal Nocturnal Haemoglobinuria (PNH).

PNH is a rare, acquired haematological disorder usually presenting in adult life.

Its features include:

- Intravascular haemolysis
- Venous thrombosis
- Marrow hypoplasia.

PNH is due to an acquired mutation giving rise to a clone of abnormal stem cells affecting erythrocytes, leucocytes and platelets.

The haemolysis, though chronic, is intermittent, occasionally paroxysmal with severe anaemia. It is mediated by complement and is made worse by reduction in blood pH as occurs at night, during exercise, or in vitro – as in Ham's test which is used in diagnosis.

The nonspecific laboratory abnormalities include:

- Anaemia, neutropenia, thrombocytopenia
- Reduced haptoglobins, reticulocytosis
- Haemoglobinuria, haemosiderinuria.

More specific tests for PNH are:

- Absence of CD59 activity on the erythrocyte cell surface
- Ham's acidified serum lysis test
- Sucrose lysis test
- CD59.

Pasteurella multocida

A gram–negative bacillus that is isolated from skin lesions. The organism is a common inhabitant in the mouths of cats and dogs and infection is frequently associated with animal bites or scratches. Regional lymphadenopathy, chills and fever can result. It is susceptible to penicillin and amoxycillin. Wound debridement may also be necessary.

Paternity Testing

Specimen: Buccal swab

DNA testing can provide parentage information for family reasons, immigration and forensic purposes.

Please note that these tests are not covered by Medicare.

For further information, please contact the Doctors Service Centre on 02 6285 9803.

PCR (Polymerase Chain Reaction)

A molecular method to amplify genetic material. The amplified sequence can then be detected by a variety of methods. Often used to detect small numbers of organisms in a specimen, e.g. *Toxoplasma gondii* in amniotic fluid or *M. tuberculosis* in CSF. PCR is also used to amplify sequences for human gene studies, e.g. paternity testing and some inherited genetic disorders.

Pemphigus Antibodies

Specimen: Serum – Gel

Reference Range: Not detected

Peripheral Neuropathy

Pathologies to consider include:

- Diabetes
- Viral infection: Guillain–Barré
- Alcoholism
- Connective tissue disorders
- Paraproteinaemias
- Toxins: lead
- Drugs: amiodarone, phenytoin, nitrofurantoin and others
- Nutritional disorders
- Vasculitis
- Deposition diseases e.g. amyloid
- Para-neoplastic
- Hereditary.

Pernicious Anaemia

An autoimmune disease, rare before the age of 40, causing destruction of gastric parietal cells and intrinsic factor and hence reduced absorption of vitamin B12. Laboratory features include some or all of the following:

- Anaemia, thrombocytopenia, neutropenia
- Serum vitamin B12 reduced
- Blood film features – oval (not round) macrocytes
- – hypersegmented neutrophils
- Macrocytosis – MCV up to 120
- Intrinsic factor and/or parietal cell antibodies
- – 90% are positive for one or both of these
- Folate – serum levels normal or increased
- Bone marrow – megaloblastic change
- Prominent increase in reticulocytes 10 days after commencing therapy with B12.

Pesticides

Specimen: Whole blood – Lithium heparin
Urine 20 mL random collection.

Reference Range: Supplied with report

Phenobarbitone

Specimen: Serum – Gel
Trough level is taken just before next dose (within one hour).
Peak level is taken 1–3 hours post dose.

Reference Range: Supplied with report

The half–life of phenobarbitone is 4 days.

See *Anticonvulsants*

Phenylalanine

Specimen: Blood spots on Guthrie card
Plasma – Lithium heparin for new born screening.

Monitoring PKU–quantitative phenylalanine levels is also done on blood spot specimens.

Phenytoin

Specimen: Serum – Gel
Trough level is taken just before next dose (less than one hour).
Peak level is collected between 4–7 hours post dose.

Reference Range: 40–80 umol/L therapeutic range adults

See *Anticonvulsants*

Phosphate

Specimen: Serum – Gel

Reference Range: Adults 0.80–1.50 mmol/L

Phosphate, also called inorganic phosphorus, is used diagnostically in two main situations:

1. Hypercalcaemia – levels are reduced or low normal in primary hyperparathyroidism and vitamin D deficiency, but are usually increased or normal in other hypercalcaemias
2. Renal failure – phosphate is elevated.

A wide range of other disorders cause hyper–or hypophosphataemia but diagnostic value is limited.

Phosphate levels can be falsely raised if the specimen is haemolysed.

Causes of Hypophosphataemia

- **Possible causes:**
- Hypercalcaemia
  - Associated with hyperparathyroidism or malignancy
- Aluminium antacid therapy
- Hyperalimentation
- Nutritional recovery
- Alkalaemia
- Treated diabetic acidosis
- Alcoholism
- Hypomagnesaemia.

Causes of Hyperphosphataemia

- **Possible causes:**
- Artefactual
-
  - Haemolysis
  - Delayed separation of plasma from RBC
- Renal failure
  - Phosphate rarely exceeds 4 mmol/L
  - Consider additional causes if phosphate > 4 mmol/L
- Infancy and childhood (Higher reference ranges)
- Increased intake
  - Vitamin D excess
  - IV therapy
- Cell leakage
  - Acidosis
- Hypoparathyroidism.

Platelet Antibodies

Specimen: Serum – Gel and Whole blood – EDTA

Reference Range: Supplied with report

Platelet antibody tests are most often performed in the investigation of an isolated thrombocytopenia. Both platelet–associated (PAA) and serum platelet (SPA) antibodies are measured. A positive result for either test supports an immune aetiology though immune thombocytopenia may be present in patients with negative PAA and SPA.

Platelet Count

Specimen: Whole blood – EDTA

Reference Range: 150–400 x 10^9/L

Reduced platelets – *see Thrombocytopenia*

Raised platelets – *see Thrombocytosis*

Platelet Dysfunction

This is suspected in patients presenting with a bleeding history and a normal platelet count, normal coagulation factor screen but prolonged bleeding time. The term "minimal bleeding disorders" is sometimes used. Occasionally, platelet aggregation and other studies are used where further investigation is required.

Platelet function disorders can be:

- **Congenital** rare defects of aggregation, adhesion or the platelet release reaction.
- **Acquired**
  - drug effects, e.g. NSAIDs
  - haematological malignancies
  - immune thrombocytopenia (ITP)
  - myeloproliferative disorders
  - renal failure.

Pneumocystis carinii

Pneumocystis carinii was considered to be a protozoan parasite but genetic studies suggest it is most likely related to the fungi.

It causes an acute to sub–acute, often fatal, pulmonary disease in the immunocompromised. It is a major disease problem in those infected with HIV.

Diagnosis is by detecting organisms in bronchial brushings, open lung biopsy and lung aspirates. A variety of stains may be used to show the organism.

Treatment is high dose cotrimoxazole and should be discussed with those who have experience in treating this condition.

Pneumonia

At the time of clinical diagnosis an attempt, often unsuccessful, should be made to obtain a sputum sample. Culture may show *Strep. pneumoniae* in classical lobar pneumonia, or *Haemophilus influenzae* and/or *S. pneumoniae* in pneumonia associated with chronic bronchitis.

Antibiotic therapy is usually started immediately on an empirical basis. A sputum specimen obtained after antibiotic treatment has started, will not be useful.

Atypical pneumonias
These are almost as common as the classical bacterial pneumonias.

- Mycoplasma pneumoniae is the commonest particularly in children and young adults
- Psittacosis affects those who have occupational contacts with birds, including poultry
- Legionnaire's disease – *See Legionella antibodies*
- Q fever
- Brucella
- Influenza.

For initial investigation (screen), the following tests are recommended: serology for Mycoplasma pneumoniae, Q fever, Legionella, Chlamydia pneumoniae, Chlamydia psittaci and influenza.

See *Chlamydia psittaci*
Chlamydia pneumoniae
Legionella antibodies
Mycoplasma pneumoniae
Q Fever antibodies

Polycystic Ovary Syndrome (PCOS)

This common endocrine disorder is characterised variably by:

- Amenorrhoea or oligomenorrhoea
- Infertility / anovulation with oestrogen present
- Hirsutism, acne, alopecia
- Mildly elevated testosterone – produced by the ovaries
- Increased extra–ovarian production of oestrogen
- LH elevated, FSH depressed, LH/FSH ratio > 2
- Prolactin elevated
- Obesity
- Ovaries enlarged with multiple cysts visible on ultrasound
- Insulin elevated due to insulin resistance
- Family history of PCOS.

Not all these features are necessarily present. The combination of androgen excess and obesity leads to increased extra–ovarian oestrogen production which increases LH and decreases FSH.

Polycythaemia

Defined as a haemoglobin concentration above the reference range for age and sex.

The approach to diagnosis is to distinguish between polycythaemias due to an increased red cell mass (primary and secondary polycythaemias) and those due to a reduced plasma volume (relative polycythaemias).

1. Polycythaemia rubra vera (primary erythrocytosis)
A myeloproliferative disorder usually occurring in middle or older age in which red cells, and often white cells and platelets, are increased.

Laboratory features
- Red cells – Hb and PCV are increased.
- The red cell mass is elevated (usually > 36ml/kg in males) but the plasma volume is normal. By convention the packed cell volume is used to monitor the treatment response.
- White cells – there is an increase in neutrophils often with a shift to the left with band neutrophils, metamyelocytes and occasionally myelocytes.
- Platelets – 400–800x10^9/L in most cases with counts over 1000x10^9/L occasionally. Giant forms may be present.
- NAP score – increased in 70%.

2. Secondary polycythaemia

Increase in the red cell mass due to tissue hypoxia caused by:

- Chronic respiratory disease
- Congenital heart disease
- High altitude.

Secondary polycythaemia also occurs in some renal disorders, particularly tumours, where there is increased erythropoietin production.

3. Relative or "stress" polycythaemia

The commonest form of polycythaemia, the elevated haemoglobin being secondary to a depletion in the plasma volume. By definition the red cell mass (volume) is normal. Clinical associations are:

- Smoking
- Alcohol
- Stress
- Dehydration
- Diuretics.

This condition is diagnosed by demonstrating a normal red cell mass and a reduced plasma volume.

Polydipsia / Polyuria

Urinary frequency due to UTI (urinary tract infection) or lower urinary tract problems, can be distinguished from polyuria by collection of a 24 – hour specimen to document urine volume.

Causes of polyuria include:

- Diabetes mellitus (random glucose levels usually above 15 when polydipsia/ polyuria are present)
- Hypercalcaemia
- Hypokalaemia
- Lithium therapy
- Psychogenic polydipsia (compulsive water drinking)–not uncommon
- Diabetes insipidus.

To differentiate between the latter two, see Diabetes insipidus.

Polymyalgia rheumatica

A relatively common disorder, particularly in the elderly, characterised by distressing shoulder and pelvic girdle pain originating from muscles rather than joints. It is often associated with temporal arteritis.

ESR and CRP are almost invariably (but not always) raised.

Response to steroids is dramatic.

Porphyrias

Specimens:	All specimens must be protected from light and delivered to the lab as soon as possible.
Urine:	During an attack of suspected acute porphyria, collect a random urine for PBG 24–hour urine.
Faeces:	Casual specimen.
Red cells:	EDTA tube

There are two main syndromes:

Acute porphyria

- Abdominal pain (autonomic neuropathy)
- Peripheral neuropathy
- Neuropsychiatric manifestations.

Attacks are provoked by drugs that induce liver enzymes – barbiturates, alcohol, oestrogens, sulphonamides, anticonvulsants, and others. In women, neuropathic symptoms may be cyclical, associated with oestrogen peaks.

Cutaneous porphyria

Presents with photosensitivity and blistering in sun–exposed areas, notably backs of hands, forearms, face, where accumulated porphyrins react with light to produce skin damage.

Symptoms are provoked and exacerbated by oestrogen, alcohol, and iron supplements.

Investigations

During a suspected acute attack, e.g. recurrent acute abdominal pain not otherwise explained, a fresh urine sample should be taken to the lab urgently for PBG.

During latent phases of acute porphyrias, abnormalities may be undetectable.

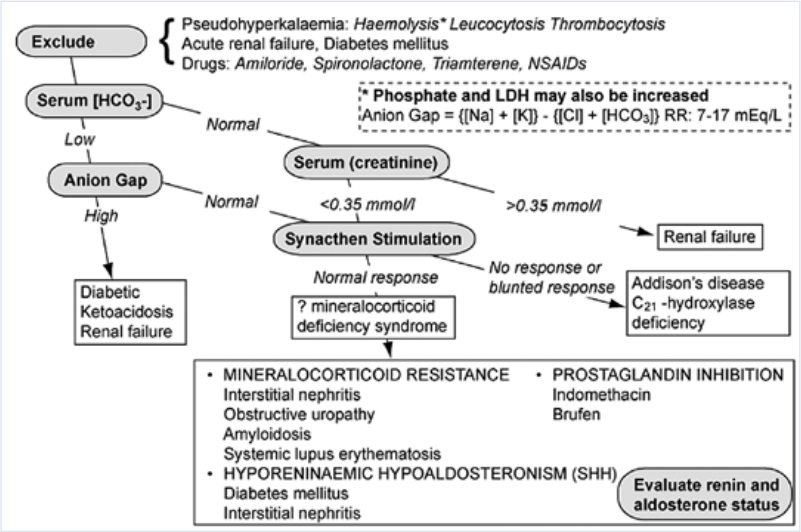
Cutaneous photosensitivity is most often due to PCT (Porphyria Cutanea Tarda) which is diagnosed by increases in urine and faecal porphyrins. Diagnosis of the rare erythropoietic porphyrias requires a blood specimen.

Potassium (K+), serum

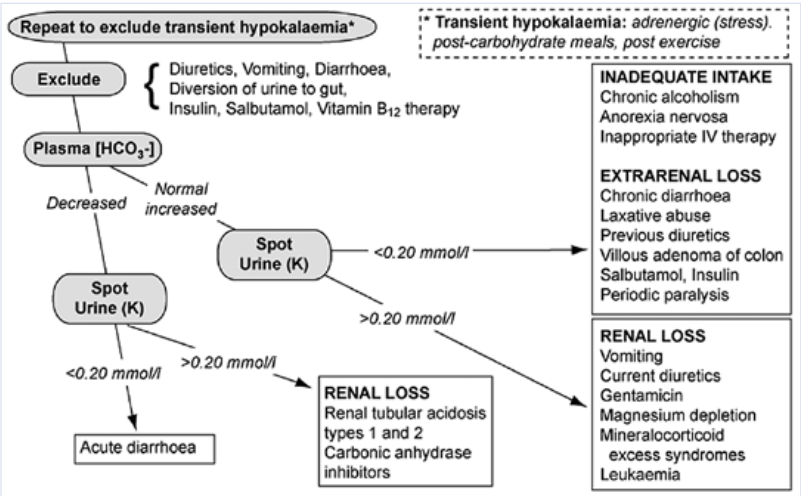
Specimen: Serum – Clot or gel

Reference Range: Adults 3.5–5.5 mmol/L

Hyperkalaemia



Hypokalaemia



Pre–Natal Testing

1. First trimester screening

A test is available to calculate the risk of Down's Syndrome and neural tube defects in the first trimester. The first trimester Down's Syndrome risk assessment combines measurement of serum free beta HCG (free BHCG), serum pregnancy associated placental protein A (PAPP–A), alphafetoprotein and oestridiol and measurement by ultrasound of nuchal fold thickness.

Together these parameters are able to give an adjusted risk for Down's Syndrome. The information from first trimester screening can be presented in a number of ways, however the maximal detection rate is achieved through the combined assessment of the biochemical markers and nuchal thickness.

Patient requirements for combined first trimester screen:

a) Serum for biochemistry collected from between 9 weeks until 14 weeks, 2 days inclusive.

b) Ultrasound performed from 11 to 13 weeks gestation.

The final combined risk assessment is provided by the radiology practice who performed the ultrasound.

2. Second trimester screening

The combination of maternal serum alpha–feto protein (AFP), unconjugated oestriol (uE3) and human chorionic gonadotrophin (HCG) taken at 15 to 20 weeks gestation together with gestational age, weight, maternal age, the presence or absence of diabetes and the previous obstetric history can be used in a calculation to obtain a risk assessment for fetal Down's Syndrome and open Neural Tube Defects.

An accurate estimate of gestational age is critical as circulating levels of all three serum markers vary with gestational age.

Assay results which may appear abnormal (and therefore high risk) for a patient with a gestational age of 18 weeks may be normal and therefore, low risk if, following ultrasound, the gestational age is determined to be 15 weeks.

Note: New or additional information can be put into the computer to give another assessment using the same assay value if, for instance, the gestational age is found to be different to that originally stated on the request form. No charge is made for this additional service.

The test is solely a SCREENING TEST and indicates the need for a more specific test (ultrasound, amniocentesis or chorionic villous sampling) to determine the outcome of the pregnancy.

The cut–off for a "screen positive" result requiring further assessment is arbitrarily set at 1 in 250. At a cut–off of 1:250, the test will identify approximately 60% of Down's Syndrome babies.

Neural tube defect risk assessment uses the AFP level to calculate the risk.

Comparison of Antenatal Screening Methods

Screening test	Gestation (weeks)	Down's Syndrome detection rate (5% FPR)
First trimester serum Biochemistry (free beta–HCG and PAPP–A) (FTS)	9–14 / 2 days inclusive	60
First trimester ultrasound Nuchal translucency (NT)	11–14	80
NT plus FTS	11–13	90
Second trimester Biochemistry (HCG, oestridiol and alphafetoprotein)	14–18	60–70
Second trimester ultrasound markers (e.g. echogenic cardiac foci, pyelectasis, nuchal fold thickening)	18–20	20–30 (low sensitivity and high FPR)

Reference : Australian Family Physician, Vol 32, No.3, March 2003, Advances in prenatal screening, pg 108

Primidone

Specimen: Plasma – Lithium heparin
Trough level is taken just before next dose (less than one hour).
Peak level is collected 1–3 hours post dose.

Reference Range: Supplied with report

Progesterone

Specimen: Serum – Gel

Reference Ranges: Follicular < 1.0 mmol/L
Luteal 3.7–45 mmol/L

- To detect ovulation a specimen is taken 6–9 days after presumed ovulation, i.e. days 20–23 of a 28–day cycle. Serum levels rise sharply during the luteal phase to reach a plateau.
- Because hormone release is episodic, levels can vary considerably throughout the day.
- Pregnancy – levels rise steadily to reach a peak of 200–700 mmol/L by term.
- Ectopic pregnancy – the diagnosis of ectopic pregnancy is supported by a level below 15 mmol/L in the presence of symptoms.

Prolactin

Specimen: Serum – Gel

Reference Ranges: Adult female < 540 mIU/L
Adult males < 390 mIU/L

Specimens should be collected at least 3 hours after awakening. Elevations of up to 1000 can usually be ignored or even up to 2000 in normally menstruating women. Repeat sampling 30 and 60 minutes after the first specimen will help show up elevations due to stress alone. Prolactin levels above 5000 are almost always associated with prolactinomas or pregnancy.

In women, hyperprolactinaemia is an important factor in infertility, amenorrhoea and galactorrhoea (though 2/3 of women with galactorrhoea have a normal serum prolactin).

In men, hyperprolactinaemia lowers testosterone levels and is an uncommon cause of impotence or galactorrhoea as well as infertility.

Causes of hyperprolactinaemia

Physiological
- Stress
- Sleep
- Pregnancy – starts to rise at 6 weeks, peaks at up to 10,000 mU/L by term
- Lactation – peaks occur during episodes of breast–feeding with levels up to 5000 mU/L. prolactin is essential for normal lactation.
- Early morning collection – trough levels occur late morning
- Eating – small rises.

Drugs
- Androgens
- Cimetidine
- Haloperidol
- Methyldopa
- Metoclopramide
- Oestrogens, including oral contraceptives
- Opiates
- Psychotropics
- Reserpine
- Tricyclic antidepressants.

Physiological
- Pituitary adenomas and micro–adenomas
- Other hypothalamic–pituitary disorders
- Polycystic ovary syndrome
- Hypothyroidism
- Renal failure
- Seizures
- Anorexia nervosa
- Addison's disease
- Hypoglycaemia
- Idiopathic.

Prostate Specific Antigen (PSA)

Specimen: Serum – Gel

Reference Range: Age related, total PSA

40–50 years	< 2.6 µg/L
60 years	< 3.6 µg/L
70 years	< 4.6 µg/L
80 years	< 6.6 µg/L

Risk Assessment–Summary of Approach

Consider the age–related risk of prostate cancer

The prevalence of prostate cancer, as demonstrated in autopsy studies, is up to 80% by the age of 80. The probability of clinical prostate cancer is under 1% for men at age 40 but rises progressively with age.

Consider the Total PSA

Assess PSA level according to age related reference values. A PSA greater than 20 µg/L is highly suggestive of cancer. If the PSA is less than 20ug/L but above normal for age, check the Free/Total PSA value. Remember – a normal PSA does not exclude cancer.

Consider the Free/Total PSA value

If the value is above 20% cancer is unlikely, though not excluded. If the value is less than 8% there is a high probability of cancer.

On the basis of this approach definitive diagnosis by multiple prostate needle core biopsies may be performed.

Further appropriate urology treatment and follow up can then occur.

Protein C

Specimen: Plasma – Sodium Citrate
Double spin, separate and freeze plasma.

Reference Range: Supplied with report

Deficiency of Protein C is an inherited cause of hypercoagulability and is found in about 5% of cases of venous thromboembolism. The incidence of the deficiency is 1:5000–1:5,000. Approximately half of these will develop a thrombosis by the sixth decade. Inherited deficiency is usually an indication for long–term anticoagulation.

Severe homozygous deficiency is associated with purpura fulminans in neonates.

Acquired causes of Protein C deficiency are liver disease, warfarin therapy, vitamin K deficiency.

Protein S

Specimen: Plasma – Sodium Citrate
Double spin, separate and freeze plasma.

Reference Range: Supplied with report

Deficiency of Protein S, a cofactor for activated Protein C, is an inherited cause of hypercoagulability and is found in up to 5% of patients with inherited thrombolic disease. The lower end of the reference range is not well defined.

Acquired causes of Protein S deficiency are liver disease, warfarin therapy, vitamin K deficiency, oral contraceptives, pregnancy.

Proteins, total, serum

Specimen: Serum – Gel

Reference Range: Adults 60–80 g/L

Total proteins, consisting of albumin and globulins, is a crude test, but a raised value can be a pointer towards raised immunoglobulins; a low value can be due to reduced albumin or globulins or both. Variations in protein concentration can be due to dehydration, diuretics, fluid retention or diurnal changes. On changing from the recumbent to the upright position, fluid is redistributed to tissues from the circulation causing an increase of up to 10% in protein concentrations.

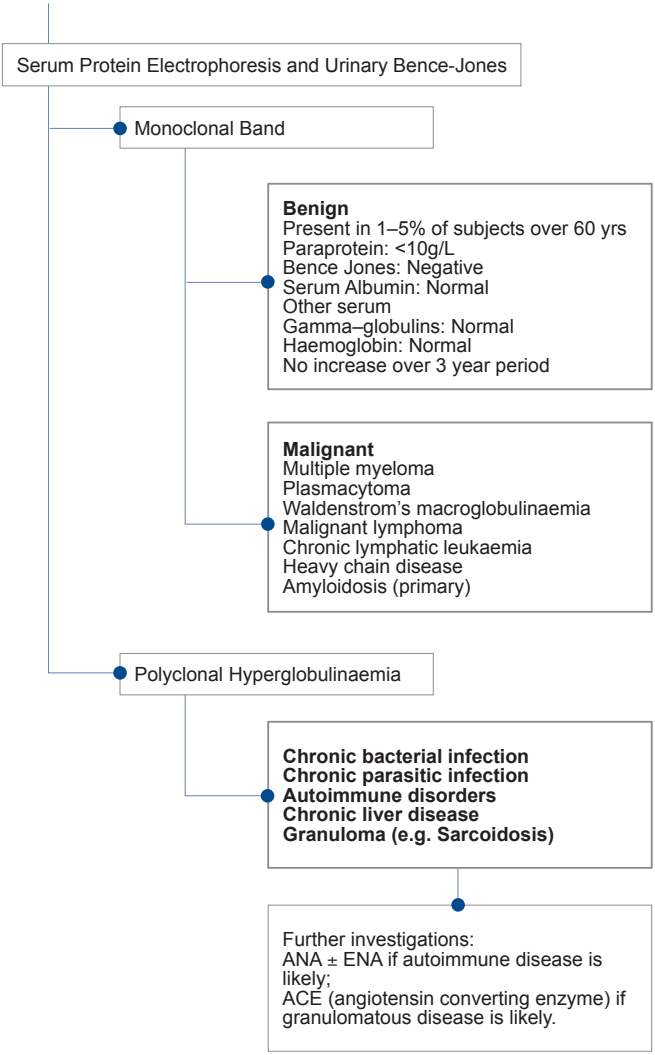
Protein–binding in serum

Many serum constituents exist in two forms, a protein–bound form, usually to an alpha or beta globulin or albumin; and a free form, often a tiny percentage of the total but this is the metabolically active form.

Examples of protein–bound constituents are thyroxine, tri–iodothyronine, cortisol, testosterone, vitamin B12, iron, copper, calcium–and many drugs.

Total globulins=total protein + albumin

Evaluation of Hyperglobulinaemia (Suggested scheme for evaluation of Hyperglobulinaemia)



Proteins, urine (Proteinuria)

Specimen: Random urine – Timed 10,12 or 24 hours

Reference Range: Supplied with report

Test methods

These depend on the clinical situation:

- *Urine dipstick*, as part of routine urinalysis
- *Total protein concentration on spot urine* – this gives a more accurate measurement than a dipstick and also detects non–albumin proteins such as free light chains.
- *24–hour protein quantitation*–this test is used as follow – up when a dipstick shows 1+ (or more) positive, or when monitoring known proteinuria.
- *Urine microalbumin* – a sensitive test measuring small quantities of albumin in the range 0.01–0.20 g/L. The test is used as an early indication of diabetic nephropathy.
- *Electrophoresis* of concentrated urine – used mainly in detection of free light chains (Bence Jones Proteins).

Renal proteinuria

Causes include the whole differential diagnosis of renal disease.

Glomerular proteinuria is by far the most common and serious type. The protein is predominantly albumin and when daily output > 3g/day, nephrotic syndrome develops, comprising oedema, albuminuria and hypoalbuminaemia. Serum lipids become elevated.

In *tubular* proteinuria, electrophoresis shows a non–selective pattern with bands representing the wide range of normal serum proteins normally reabsorbed by the tubules. Causes include drugs, inherited disease (e.g. Wilson's disease), autoimmune disease (e.g. SLE) or chronic infection.

Proteinuria of > 1.0 g/day requires renal investigation including consideration of renal biopsy.

Benign and non–renal proteinuria

40% of our routine urinalyses show a trace or more of protein and most of these are benign.

Causes include:

- Infection
- Fever
- Stress
- Exercise
- Heart failure
- Orthostatic proteinuria, found particularly in young men, disappears when the patient is recumbent. Protein is absent from a morning specimen collected on first getting up.
- Idiopathic

- 24–hour protein in these is usually < 0.5 g/day.
- Levels of 0.2–0.5 g/day (or even up to 1 g/day) cannot be described as normal but often remain of unknown aetiology when found as isolated abnormalities. Intensive follow–up is usually unrewarding.

Prothrombin mutation (20210G–A)

Specimen: Whole blood – EDTA

The prothrombin gene mutation 20210G–A, found in 1–3% of the population, is associated with levels of prothrombin which exceed those in the normal population by an average 30%.

Clinically the mutation is associated with:

- Thrombophilia
- 3–6 fold increase in VTE
- Cerebral vein thrombosis
- Myocardial ischaemia in patients age < 50.

Pseudomonas aeruginosa

In general practice, *Pseudomonas* is encountered mainly in chronic otitis externa where it usually responds to local toilet and topical polymyxin. The most commonly used agent in the past, Chloromyxin, which contained polymyxin B, is no longer available. Another ear preparation, Colymycin S Otic, containing colistin, which has the same spectrum of antibacterial activity as polymyxin, is currently the most widely used topical preparation for *Pseudomonas* otitis externa.

P. aeruginosa is often recovered from ulcer lesions on the lower limbs of the elderly. It is frequently a coloniser rather than a pathogen in this situation. If, however, the gram–stain reveals many WBCs with gram–negative bacilli and the ulcer has deteriorated clinically *Pseudomonas* may be the cause of infection. In this instance ciprofloxacin treatment may be indicated.

Pseudomonas can cause more serous infections in burns, in immunocompromised patients and in nosocomial infections. In these situations, early treatment with carefully selected antibiotics is required because of the organism's pathogenicity and resistance to a variety of agents.

Q

Q Fever Antibodies

Specimen: Serum – Gel

Reference range: Supplied with report

Queensland Tick Typhus Antibodies

Specimen: Serum – Gel

Reference range: Supplied with report

R

RAST (Radioallergosorbent Test)

Specimen: Serum – Gel

RAST tests use the patient's serum to diagnose specific allergies by identifying the presence of an IgE antibody against that allergen. Specify which allergens are clinically relevant. A general Alatop screen can be performed, as well as a limited number of individual allergens per test episode.

Red Cell Folate

Investigation of serum and red cell folate levels has been useful for the investigation of macrocytosis, megaloblastic anaemia and nutritional status along with assessment of vitamin B12 stores. Whilst red cell folate provides an accurate assessment of tissue folate status, serum folate is of little diagnostic value since it can be reduced with a short term decrease in dietary intake or in systemic illness. For these reasons Capital Pathology will no longer be performing serum folate as part of B12 and folate assessment. Red cell folate measurement will continue, and should now become the main indicator for assessment of folate status.

Reducing Substances in Faeces

Specimen: Fresh sample of faeces delivered to lab as soon as possible, stored in fridge if delay is unavoidable.

Reference Range: Not detected

The test is a simple qualitative test for reducing substances which include lactose, glucose and fructose but not sucrose.

Acquired lactase deficiency, in which the disaccharide lactase is not digested, presents as frothy diarrhoea with failure to thrive in infants after infectious diarrhoea.

The diagnosis can be confirmed by improvement after withdrawal of lactose from the diet.

Renal Calculi

Most calculi are of unknown origin but four aetiologies need to be remembered:

1. Hypercalcaemia – check serum calcium, vitamin D
2. Uric acid excess in urine
3. Cystinuria
4. Mixed "triple–phosphate" stones, often due to chronic, occult urine infections with gram–negative bacteria such as Proteus.

The reason for analysing calculi is to detect pure uric acid (5–10% of all stones) or cystine. In the case of uric acid stones, the causes of increased uric acid excretion need to be looked for. Allopurinol is effective in preventing further stone formation.

The remaining 90% of stones are composed of calcium oxalate or phosphate and in half of these there is hypercalciuria due to increased intestinal absorption of calcium. A 24–hour urine should be analysed for calcium, phosphate, urate and oxadate.

Renin

Specimen: Plasma – EDTA x 2
Spin,separate and freeze plasma within 2 hours of collection.

Reference Range: Supplied with report

Reticulocyte Count

Specimen: Whole blood – EDTA

Reference Range: Adults absolute count 20–100x10^9/L

Absolute counts are preferred to percentages because they are independent of anaemia. Polychromasia is the morphological indicator of increased reticulocytes.

The reticulocyte count is increased in:

- Chronic or acute blood loss
- Haemolytic anaemias
- Deficiency anaemias treated with iron, B12 or folate (treatment response)
- Marrow infiltration.

A reduced reticulocyte count suggests a hypoplastic basis for an anaemia.

Rheumatic Fever

Diagnosis is based on the revised Jones criteria:

Major manifestations
- Carditis
- Polyarthritis
- Chorea
- Erythema margination
- Subcutaneous nodular

Minor manifestations
- *Clinical*
  - previous rheumatic fever
  - or rheumatic heart disease
  - or arthralgia
  - fever
- *Laboratory*
  - raised ESR, or CRP, or WBC
- *ECG*
  - prolonged P–R interval.

Evidence of streptococcal infection
- Increased streptococcal antibodies
- Positive throat culture of GpA streptococcus
- Recent scarlet fever.

The diagnosis requires:

- Two major criteria

or

- One major and two minor criteria.

plus evidence of streptococcal infection

Absence of evidence of streptococcal infection makes the diagnosis doubtful. A serum sample should be collected for streptococcal antibodies whenever the diagnosis or rheumatic fever is suspected and a second sample collected at least two weeks later, looking for rising titres.

See *Streptococci*

Rheumatoid Arthritis (RA)

The commonest of the connective tissue diseases with an incidence of 1–2% in the general population. Patients may show:

- Rheumatoid factor (positive in 80%)
- Morning stiffness
- Arthritis of hand joints
- Symmetric arthritis
- Arthritis of three or more joint areas (of 14)
- Rheumatoid nodules
- Radiographic changes.

Note that 20% of patients with rheumatoid arthritis are negative for rheumatoid factor.

See *Rheumatoid Factor*

Rheumatoid Factor (RF)

Specimen: Serum – Gel

Reference Range: Supplied with report

Rheumatoid factor is an antibody, or rather a group of IgM, IgG and IgA antibodies, directed against IgG. Most methods detect the same IgM rheumatoid factor but specificities vary somewhat and for this reason a test that is positive in one laboratory may be negative in another and the quantitations may differ.

2% of the healthy population are positive for RF, rising to 5–10% in the elderly.

In rheumatoid arthritis, RF is present in only 80% of cases. RF on its own does not signify rheumatoid arthritis and conversely a clear clinical presentation of RA does not require RF to establish the diagnosis. RF is often absent during the first 6 months of disease. Higher levels tend to be associated with more aggressive disease.

Conditions, other than RA, with a high incidence of RF are:

- Sjogrens (90%)
- SLE (30%)
- Other connective tissue disorders
- Chronic inflammatory/infectious disorders
- Malignancy.

Rickettsial Serology

Specimen: Serum – Gel

Reference Range: Supplied with report

Ross River Virus Antibodies

Specimen: Serum – Gel

Reference Range: Supplied with report

Rotavirus

Specimen: Fresh faeces

Rotavirus is the commonest cause of diarrhoea in young children – 50% of children under five hospitalised for gastroenteritis are infected with rotavirus. Essentially all children are infected by the age of three with the peak incidence between 6 and 24 months. Adult infections are much less frequent. There is a clear winter peak. Spread is by the faecal–oral route. It has a short incubation period, 24–72 hours.

Symptoms may be severe but usually settle within 7 days with oral fluid and electrolyte replacement.

Rubella Antibodies

Specimen: Serum – Gel

Reference Range: Supplied with report

Tests: IgG for immune status
IgM for current infection.

Immune Status

The person with an IgG antibody level > 10 u/ml, can be assumed to have immunity.

With levels between 5 and 10, immunity may be present.

Below 5, immunity is either non–existent or slight and revaccination may be indicated, though not in pregnancy where immunisation should be delayed until after delivery.

It is essential that pregnant women found to be non–immune be vaccinated after delivery.

Infection during pregnancy

Because 85% of mothers infected during the 1st trimester give birth to infants with congenital defects (eyes, ears, heart, brain), termination of pregnancy must always be discussed when infection has been established.

The mother who has had contact with suspected rubella and whose immune status is susceptible or unknown, should immediately have blood collected for IgM and IgG antibodies with at least one more specimen three weeks later.

S

Salicylate

Specimen: Plasma – Lithium Heparin
Optimal collection time is just prior to next dose.
Please supply time of last dose.

Reference Range: Supplied with report

Salmonella Antibodies (Widal Test)

Specimen: Serum – Gel

A test of limited value in the diagnosis of typhoid fever, blood culture being the preferred diagnostic method.

Interpretation:

O antigens	titre > 1:160 and rising sharply over 7–14 days suggests current infection
H antigen	> 1:160 suggests immunity
Vi antigen	high titre sometimes indicates the carrier state

Titres can be nonspecifically elevated in diseases not caused by Salmonella and vaccination can give positive titres.

Salmonella Culture

Specimen: Faeces for diarrhoea
Blood culture for suspected typhoid fever.

Enterocolitis
Salmonella species make up 15% of the pathogens cultured from faeces in this laboratory, the main reservoir being domestic animals (including poultry and eggs) and infected humans, both symptomatic and carriers. Species causing enterocolitis include *S. typhimurium*, *S. choleraesuis* and *S. enteritidis*. In healthy persons the episode usually resolves in 2–3 days. Patients with impaired defences may require antibiotic treatment, usually with cotrimoxazole, amoxycillin or ciprofloxacin. The chronic carrier state can be treated with ciprofloxacin.

Typhoid fever
During the early stages, diagnosis is by blood culture which should be done repeatedly when clinical suspicion is strong. From the second week on, faeces cultures may be positive.

Sarcoidosis

A systemic disorder of unknown cause characterised by non–caseating granulomata. Diagnosis usually requires histological examination of lung, skin, lymph nodes or liver.

Serum ACE (Angiotensin Converting Enzymes) is elevated in 2/3 of patients but the test is not specific enough to be of diagnostic value, nor does it have prognostic significance.

Serum calcium may be elevated.

See *Angiotensin Converting Enzyme (ACE)*

Scabies

Caused by the mite *Scarcoptes scabiei* and transmitted by direct skin to skin contact. Transfer from clothes is possible but only if worn by infected people immediately beforehand.

Incubation period is 2–6 weeks without previous exposure but only 1–4 days in those who have been infected before.

Burrows are characteristic in scabies. These consist of skin–covered ridges 0.5–1.0 cm in length, often with a small vesicle at the end.

The diagnosis is usually made clinically. Treatment is usually with permethrin 5% cream. Gamma benzene hexachloride 1% cream or lotion, and crotamiton 10% cream or lotion, are alternatives. Pruritis may persist for some time after successful treatment.

Schistosomiasis

Specimen: Urine, full volume preferably collected between noon and 4pm when there is peak egg excretion.

Serum for antibody tests.

Request examination for schistosome ova. Eggs may be found in urine and stool as early as five weeks after infection. Patients with a low worm burden may have few or no eggs in urine or stool.

Although over 200 million people in Africa, the Middle East, S. America and the Caribbean are infected with these blood flukes, schistosomiasis is rare in travellers to these areas. Infection requires skin contact with contaminated fresh water as in swimming or wading barefoot in paddy fields, waterholes, local streams, etc. The infective form penetrates human skin, passes through a migratory phase in the lung and liver and then moves to its final habitat in the portal venous system (*S. mansoni*) or urinary bladder venous plexus (*S. haematobium*). Adult male and females live for 5–10 years.

Many travellers have acquired infection from Lake Malawi in South East Africa. It is stated in some travel guides that this lake is free of infection risk. This not the case and travellers should be advised against swimming in this lake.

The usual problem is the traveller who requires reassurance. Examination of faeces for the highly characteristic ova; urine for ova and red cells; serum for serology, and perhaps liver function tests, are adequate for this purpose.

Where more serious investigation is required, up to six specimens of urine and faeces should be examined. Further investigation might include appropriate tissue biopsies (rectum, bladder, liver).

Positive serology indicates present or past infection. Persons with negative stool and/or urine tests but positive serology require treatment to avoid the uncommon but catastrophic spinal cord complication of transverse myelitis.

Treatment with praziquantel and follow–up are best managed by those with experience treating schistosomiasis.

Scleroderma

Scleroderma Diffuse
Also called systemic sclerosis, this is a connective tissue disease characterised by diffuse cutaneous and/or systemic fibrosis.

Raynaud's phenomenon is present in 95% of cases. It has an incidence of about 1:50,000.

Tests that are often positive include:

- ANA (95%) usually with a nucleolar pattern
- RF (25%)
- ENA anti Sci–70 (30%).

Scleroderma Limited

- Previously called CREST syndrome
- Calcinosis
- Raynaud's phenomenon
- Esophageal dysmotility
- Sclerodactyly
- Telangiectasia.

Tests that are often positive include ANA (80%) usually centromere pattern.

Scoline (Suxamethonium) Sensitivity

This autosomal recessive disorder renders the carrier apnoeic for an abnormally long period after administration of scoline. Family members of an affected individual should be tested. The dibucaine number is an additional test used in identifying carriers.

See *Cholinesterase*

Selenium

Specimen: Serum – Whole blood – Trace element tube.
24-hour urine (nil preservative).

Reference Range: Supplied with report

Selenium is an antioxidant and is an essential trace element in humans and animals, entering the food chain through plants. In the Keshan province of China, an endemic cardiomyopathy has been attributed to the region's severe soil selenium deficiency.

Brazil nuts are a rich natural source of selenium.

Seminal Fluid

1. Post–vasectomy specimens

This is a test for sperm count only, not motility. The specimen should be delivered to the laboratory on the day of collection but without the urgency necessary for fertility specimens.

Most post–vasectomy specimens have a zero sperm count but a small percentage have a low count of non–motile spermatozoa even months after successful surgery. They are believed to be sequestered spermatozoa stored in the recesses of seminal vesicles or other parts of the male GU system.

2. Infertility specimens

The specimen should be delivered to the main laboratory within 2–3 hours of collection and preferably before 3pm. Many people collect the specimen at home at 7–8am and deliver the specimen before work. Please ensure specimen is kept warm. Patient information sheets are available from the main laboratory, collection centres and the website www.capitalpath.com.au

Serotonin (5–hydroxy tryptamine)

Specimen:	Serum – Gel or 24-hour urine (HCL preservative).
Reference Range:	Supplied with report

Sex Hormone Binding Globulin (SHBG)

Specimen:	Serum – Gel	
Reference Range:	Adult female	20–155 nmol/L
	Adult male	15–100 nmol/L

SHBG is a serum globulin that binds testosterone and, to a lesser extent, oestradiol. On its own, it has little diagnostic value but it is required when measuring the biologically active free testosterone fraction as distinct from the 98% of total testosterone which is bound and inactive.

SHBG is increased by: oestrogens, pregnancy, androgen deficiency, thyrotoxicosis and cirrhosis.

It is decreased by: obesity, hypothyroidism, androgen excess, polycystic ovary syndrome and hirsutism.

Sexually Transmitted Disease

1: Common presentations of sexually transmitted infections and recommended investigations*

Presentation	Possible causes	Investigations*
Urethritis (dysuria, discharge, frequency)	Common causes: Chlamydia trachomatis (commonly asymptomatic) Neisseria gonorrhoeae Trichomonas vaginalis Mycoplasma genitalium, Mycoplasma hominis, Ureaplasma urealyticum (importance as pathogens not clear: maybe associated with urethritis) Herpes simplex virus (HSV) types 1 and 2 (can cause intermittent, recurrent attacks of urethritis)	• First-catch urine (does not have to be first void of the day) for PCR for C. trachomatis (± N. gonorrhoeae) • Urethral bacterial swab for microscopy and culture (culture is necessary for antibiotic susceptibility testing of N. gonorrhoeae) • PCR is available and can also be done off *ThinPrep* specimen • Microscopy of wet preparation • Urethral viral swab for HSV PCR.
Cervicitis (usually asymptomatic)	C. trachomatis N. gonorrhoeae	• Urine, cervical or vaginal swab for PCR for C. trachomatis and N. gonorrhoeae • Cervical swab for microscopy and culture for N. gonorrhoeae
Vaginal discharge (usually not sexually transmitted)	Candida albicans (most common cause) Bacterial vaginosis (caused by changes in normal vaginal flora) T. vaginalis (sexually transmitted)	• High vaginal swab for microscopy and culture • Microscopy of wet preparation
Pelvic inflammatory disease (pelvic pain, menstrual irregularities, dyspareunia, may be silent)	STI-related C. trachomatis N. gonorrhoeae Non-STI-related Mixed pathogens — anaerobes, facultative bacteria and Mycoplasma spp.	• Urine, cervical or vaginal swab for PCR for C. trachomatis and N. gonorrhoeae • Cervical swab for microscopy and culture for N. gonorrhoeae • Pelvic ultrasound examination • Possible laparoscopy in severe cases
Genital warts (typical warty lesions in anogenital area)	Human papillomavirus (most commonly types 6 and 11; occasionally other types)	• Clinical diagnosis • Biopsy if atypical lesions or doubt about diagnosis
Anogenital ulceration	**Infective causes** • HSV-1 and HSV-2 • Syphilis (Treponema pallidum) • Donovanosis • Chancroid (Haemophilus ducreyi) • Lymphogranuloma venereum **Non-infective causes** • Aphthous ulceration • Behçet's disease • Trauma • Drug reaction • Inflammatory Bowel Disease	• Viral swab for viral culture ± PCR for herpes • Routine microsocopy and culture of swab specimen (H. ducreyi is difficult to culture on routine media) • Serological tests for syphilis • Biopsy for histopathological examination

PCR = polymerase chain reaction. LCR = ligase chain reaction. STI = sexually transmitted infection. * Routine screening for other STIs (including HIV) should also be offered to patients with any of these presentations.

Shigella

Specimen: Fresh faeces

Shigella, the cause of "bacillary dysentery", is predominantly found in third world countries where it is transmitted by contaminated food and water. Children under 5 years are particularly affected.

Mild cases are treated with oral rehydration alone but more severe infections may require antibiotics and IV fluids. Shigellosis is a notifiable disease.

S. I. Units (Systeme Internationale)

In 1975–77, Australian, New Zealand and UK laboratories switched from traditional mass units (mg/100ml) to SI units (mmol/L). United States laboratories have mostly retained mass units which may need to be converted when American patients present in Australia with results from back home.

	SI Unit	Mass Unit	Conversion Factor
Alcohol	1 mmol/L	= 4.5 mg/100ml	0.22
Bilirubin	1 umol/L	= .06 mg/100ml	17.1
Calcium	1 mmol/L	= 4 mg/100ml	0.25
Cholesterol	1 mmol/L	= 38.5 mg/100ml	0.026
Glucose	1 mmol/L	= 17.9 mg/100ml	0.056
Potassium	1 mmol/L	= 1 meq/L	1.0
Protein	1 g/L	= .1 g/100ml	10.0
Sodium	1 mmol/L	= 1 meq/L	1.0
Triglyceride	1 mmol/L	= 91 mg/100ml	0.011
Urate	1 mmol/L	= 16.7 mg/100ml	0.060
Urea	1 mmol/L	= 5.9 mg/100ml	0.17

To convert mg/100 mL to mmol/L: multiply by conversion factor

To convert mmol/L to mg/100ml: divide by conversion factor

An advantage of SI units is that osmolality (in mmol/L) of serum or urine can be simply calculated by adding concentrations (in mmol/L) of the constituents, assuming all the main ones have been measured.

Sickle Cell Test

Specimen: Whole blood – EDTA

Sickle cell haemoglobinopathies are hereditary disorders due to HbS, found in blacks of African descent. The homozygous state (sickle cell disease) causes a chronic haemolytic anaemia and during crises, episodes of pain and multisystem organ damage. Heterozygotes (sickle cell trait) are free of clinical disease and have normal red cell indices but HbS can be demonstrated by appropriate tests including Hb electrophoresis.

Sideroblastic Anaemia

This is an acquired disorder of porphyrin metabolism seen in older patients and associated with anaemia, either microcytic or dimorphic. It is a preleukaemic condition and is part of the myelodysplastic (MDS) spectrum of disorders. The hallmark is a marked increase in bone marrow iron, ring sideroblasts or Perl stain in the bone marrow and an elevated ferritin. The disorder may be primary (in MDS) or secondary to drugs, particularly antituberculous therapy.

Sjogren's Syndrome

A connective tissue disease characterised by salivary and lacrimal gland involvement resulting in dry mouth and dry eyes.

It may exist on its own or be associated with rheumatoid arthritis or other connective tissue disease.

Tests that are usually positive include:

- ANA usually with a speckled pattern
- RF (90%)
- ENA (60%) anti SSA (Ro) or SSB (La).

Skin and Wound Swabs

When collecting the swab, aim to get pus or exudate from the base of an actively inflamed lesion. Remove scab if present and open infected blisters. Avoid getting bacteria from normal skin. 80% of skin/wound/pus swabs in the community grow *Staph. aureus*. *Strep. pyogenes* makes up most of the remainder. The latter is the usual pathogen in impetigo and cellulitis.

Chronic leg and foot ulcers are commonly associated with vascular insufficiency, and, in the diabetic foot, with loss of pain and touch sensation. Swabs usually show a mixed growth of gram–positive cocci and gram–negative bacilli. Cellutitis confined to the rim of the ulcer can be treated with an oral agent such as amoxycillin–clavulanate but a spreading cellulitis, particularly in a diabetic foot, needs urgent admission for parenteral antibiotics.

All chronic ulcers require careful daily cleansing with warm saline and debridement of dead tissue down to a healthy base.

Skin Biopsy

Method

1. Excisional skin biopsy

This technique should be used for:

- Atypical pigmented lesions
- Deep dermal/subcutaneous nodules
- Where evaluation of margins is important
- Where panniculitis (inflammation of subcutaneous fat) is suspected.

2. Punch biopsy

This technique is useful for:

- Inflammatory dermatoses including alopecia
- Suspected basal or squamous cell carcinomas prior to definitive treatment.

Excisional biopsy should be performed rather than punch biopsy if malignant melanoma is suspected. The punch biopsy should include subcutaneous fat. Crush artefact, when forceps are used to extract the biopsy, should be avoided. The specimen should be immediately placed in 10% formalin.

3. Shave biopsy

This usually results in epidermis only or epidermis and superficial dermis. Shave biopsy should not be used if there is any suspicion of malignant melanoma. In acral sites (soles of feet, palms or hands) usually only keratin is obtained in a shave biopsy.

4. Curettage

This is the least satisfactory form of skin biopsy, as the tissue is typically fragmented and may not be satisfactory for histology. Curettage is suitable for some superficial lesions.

Site Selection

This is of critical importance particularly in:

Inflammatory dermatoses – biopsy should be from a fully developed lesion and if the lesions are in different stages of evolution, multiple biopsies should be taken. Treated areas should be avoided. For alopecia investigation two biopsies should be supplied for vertical and horizontal sections.

Vesiculobullous lesions, ulcers, pustules – biopsies should be taken from very early lesions and should include normal skin.

Direct immunofluoresence microscopy

Biopsies should be placed in transport medium (NOT in formalin). This may be obtained from the lab by phoning 02 6285 9857. Alternatively the specimen may be submitted to the laboratory fresh on damp saline–soaked gauze.

The specimen should arrive in in the laboratory within 2 hours of being taken.

See *Histopathology*

Skin Antibodies

Specimen: Serum – Gel

Reference Range: Not detected

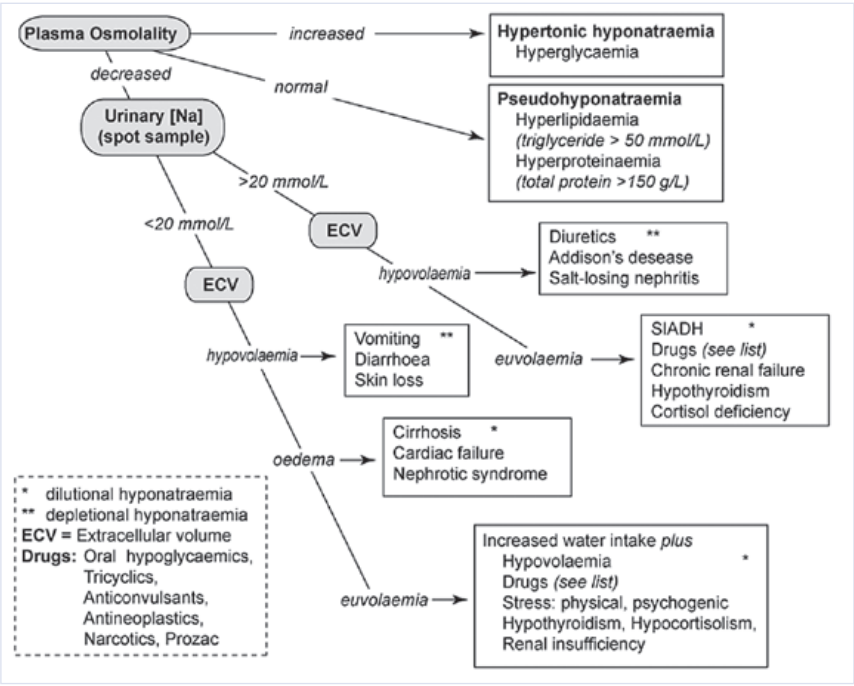
Specify whether pemphigus or pemphigoid

Sodium, serum

Specimen: Serum – Gel

Reference Range: Adults 135–145 mmol/L

Hyponatraemia



Sodium, urine

Specimen: Spot urine or 24-hour (nil preservative)

Reference Range: Supplied with report

In a well person, urine sodium output reflects dietary intake. A low output, < 20 mmol/day, may be an indicator of a total sodium deficit but interpretation requires consideration of all fluid and electrolyte parameters.

Sperm Antibodies

Specimen: Semen sample and serum – Gel

Reference Range: Not detected

Antibodies in seminal fluid are a marker for immunological male infertility.

Spherocytes

Spherocytosis is associated with haemolysis:

- Congenital spherocytosis
- Autoimmune haemolytic anaemia
- Microangiopathic haemolysis
- Severe burns
- After splenectomy.

Sputum Culture

Sputum is examined in three clinical situations:

1. Suspected TB

Where infectious TB is a possibility, ZN stain and culture are essential.

See *Tuberculosis*

2. Pneumonia

If the patient can produce sputum – often a problem – the examination is worthwhile provided it is collected before antibiotics are commenced.

3. Acute or chronic bronchitis

A sputum examination is seldom useful though the matter is debated. In exacerbations of chronic bronchitis the important organisms are *Strep. pneumoniae* and *Haemophilus influenzae*. *Moraxella* (formerly *Branhamella*) *catarrhalis* may also be involved.

Sputum Cytology

Sputum cytology is useful for the investigation of bronchopulmonary and metastatic tumours. Centrally located tumours have a higher rate of pick up on sputum cytology than peripheral tumours.

Sputum cytology may also be useful for the investigation of asbestosis and various infections, particularly in immunocompromised patients.

Patient Information for Collecting Sputum for Cytology

Routine sputum cytology series incorporates a separate specimen collected on each of three consecutive days (the series is covered by one Medicare item number and hence requires only one request form marked x 3).

Specimens should be collected early in the morning to maximise the yield of pooled endobronchial material.

The mouth should be rinsed thoroughly with water prior to collection.

Deep cough specimens are important to ensure bronchopulmonary origin of the specimen (oral and nasopharyngeal material are of little diagnostic value).

At least 2mL of sputum should be expectorated into a specimen container and refrigerated pending delivery to the laboratory or nearest Collection Centre.

Each container needs to be labelled clearly with full name, collection date, date of birth, the number in the series (i.e. 1, 2 or 3) and should be delivered to the nearest Collection Centre as soon as practicable on the day it is collected. (Best results are obtained on fresh specimens.)

Staphylococci

These organisms are divided into two groups according to whether they produce coagulase, an enzyme–like protein that bestows invasive potential.

1. Coagulase-positive staphylococci

Staph. aureus The common pyogenic organism of skin and wound infections and a variety of serous systemic infections. About 30% of normal people carry *Staph. aureus* in their anterior nares.

MRSA–methicillin resistant Staph. aureus

MRSA are always resistant to methicillin/oxacillin, penicillin, amoxycillin, amoxycillin–clavulanate, and all cephalosporins.

With regard to other sensitivities, it is important to distinguish between two different forms of MRSA:

Multi–resistant MRSA

Until 1995 these were the only variety known. They are resistant not only to penicillin and cephalosporins but also alternative oral agents such as erythromycin, tetracycline and cotrimoxazole.

They are found infrequently but when identified require action:

– Isolate the patient
– Minimise staff contact
– Screen staff if there is evidence of cross infection
– Treat infection
– Consider eradication therapy for those who are colonised
– Consult Infection Control Officer.

Non multi–resistant MRSA

In 1995 new strains of MRSA were identified that were seldom resistant to erythromycin, tetracycline or cotrimoxazole. They do no require extensive infection control procedures but should be treated with standard precautions as for other *S. aureus*.

2. Coagulase-negative staphylococci

Staph. saprophyticus A relatively non–pathogenic organism. It is of clinical interest mainly as a cause of urinary tract infections, particularly in young women where it is second in frequency to E. coli.

Staph. epidermidis A universal skin commensal, non–pathogenic except where host defences are seriously compromised or when introduced by instruments or prostheses.

S. epidermidis Is not infrequently grown from an MSU but is of doubtful pathogenicity. If accompanied by pyuria, treatment could be considered bearing in mind the other causes of pyuria.

Streptococci

Streptococci causes a wide spectrum of common diseases. Three species or groups are commonly isolated.

1. Streptococcus pyogenes

Also known as beta–haemolytic group A streptococcus, this organism is the common cause of bacterial pharyngitis. It also causes 20% of the skin and wound infections, particularly cellulitis and impetigo where it is the most frequent pathogen. There are a wide variety of other less common sites of infection.

2. Streptococcus pneumoniae

Often called the pneumococcus, a common commensal in our upper respiratory tract, with up to 60% of people carrying it. It is a gram–positive diplococcus which causes alpha–haemolysis on blood agar (greening of the agar due to partial haemolysis of the red blood cells) and often has a capsule conferring resistance to host defences. More than 80 different serotypes are known but most disease is caused by a limited number of serotypes. Transmission is from person to person by droplet spread.

It causes a wide range of infections: pneumonia with or without bacteraemia, otitis media, sinusitis, meningitis, and other types of invasive disease. It does not cause pharyngitis or tonsillitis. 30–50% of community–acquired pneumonia is due to pneumococci and around 10% of nosocomial pneumonia.

Several conditions are associated with more frequent and severe pneumococcal pneumonia: alcoholism, diabetes, chronic renal disease, heart failure and some malignancies.

In the eye, *S. pneumoniae* is susceptible to chloramphenicol ointment but is intrinsically resistant to neomycin.

Pneumococcal vaccine

Splenectomised patients are at significant risk for invasive pneumococcal disease and should receive vaccine.

Pneumococcal vaccine contains 23 of the most common serotypes and covers 90% of strains causing invasive diseases. 90% of adults respond to a single dose. The vaccine is also recommended for persons > 65 years, persons at increased risk of complications such as those mentioned above, immunocompromised patients, including those with HIV

3. Streptococcus faecalis (Group D streptococcus)

Strongyloidiasis

Intestinal infestation with *Strongyloides stercoralis* is found worldwide in tropical regions, particularly SE Asia and Central America. Clinically it can present with abdominal discomfort, diarrhoea or malabsorption. A life–threatening disseminated hyperinfection occurs in immunocompromised patients. Diagnosis relies on finding larvae in faeces or, in disseminated disease, respiratory secretions. It tends to be less easy to eradicate than hookworm which is a related nematode.

Synacthen Stimulation Test

Specimen: Serum – Clot or gel
Collect baseline specimen for cortisol.
Inject 0.25mg Synacthen (synthetic ACTH) IM.
Collect specimens 1/2 hour and 1 hour after Synacthen injection.

Indications:
Used in suspected Addison's disease or for assessment of adrenal reserve after long term steroid therapy.

Precaution:
Synacthen can cause a severe anaphylactic shock reaction leading to profound shock and collapse. Test should only be performed by a clinician with access to full resuscitation facilities. Test not performed by pathology laboratory or on outpatients.

Synovial Aspirate

Specimen: Collect into a plain tube
If sufficient fluid is available, collect a sodium citrate or EDTA.

Synovial fluid examination provides definitive diagnosis of septic arthritis, gout and pseudogout, and places other effusions into the broad categories of non–inflammatory lesions (trauma, osteoarthritis, etc.) and inflammatory non–infective disease (rheumatoid arthritis, etc.).

Examination covers the following:

- Gross appearance – clear, cloudy, purulent, blood–stained (trauma)
- Volume
- White cell count
- White cell differential–polymorphs predominate in septic arthritis, gout, pseudogout and sometimes in rheumatoid arthritis; mononuclear cells usually predominate in the inflammatory non–infective disorders.
- Gram stain
- Crystals – polarised light is used to look for the urate crystals of gout or the pyrophosphate crystals of pseudogout.
- Culture – aerobes, gonococci and anaerobes.

Other examinations will be performed on request.

Syphilis

Where there is a possibility of syphilis, serological tests should be performed. If a lesion is strongly suspicious of syphilis with the typical painless indurated ulcer, direct fluorescent stain can be arranged with the pathologist.

Specimen: Serum – Gel

Reference Range: Non–reactive

1. Screen test

The initial enzyme–immunoassay test is a screen for both non–treponemal and treponemal antibodies. A positive screen test will be followed by the RPR (Rapid Plasma Reagin), TPHA (Treponema Pallidum Haemagglutination) and FTA (Fluorescent Treponemal Antibody).

2. Non–treponemal (reagin) tests, RPR and VDRL

The RPR and VDRL (Venereal Disease Research Lab) are the most widely used of the older, nonspecific reagin tests. They can be transiently positive in a wide range of viral and autoimmune diseases as well as syphilis and yaws. After successful treatment of syphilis, the RPR/VDRL titre declines and may become negative.

The RPR/VDRL is used as a screening test for syphilis and for monitoring response to treatment. A positive result may be due to current infection.

3. Treponemal tests TPHA and FTA

The TPHA and FTA are much more specific for treponemal infections but unexplained, weakly reactive, false positives of one or both tests are sometimes found.

After a treponemal infection, the TPHA and FTA stay positive indefinitely without indicating whether disease is current or past.

In true treponemal infections, all three tests, VDRL or RPR, TPHA and FTA, will usually be clearly reactive.

Antibody tests cannot distinguish between yaws and syphilis infections which are both due to *Treponema pallidum* species.

Systemic Lupus Erythermatosus (SLE)

A relatively common (1:1000) connective tissue disease which can affect a wide variety of systems and is characterised by autoantibodies directed against nuclear components. For initial investigation (screen), the following tests are recommended:

ANA, Anti- DNA, Lupus inhibitor, Rheumatoid Factor (RF).

There is no single specific test but rather a list of diagnostic criteria. The American Rheumatology Association (1982) recommends that if four of the following criteria are positive, the diagnosis can be made:

- ANA positive (95%)
- Anti–DNA, or ENA Sm, or biological false positive VDRL
- Haemolytic anaemia, or leukopenia, or thrombocytopenia
- Nephropathy (proteinuria > 0.5 g/day)
- Arthritis
- Pleuritis or pericarditis
- Malar rash
- CNS involvement (psychosis, seizures).

During active SLE, complement (C3 and C4) are reduced and the ESR is raised.

See *Antinuclear Antibodies (ANA)*
Extractable Nuclear Antigens (ENA)

T

T3, free (Tri–iodothyronine)

Specimen: Serum – Gel

Reference Range: 1.6–6.5 pmol/L

T3 is the active thyroid hormone with T4 being effectively a “prohormone”. 80% of T3 is formed from T4 in the tissues, the remainder being directly secreted by the thyroid.

Like T4, T3 is protein–bound in the blood and protein–binding abnormalities can elevate both free and total T3 without causing hyperthyroidism.

Elevated by:

- Hyperthyroidism, including T3 thyrotoxicosis
- T3 therapy – because T3 has a short half–life, levels fluctuate up to 30%, depending on time of last dose
- Subacute thyroiditis – early stage
- Hashimoto’s thyroiditis – occasionally in early stage
- Heterophilic antibodies, protein – binding abnormality.

Lowered by:

- Hypothyroidism – but is a poor test for this
- T4 therapy – though T3 is usually within the reference range when T4 dose is appropriate
- Non–thyroidal illness
- Drugs: amiodarone, propranolol, steroids, lithium, iodine in tonics or contrast medium
- Hypopituitarism (secondary hypothyroidism).

T4, free (Thyroxine)

Specimen: Serum – Gel

Reference Range: 9.6–23.4 pmol/L

Although T4 is the principal thyroid hormone, it is converted to T3 in the tissues. It is the preferred thyroid replacement therapy in hypothyroidism.

Elevated by:
- Hyperthyroidism
- T4 therapy–because the half–life is long, time of sampling for monitoring is unimportant
- Non–thyroidal illness
- Drugs: amiodarone, NSAIDs, propranolol, steroids, iodine–containing contrast medium, heparin
- Heterophilic antibodies, protein–binding abnormalities
- Sub–acute thyroiditis – early stage
- Hashimoto's thyroiditis – early stage
- Self–administration of T4 e.g. for attempted weight reduction.

Lowered by:
- Primary hypothyroidism
- Non–thyroidal illness (uncommon)
- Hypopituitarism (secondary hypothyroidism)
- Hashimoto's thyroiditis
- Sub–acute thyroiditis (recovery stage)
- Drugs: T3 (Tertroxin), phenytoin, lithium, carbamazepine.

Tegretol (Carbamazepine)

Specimen: Serum – Gel

Trough level should be taken just before next dose (within one hour).
Peak level should be collected 3 hours post dose.

Therapeutic range: 15–40 umol/L

Testosterone

Specimen: Serum – Gel

Reference Range: F <4.6 nmol/L
M 8 yrs < 2.5 nmol/L
M 14 yrs 3.0–10.0 nmol/L
M Adult 8.0–38.0 nmol/L

Free and total testosterone
Free testosterone, the biologically active fraction, is derived from total testosterone using the value for SHBG which is the serum binding protein. Where the increase in testosterone is small, as in minor degrees of hirsutism in the polycystic ovary syndrome, it may be found that only the free testosterone is above its reference limit.

Age changes in males
The male embryo has high levels of testosterone falling to female levels at time of birth but with a second peak during the first few months of life. Throughout childhood male

levels are not much higher than in females. Until at about age 11 the pubertal rise commences, reaching adult levels at about age 17. Levels decline slowly from about age 40 on, the decline being more marked in free testosterone than total testosterone, which is partially sustained by a rise in SHBG.

Thalassaemias

The thalassaemias are common hereditary conditions in which there is a reduction in the synthesis of one or more of the four globin chains of the Hb molecule. Adult haemoglobin, HbA, which makes up > 96% of normal Hb, contains two alpha and two beta globin subunits and these define the two main groups of disorders, the alpha thalassaemias and the beta thalassaemias.

The genes for thalassaemia are found commonly in Asians, Polynesians, Africans and Mediterranean people. Clinically, the thalassaemias range from the clinically undetectable heterozygous state, through mild microcytic, hypochromic anaemias, (thalassaemia minor) to severe transfusion–dependent anaemias or fetal death (thalassaemia major).

Clinical Presentation

The major thalassaemia syndromes usually come under specialist care early in life with moderate or severe haemolytic anaemias.

- The thalassaemia trait disorders, which are usually asymptomatic, will for the most part be discovered incidentally on a routine blood film.
- Anaemia, usually mild, sometimes moderate, sometimes lower end of the reference range
- Microcytic, hypochromic blood film, resembling an iron deficiency anaemia but with an MCV which is disproportionately low compared with the anaemia
- Normal ferritin, s. iron, iron–binding capacity
- The blood film may show other suspicious abnormalities; and the histogram of red cell size distribution may show a pattern typical of thalassaemia.

Diagnosing the Type of Thalassaemia

The thalassaemias are complex and diverse and under certain circumstances may require specialised DNA and globin chain analyses to characterise them fully. In ordinary clinical practice, once the diagnosis of thalassaemia has been suspected because of microcytosis in the absence of iron deficiency, a group of tests is applied to broadly separate alpha from beta thalassaemias.

These tests are:

- HbH inclusion bodies (alpha)
- Hb electrophoresis
- HbA2 % (beta)
- HbF % (beta)
- Kleihauer stain (beta).

Alpha Thalassaemias

Each parent contributes two alpha genes giving a total of four. Alpha thalassaemia results from deletion of one or more of these four genes.

Deletion		Hb	MCV	HbH bodies	clinical
– alpha/alpha alpha	single gene	N	N	Absent	'silent carrier'
–alpha/–alpha	two gene (trans)	N	N (usually)	Very occasional	Polynesian type
–/alpha alpha	two gene (cis)	±/↓	↓	Occasional	more severe than trans. asian type
–/–alpha	three gene	↓	↓	Numerous	HbH disease
–/–	four gene	–	–	–	Intrauterine death

Beta Thalassaemias

Beta thalassaemias arise from point mutations in the coding genes rather than deletions and more than 300 mutations have been identified. The first thalassaemia to be described, by Cooley in 1925, was severe beta thalassaemia found in a patient of Mediterranean descent, thalassa being the Greek for sea.

Alpha and beta thalassaemias occur with roughly equal frequency.

Theophylline (Nuelin)

Specimen: Serum – Gel

Therapeutic range: 55–110 µmol/L

The therapeutic range refers to peak levels. The specimen is collected 4–6 hours after the last dose for long–acting preparation, 2 hours after those that are short–acting.

ThinPrep

See *Cervical Cytology*

Throat Swabs

Swabs are taken from the tonsillar area for a sore throat, or the posterior pharyngeal wall for sinus trouble so as to collect post–nasal discharge.

The most common indication for a throat swab is to detect Group A streptococcus (*S. pyogenes*). Culture results take 1–2 days to be reported.

Rapid streptococcal antigen tests are not performed in the laboratory. Their general use increases the cost of testing as it is commonly recommended that all patients with a negative rapid antigen test require a culture.

Streptococcus pyogenes, which is 100% sensitive to penicillin, is by far the commonest bacterial pathogen in pharyngitis in immune competent patients. Occasional pathogens include *Arcanobacterium haemolyticum*, *Bordetalla*, gonococci, groups C and G streptococci, diphtheria, and anaerobic organisms in quinsy.

Thrombocytopenia

A reduction in platelets below $150x10^9$/L. The lower the platelet count, the stronger the possibility of spontaneous bleeding.

$0–20x10^9$/L	Severe thrombocytopenia which can be life–threatening. Should always be referred to a haematologist.

Bleeding is not directly proportional to the degree of thrombocytopenia.

Thrombocytopenia in pregnancy can put the fetus at risk and should be referred for a specialist opinion.

Causes of thrombocytopenia:

Acute infection

Transient, often marked, thrombocytopenia may be seen in association with acute viral illnesses in children. In this setting, platelet levels often recover rapidly. Post–viral thrombocytopenia in adults may persist at mild to moderate levels and is assumed to have an immune–mediated mechanism. Acute HIV infection may be a cause of thrombocytopenia.

Drugs

A long list including salicylates, sulphonamides, trimethoprim, penicillins, cephalosporins, methyldopa, chlorthiazide, frusemide, tolbutamide, heparin, phenytoin, phenobarbitone, carbamazepine, phenothiazines, phenylbutazone, gold, penicillamine – and others.

Alcohol and liver disease

ITP

Idiopathic Immune Thrombocytopenic Purpura

Leukaemias

Marrow infiltration

Malignancy, myelofibrosis

Other

Hypersplenism, SLE, B12 or folate deficiency, DIC, post–transfusion, post–partum

Pregnancy

ITP, dilutional or gestational thrombocytopenia, GPH syndromes

Thrombocytosis

Refers to an increase in the platelet count above $450x10^9$/L.

Transient reactive thrombocytoses up to about $800x10^9$/L are common. Causes are:

- Blood loss
- Surgery, trauma
- Infection, including viral infection
- Other inflammatory disorders
- Malignancy – an important cause of persistent thrombocytosis
- Myeloproliferative disorders
- Essential thrombocythaemias
- Polycythaemia vera
- Myelofibrosis
- Myelodysplasia.

Thrombophilia

Patients with thrombosis may be further investigated, especially those with thrombosis at a younger age, thrombosis at an unusual site, those with a family history of thrombosis or those with recurrent thrombosis.

Non-genetic risk factors for thrombosis must also be assessed for cumulative risk assessment such as hypertension, smoking, diabetes mellitus, obesity and the oral contraceptive .

Tests may include:

- Antithrombin III (ATIII)
- Protein C
- Protein S
- Lupus Inhibitor
- Anticardiolipin Antibodies
- Homocysteine levels
- Activated Protein C Resistance
- Factor V Leiden Mutation
- Prothrombin Gene Mutation.

Medicare benefit is available where the request for testing specifically identifies that the patient has a history of venous thrombosis or pulmonary embolism or is a first degree relative of a person who has a proven diagnosed defect.

Please specify individual tests requested.

Thyroid antibodies

Specimen: Serum – Gel
Please specify which thyroid antibody is required.

Reference Range: Not present

Graves' disease and primary hypothyroidism are both autoimmune diseases and are associated with a variety of antibodies.

Anti-TPO and anti-thyroglobulin antibodies

Elevated in:

- Hashimoto's thyroiditis – antibodies in high titre will differentiate non–toxic goitre from Hashimoto's
- Primary hypothyroidism – antibodies present in 90%, often in high titre
- Autoimmune disease marker – there is an association with diabetes and pernicious anaemia
- Euthyroid normals – 10% have antibodies usually in low titre. Annual follow–up will show progression to thyroid disease in some, indicating that these elevations can be a marker for an early autoimmune state.

Thyroid Stimulating Antibody (TS antibody, also called TSH receptor antibody)

This IgG antibody, formerly known as LATS, is the marker for autoimmune thyrotoxicosis (Graves' disease).

Thyroid Stimulating Hormone (TSH)

Specimen: Serum – Gel
Please specify which thyroid antibody is required.

Reference Range: Adults 0.50–6.3 mIU/L

Abnormal TSH levels, whether high in hypothyroidism or low in hyperthyroidism, respond slowly to appropriate therapy with the new equilibrium level not reached for 2–8 weeks.

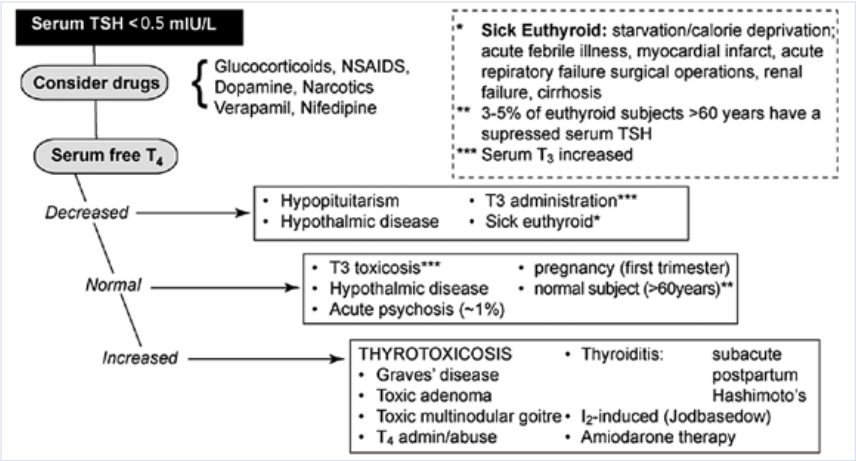
TSH is elevated by:

- Primary hypothyroidism, subclinical or clinical
- Hashimoto's thyroiditis
- Subacute thyroiditis, recovery phase
- Non–thyroidal illness – occasionally during recovery phase
- Ectopic TSH from tumours of lung, breast etc.
- Drugs: lithium, metoclopramide, clomiphene, domperidone, iodides: kelp tablets, amiodarone, contrast medium
- TSH release is pulsatile – minor elevations may simply be detecting the transient peak of a pulse.

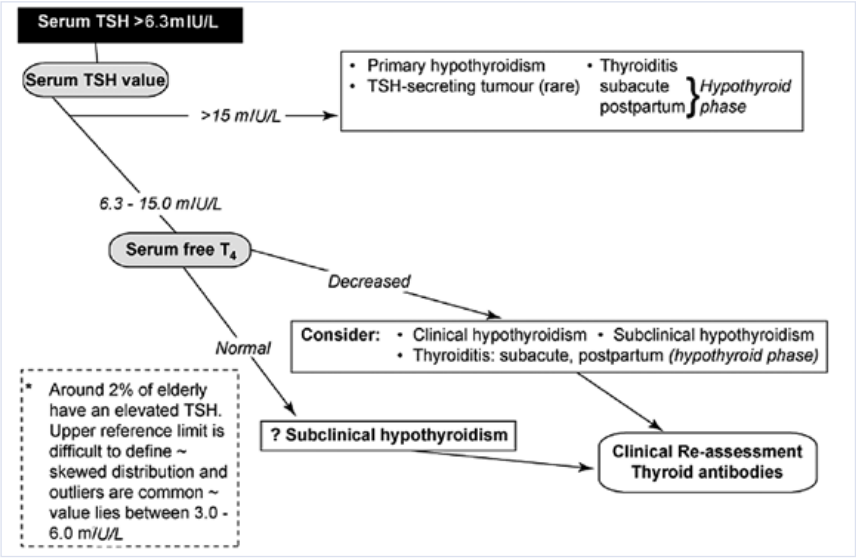
TSH is decreased by:

- Hyperthyroidism – usually the TSH is < 0.03 in clinical thyrotoxicosis
- Thyroid autonomy/sub–clinical hyperthyroidism
- Patients on supra–optimal T4 or T3 therapy
- Drugs: steroids, L–dopa, bromocriptine, heparin
- Non–thyroidal illness.

Suppressed TSH



Elevated TSH



Tissue Typing

Specimen depends on the history – Whole blood ACD tube, clot tube, EDTA plasma or SST tube. Keep specimen at room temperature.

Can be collected on Mondays to Thursdays, however, a booking needs to be made with Red Cross by the doctor or Collection Centre prior to specimen collection.
Red Cross: 02 9234 2332

TORCH Screen Antibodies

This is an acronym derived from infections causing intrauterine growth retardation or death: Toxoplasma, Other, Rubella, CMV, Herpes (and Syphilis).

"TORCH antibodies" is no longer regarded as an appropriate block of tests – individual tests should be requested according to indications.

Toxoplasma Antibodies

Specimen: Serum – Gel

IgM antibodies become detectable 5 days after infection and remain for months or occasionally years. A positive IgM result does not separate current from past infection except when a rising titre can be demonstrated. Approximately 2% of women tested antenatally are +ve for IgM but most do not have active infection.

IgG antibodies become positive 1–2 weeks after infection and remain positive for life. A strongly rising titre over a 3–week interval is good evidence of current infection. 30–60% of the population have IgG antibodies.

Life–cycle and clinical disease
Toxoplasma gondii is an intracellular protozoan found in cats, humans, sheep, pigs and other mammals. Cats are the primary hosts spreading cysts in their faeces to be accidentally ingested by cat–lovers and grass–eating animals. In the secondary host (man, domestic animal) the infection is usually subclinical but with lymphadenopathy, variant lymphocytes in the blood film and a lymphocytosis. Viable parasites in the tissues of domestic animals cause infection when their meat is eaten undercooked. In the immunocompromised human, quiescent lesions can be reactivated causing serious disseminated infections.

Toxoplasmosis in pregnancy
About one third of women acquiring toxoplasmosis during pregnancy will transmit the parasite to the fetus. In the first trimester the incidence of infection is about 10%, but with a high risk of serious or fatal disease in the fetus. In the second and third trimesters the fetal infection rate rises to 30% and 60%, respectively, but with less serious effects in the fetus where the disease may not be apparent until later in childhood with CNS impairment or chorioretinitis.

If antibodies were known to be present at least one month before conception, the fetus will be safe.

Because infection, whether in or out of pregnancy, is usually subclinical, the diagnosis is often made only when an affected fetus or child is encountered.

Sometimes a mother will ask for toxoplasma tests during pregnancy and about 2% of these will test +ve for IgM antibodies, most of them derived from pre–pregnancy infections.

If a mononucleosis–like illness occurs in pregnancy, or if there is any other reason to suspect acute maternal infection, serial testing of antibodies is obligatory followed by toxoplasma DNA testing of amniotic fluid or fetal blood. DNA testing of amniotic fluid at around 18 weeks of pregnancy is highly predictive of the presence or absence of fetal infection. Discussion with a microbiologist is recommended when toxoplasmosis in pregnancy is suspected.

Transferrin

Specimen: Serum – Gel

Reference Range: 2.0–3.2 g/L

Transferrin is the serum iron–binding transport protein which is measured by "iron–binding capacity" and is described under that heading.

As an approximation, transferrin = IBC ÷20.

Trichinosis

An intestinal and muscular infection caused by the nematode *Trichinella spiralis*. Pigs are the main reservoir, and eating under–cooked pork the principal mode of infection. Although T. spiralis is common worldwide, particularly in North America and Europe. Australia is considered essentially free of infection. Infected pigs probably still exist. It is likely that rats and wild cats serve as a reservoir.

Diagnostic tests include eosinophil count, CK, serological tests and muscle biopsy.

Triglyceride

Specimen: Fasting Serum – Gel

Desirable Range: 0.5–2.0 mmol/L (fasting)

Triglyceride in the fasting state comes from the liver in VLDL particles and their smaller IDL products. When triglyceride metabolism is markedly impaired, chylomicrons can be present in the fasting state. If the fasting triglyceride is above 5 mmol/L, the specimen will appear cloudy due to raised VLDL and/or chylomicrons which rise to form a creamy layer on standing.

Elevated fasting triglyceride

1. **Primary**
- Familial combined hyperlipidaemia
- Familial hypertriglyceridaemia
- Type III ("remnant removal disease") hyperlipoproteinaemia.

2. **Secondary**
- Obesity
- Alcohol
- Diabetes
- Hypothyroidism
- Liver disease, particularly obstructive
- Nephrotic syndrome
- Pancreatitis

- Pregnancy
- Significant illness
- Drugs: oestrogen, oral contraceptives, beta blockers, corticosteroids, thiazides, retinoic acid, antiviral agents, valproic acid.

Very high triglycerides

A patient with a triglyceride above 10.0 mmol/L is at risk for acute pancreatitis and requires immediate restriction of dietary fat and alcohol, treatment of any other underlying cause such as diabetes, and addition of a triglyceride–lowering drug such as a fibrate if other measures fail.

With massive hypertriglyceridaemias, serum can have the appearance and consistency of cream. Often there is more than one aetiology, e.g. diabetes and alcohol.

Triglyceride levels can rise and fall very quickly. Above a level of 6.0 mmol/L, lipoprotein lipase clearance mechanisms are saturated which means that dietary fat can rapidly raise triglyceride to surprising levels. Dietary restrictions may cause it to fall equally quickly.

See *Lipid Disorders*

Troponin

Specimen: Canberra + Cooma region: Serum – Gel
Goulburn + Bega region: Whole Blood – Lithium heparin

Reference Range: Troponin T < 0.03 ng/mL
Troponin I < 0.03 ng/mL

The markers of myocardial damage are, in decreasing order of specificity, troponins I & T, CK–MB, total CK, myoglobin, AST and LD.

AST and LD no longer have a useful role.

Myoglobin, though relatively nonspecific, is the earliest to rise and can appear within the first six hours.

The remaining four typically start to rise 4–12 hours after infarction, reach a peak and return to baseline over a period which differs between the four.

	Elevated for
CK–MB	2–3 days
Total CK	2–3 days
Troponin I	4–7 days
Troponin T	4–10 days

Of these, the troponins are the most specific and usually, when elevated, indicate infarction. Small elevations found in unstable angina may indicate ischaemic damage and carry a poorer prognosis than a normal level.

Troponins do not rise earlier than CK but their longer period of elevation is useful, e.g. when chest pain occurred several days earlier.

CK–MB is falling into disuse since the troponins arrived, though it may be useful in plotting successive reinfarctions.

An elevated total CK is nonspecific but if serial CK measurements follow the typical time course of an MI, they support the diagnosis. A small MI may occasionally be suggested by CK changes that stay within reference range but show a typical peak over 2–3 days.

Elevation of cardiac troponin is more sensitive and specific for myocardial infarction than CK–MB. Concentrations rise with 4–12 hours of commencement of cardiac pain and remain elevated for up to 10–14 days.

Unstable angina can also cause elevations due to minimal myocardial damage not detected by CK–MB. It is important to recognise that angina without myocardial necrosis will not elevate troponins.

If the specimen was obtained less than 6 hours after commencement of chest pain, a follow–up should be collected.

Tuberculosis (TB)

One third of the world's population is infected with TB, the majority of them in the developing world.

High risk groups are those who have arrived from Asia or the Pacific Islands in the past few years and those with other family members who have been infected.

Infection is by inhalation of droplet nuclei. Local replication leads to acute disease in about 5% of those infected. It is higher in infants and those more than 60 years old. About 10% of infected people reactivate during their lifetime. This is more common over the age of 50 and in men more than women.

Diagnosis

Specific diagnosis requires isolation of bacilli from an appropriate specimen.
The tuberculin test can be useful provided its limitations are recognised.

Specimens:

Urines	The whole of an early morning specimen should be collected on three different days. A special collection jar is provided and each should be returned to the lab on the day of collection.
Sputum	ZN stain and culture of sputum is the most effective test for diagnosing infectious TB. Three specimens should be examined, preferably early morning on separate days. Automated liquid–based culture methods grow TB from most smear–positive specimens within a week.
Bronchial washings	Collect into a sterile container.
Tissue	Specimens for culture must be placed in sterile saline or water, not in formalin.

See *Mantoux Test*

Tumour Markers

Specimen: Serum – Gel

The principal value of tumour markers is in monitoring patients with known malignant disease. The lack of tissue specificity for many markers as well as the occurrence of false positive and false negative results limits their use as a screening or diagnostic test. Some of the markers applied to tumour surveillance are shown below.

Tumour Marker	Condition
Alpha–fetoprotein (AFP)	Testicular cancer (non–seminomatous) Hepatocellular carcinoma Gastrointestinal tract cancers with and without liver metastases
CA 125	Ovarian carcinoma (non–mucinous) liver, pancreatic, lung, colon, uterine, fallopian tube
Carcino Embryonic Antigen (CEA)	Colorectal cancer Pancreatic, breast, lung, small intestine, stomach, ovaries
Prostate Specific Antigen (PSA)	Prostate cancer
Human Chorionic Gonadotropin (HCG)	Choriocarcinoma Hydatidiform mole Trophoblastic diseases

Turner's Syndrome

In Turner's syndrome, phenotypic females have only one X chromosome rather than the usual two. Clinical features include short stature, sexual infantilism and primary amenorrhoea. Gonadal dysgenesis is indicated by raised FSH and LH and low oestradiol.

Diagnosis is based on demonstration of the abnormal karyotype by the cytogenetics laboratory.

U

Urate, serum

Specimen: Serum – Gel

Reference Range: Supplied with report

Urate is a breakdown product of cell nuclei, one third from the diet, two thirds from endogenous tissue catabolism. 20% of urate is excreted by the kidney, the remainder in the gut.

Elevated Urate

Like cholesterol, baseline serum urate levels are genetically determined but elevated by secondary factors:

- High purine diet – liver, kidney, shellfish, fish roe, kina
- Alcohol
- Renal insufficiency
- Drugs: diuretics, salicylates in low dose, steroids, chemotherapeutic agents, niacin, ethambutol, pyrazinamide
- Polycythaemia vera
- Malignancies, particularly leukaemias or lymphomas being lysed by chemotherapy
- Hypothyroidism
- Rare enzyme defects – can present as gout or urate nephropathy in children, young adults or pre–menopausal women
- Down's syndrome
- Metabolic syndrome – hypertension, dyslipidaemia, diabetes, obesity
- Pregnancy – compared with the non–pregnant base–line, levels are 20% lower in the 1st trimester and 20% higher in the 3rd
- Gout – an acute inflammatory arthritis precipitated by urate deposition in synovial tissue and occurring mainly in middle–aged and older men and in post-menopausal women.

Demonstration of urate crystals in aspirated synovial fluid establishes the diagnosis beyond doubt.

Raised serum urate levels are contributory but not diagnostic. During an acute attack of gout, serum urate may actually fall below the levels that will be found between attacks.

Evaluation of Hyperuricaemia

Exclude:

Potentially correctable contributory factors:

- Obesity
- Alcohol
- Hypertriglyceridaemia
- Drugs (especially Thiazides, but also Diuretics, Salicylates [low dose], Nicotinic acid, Pyrazinamide, Ethambutol, Cyclosporin)
- Hypertension
- Low fluid intake

Consider:

High Purine intake

Diet (meats, yeast products)

Increased Urate production

Primary

- Idiopathic
- Enzyme defects

Secondary

- Blood dyscrasias
- Infectious mononucleosis
- Malignancy
- Cytotoxic therapy
- Psoriasis
- Alcoholism
- Prolonged exercise

Decreased renal excretion

Primary

- Syndrome–X, (insulin resistance; dyslipidaemia [increased TG, low HDL–chol]; obesity; hypertension; hyperuricaemia)
- Idiopathic

Secondary

- Renal failure

If renal failure is causing the hyperuricaemia, serum creatinine will be > 0.40 µmol/L, serum urate < 0.65 mmol/L and the ratio of urine urate: creatinine will be < 0.7. If hyperuricaemia is causing the renal failure, then serum urate > 0.7 mmol/L and urine urate: creatinine ratio > 0.7.

- Dehydration
- Diuretics
- Ketonaemia (starvation; diabetes mellitus
- Hyperlactataemia (alcohol; toxaemia or pregnancy)
- Drugs
- Hyperparathyroidism

Urea

Specimen: Serum – Gel

Reference Range: Adults 3.0–8.0 mmol/L

Elevated by:

- Renal impairment – but serum creatinine is a better test being less subject to other interferences
- High protein diet – an important determinant
- Catabolic states – any acute serious illness, particularly sepsis
- Dehydration – at low urine flow, there is increased tubular reabsorption of urea
- Bleeding into GI tract – can give elevations up to 15 mmol/L
- Prostatic hypertrophy – or other post–renal obstruction
- Drugs – steroids, diuretics.

As a routine measure of renal function, creatinine has almost entirely replaced urea. Nephrologists, however, measure urea as well as creatinine in dialysis patients, the preferred urea level being below 20–30 mmol/L.

An unexpected fall can be due to loss of albumin in dialysis fluid.

Unexpected rises in dialysis patients can be due to:

- Dietary non–compliance, too much protein
- Sepsis
- Dehydration or catabolic illness
- Steroid dose too high.

Urethral Swabs

Male
Where there is a discharge, swabs and gonococci culture should be collected by the doctor at the time of examination. In the absence of a discharge, urethral swabs are unlikely to grow a pathogen.

Female
The urethra can be swabbed for gonococci when looking for STD.

Urine Cytology

Urine cytology has a high degree of accuracy in high grade urothelial tumours and carcinoma in–situ. However the test is less accurate with low–grade neoplasia including papillomas. Infection, lithiasis and catheterisation may be difficult to distinguish from low grade urothelial neoplasia on urine cytology.

Patient Information for Collecting Urine Series for Cytology

Three samples of urine to be collected preferably on three consecutive days. (The series is covered by one Medicare item number and hence requires only one request form marked x 3).

The second urine of the morning is the preferred specimen. It should be collected each morning, and a portion of this sample placed in the “specimen container” provided. The container should be at least half filled.

Each container needs to be labelled clearly with full name, collection date, date of birth, the number in the series (i.e. 1, 2 or 3) and should be delivered to the nearest Collection Centre as soon as practicable on the day it is collected. (Best results are obtained on fresh specimens.)

If there is any delay in delivery the specimen should be refrigerated.

Urine, pigmented/colouration

Causes include:

Red urine
- Haematuria
- Phenolphthalein–containing purgatives, which are red when alkaline. Fresh urine is acid but turns alkaline on standing
- Diet–beetroot, rhubarb turn red on standing, particularly with coexisting iron deficiency
- Some porphyrias–turn red/brown on standing
- Haemoglobinuria
- Myoglobinuria.

Orange / brown urine
- Concentrated urine in febrile states may be described as abnormal by patients
- Bilirubin, or urobilinogen which turns brown on standing
- Drugs, e.g. methyldopa, de Witts pills, chloroquine, rifampicin, quinine, metronidazole, nitrofurantoin, riboflavin
- Some porphyrias turn brown on standing
- Melanin in disseminated melanoma

- Alkaptonuria, turns brown on standing
- Tyrosinaemia.

Yellow urine
- Vitamin B complex, multivitamins.

Blue / green urine
- Drugs, e.g. amitriptyline, NSAIDs, triamterene
- Biliverdin.

Milky urine
- Infection
- Chyluria
- Nephrotic syndrome
- Oxaluria.

Factitious additives (something the patient has added) are another source of pigmentation.

Urine, 24–hour collection

Some tests require preservatives. Refer to specific test for further information.

A carefully timed specimen should be collected as follows:

1. The bladder must be emptied at a set time on day one (e.g. 8.00 am) and this urine discarded.
2. All urine voided during the next 24 hours must be collected and transferred into the large 24–hour urine container provided. The container should be kept refrigerated.
3. The bladder must be emptied at the same time on day two as for the commencement of the test on day one (e.g. 8.00 am). This specimen must be included in the 24–hour specimen for assay.
4. The patient's name and the dates and times of commencement and completion of collection are to be noted on the container. If for Creatinine Clearance, include height and weight of patient and a specimen for Serum Creatinine.
5. The specimen should be forwarded to the laboratory as soon as possible after collection is completed.

Patient information leaflets are available from Collections Centres, website www.capitalpath.com.au or front reception on 02 6285 9800

V

Vaginal Swabs for Discharge

Specimen: Routine bacterial swab collected from 5 cm inside the vagina and placed in transport medium.

The normal flora of the vagina is an abundant growth of gram–positive lactobacilli.

Pathogens are of three main types:

- **Trichomonas** – detected by microscopic examination of a smear taken from the swab
- **Candida** – often obvious clinically as a curd–like growth, it is easily cultured
- **Bacterial vaginosis** – a growth of predominantly anaerobic organisms associated with a lack of normal lactobacilli. Clue cells are a feature and Gardnerella vaginalis may be present.

Metronidazole provides effective treatment for both bacterial vaginosis and *Trichomonas* infection.

Where an STD is suspected as cause of a discharge, cervical and urethral swabs should be collected. Vaginal swabs seldom grow gonococci or test positive for *Chlamydia*.

Valium (Diazepam)

Specimen: Plasma – Lithium heparin
Trough level suggested, taken before next dose (within one hour).

Reference Range: Supplied with report

See *Diazepam*

Valproic Acid (Epilim)

Specimen: Serum – Gel
Suggest trough level collected just before next dose.
If peak requested collect between 0.5–1.0 hours for Syrup, 1–3 hours for capsules and 2–6 hours for coated tablets.

Reference Range: Therapeutic 350–700 µmol/L

See *Anticonvulsants*

Vancomycin

Specimen: Serum – Gel
Trough level is taken before next dose (within one hour).
Peak level is collected 30 min post completion of IV dose, or one hour post IM injection.
Trough levels are preferred, trough should be less than 15 mg/L.

Reference Range: Pre dose trough 5.0–15.0 mg/L
Post dose peak 25.0–40.0 mg/L

Varicella–Zoster Virus (VZV)

Specimen: Viral swab taken from base of fresh vesicle or serum gel for serology.

The same virus is responsible for both chickenpox (*varicella*) and shingles (*herpes zoster*) the latter being a reactivation of dormant virus acquired during a childhood attack of chickenpox.

Diagnosis is clinically obvious in most cases but the virus can be identified by PCR, a fluorescent antibody applied to a suitable smear or by culture. PCR is the preferred method.

Venous Thromboembolism (VTE)

In hospital practice venous thromboembolism is thought to contribute to 10% of deaths. In the community, incidence varies, depending on age, between 0.1 and 1/1000 per year.

Objective diagnoses of DVT (Deep Vein Thrombosis) and PE (Pulmonary Embolism) are important for three reasons–managing the current event, identifying and managing recurrent disease, and managing prophylaxis in high–risk situations such as pregnancy or surgery. The initial objective tests are ultrasound for DVT and lung scanning or helical CT for PE.

Primary heparin therapy is essential for VTE and until recently this was administered by continuous intravenous infusion in hospital, followed by outpatient warfarin.

Low molecular weight heparins are now available which enable many patients with both DVT and PE to be treated on an outpatient basis. The low molecular weight heparin (LMWH) with dose adjusted for bodyweight, is given subcutaneously once or twice each day for a minimum period of five days, followed by warfarin. Using this approach, many patients can be managed as outpatients.

See *Thrombophilia*

Virology Swabs and Specimens

Please provide brief clinical details. If possible, state which viral infections you are considering in the differential diagnosis.

Despite the vast spectrum of disease caused by viruses, they are less often investigated by the laboratory than bacteria. They are harder to grow, harder to treat and the trivial viral infections tend to be self–limiting.

Specific tests are described under the alphabetical entry for each disease.

Identification is by direct viral or virid antigen detection (PCR, immunofluorescence, cell culture) or by detection of antibodies.

Virus or its antigen

Specimens can be faeces, throat swab, serum, vesicle fluid, CSF or aspirated respiratory fluid. Swabs for viral investigation should be placed into viral transport medium.

Antibodies

IgM antibodies usually indicate current infection. IgG antibodies indicate past or present infection.

Diagnosis is established by either a single high titre or by an increase in titre of 4–fold or more on paired sera, the first collected as early as possible in the disease, the second after 2–3 weeks of illness.

Enquiries about specific tests can be directed to the Director of Clinical Pathology on 02 6285 9895.

Vitamin A (Retinol)

Specimen:	Serum – Gel
Reference range:	Supplied with report

Vitamin B12

Specimen: Serum – Gel

Reference Range: > 170 pmol/L

Normal absorption of B12 requires a non–vegetarian dietary source, a normal stomach to produce intrinsic factor, and a normal terminal ileum to absorb the B12/IF complex. A healthy person with replete body stores has enough B12 to last 3–6 years if no more is ingested.

Low B12 levels
Causes include:

- Vegetarian diet
- Drugs: – oral contraceptives
- – metformin
- – other: methotrexate, colchicine, Slow K, anticonvulsants, cimetidine, triamterene
- Pregnancy–B12 levels are often low without tissue deficiency
- Low B12 binding protein, transcobalamin I, which can be measured, but the expense is seldom warranted
- Pernicious anaemia
- Malabsorption
- Inflammatory bowel disease: Crohn's disease, or ulcerative colitis in the terminal ileum
- Previous gastrectomy or ileectomy
- Primary folate deficiency
- Diphyllobothrium fish tapeworm infestation.

Further investigation of low B12
If haematological abnormalities are present–raised MCV, anaemia, neutropenia, thrombocytopenia, oval macrocytes and hypersegmented neutrophils in the blood film–the underlying cause must be identified.

If there are no haematological abnormalities and the patient seems healthy, there is less urgency. The level may be normal for that person or it may indicate early deficiency. It is a matter of clinical judgement and patient choice whether to investigate further at once or at follow–up. The elderly often have low B12 and folate levels and may benefit from supplements.

Elevated B12 levels

- Oral or parenteral vitamin B12 supplement
- Haematological disorders: myeloproliferative disorders, leukaemias, high white cell counts
- Liver disease.

See *Pernicious Anaemia*

Vitamin C

Specimen: Serum – Lithium heparin
Spin, separate and freeze serum.
Wrap in foil to protect from light.

Reference Range: Supplied with report

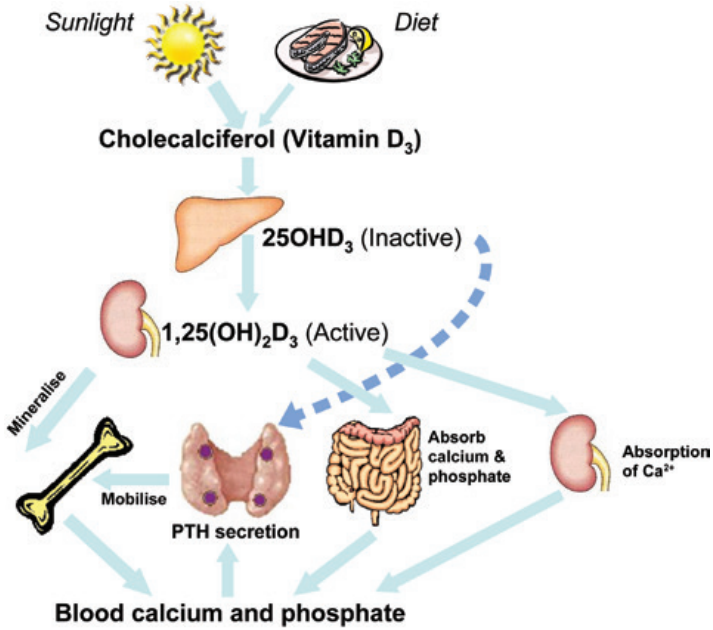
Vitamin D

Specimen: Serum – Gel
Patient should be fasting.

Reference Range: <50 Deficient
50–75 Intermediate
>75 Sufficient

Who is at risk of Vitamin D deficiency?

- People with limited sun exposure
- Dark skinned people especially if veiled
- Malabsorption
- Pregnant women
- Patients on certain medication such as anticonvulsants or corticosteroids.



Vitamin E

Specimen: Serum – Gel
Spin, separate and freeze serum.
Wrap in foil to protect specimen from light.

Reference Range: Supplied with report

Vitamin K

Vitamin K is a fat–soluble vitamin which is essential for hepatic formation of functionally active prothrombin (Factor II) and coagulation Factors VII, IX and X. Although vitamin K is found in the diet, particularly green vegetables, the more important source is the normal bacterial flora of the bowel which synthesises the vitamin. Body stores last only 1–2 weeks if absorption stops.

Deficiencies of vitamin K and Factors II, VII, IX and X can be due to pre–hepatic or hepatic causes.

The prothrombin ratio (PR) and its standardised presentation, INR, are used to monitor vitamin K–dependent factor deficiencies and the anticoagulant effect of warfarin.

Vitamin K injections correct factor deficiencies due to lack of absorption or availability and are also used to correct a prolonged INR due to excess warfarin action.

See *INR*

Von Willebrand's Disease

vWD, discovered in 1926 and the commonest inherited bleeding disorder, is due to a defect in the plasma, von Willebrand factor (vWF) causing a secondary abnormality or platelet adhesion.

vWF, like fibrinogen and fibronectin, is an adhesive protein important in platelet attachment to subendothelial surfaces and in platelet–platelet interactions at sites of vessel injury. vWF also plays an important role in the stabilisation and protection of coagulant FVIII:C in plasma.

The most commonly encountered type of vWD is transmitted as an autosomal dominant trait. These patients usually have mild bleeding symptoms and mild to moderate abnormalities in the relevant laboratory tests (type I heterozygous vWD).

Testing for vWD

FVIII:C The coagulant activity of the FVIII:C protein.

von Willebrand Factor (vWF)
Measured as the von Willebrand factor antigen (vWF:Ag). This large glycoprotein normally stabilises Factor FVIIIC and is variably reduced in vWD. vWF is essential for normal adhesion of platelets and therefore for a normal bleeding time.

von Willebrand factor activity (vWF:Activity)
This is a functional activity which measures the vWF's ability to bind to the Glycoprotein Ib platelet binding site responsible for normal adhesion.

Collagen binding assay (CBA)
The functional assay which measures the vWF's ability to bind to collagen. A discrepancy between the vWF antigen and the CBA suggests a variant form of vWD.

Ristocetin–induced platelet aggregation
The functional activity of vWF measured as the ability of plasma vWF to agglutinate platelets in the presence of ristocetin. The assay is relatively sensitive and specific for vWD.

In the initial laboratory evaluation of patients suspected of having vWD, the following should be performed:

- Bleeding time
- Platelet count
- APTT
- FVIII:C
- vWF Activity
- CBA
- Blood group
- Ristocetin platelet aggregation (not routinely performed in all patients).

One or more of these tests are usually abnormal in patients who have a vWD but the results may vary in the same patient with repeat testing. It is essential that the patient not be taking drugs which could affect the bleeding time. Most frequent offending agents are aspirin or other NSAIDs. Many conditions such as pregnancy, hypo–or hyperthyroidism, uraemia, recent exercise, infection diabetes can affect the FVIII:C activity and vWF antigen levels. Individuals with blood group O have significantly reduced levels of the vWF:Ag and vWF activity compared with blood groups A, B or AB.

W

Wound Swabs

A well collected wound swab can provide valuable information. It is important to sample from any discharging area, and to avoid touching outside the wound perimeter thus avoiding skin contaminants. In the results interpretation the clinical appearance of any wound is important, as well as considering the Gram stain result. Simply growing an organism/s from a wound does not necessarily imply isolation of a significant pathogen.

X

Xylene

Specimen: Urine post shift

Reference Range: Supplied with report

Y

Yersinia enterocolitica

Specimen: Serum – Gel

A pathogen which is being isolated with increasing frequency from cases of infectious diarrhoea–currently about 10% of the total.

Yersinia spp. are frequently present in the intestinal tract of wild and domesticated clinical healthy birds and animals. Processed meat ready for sale has only rarely been shown to be contaminated with *Yersinia*. The source of most cases of *Yersinia* infection unknown.

Some patients develop an abdominal pain syndrome which may last for weeks. The most likely cause for this is intra–abdominal lymphadenopathy.

Y. enterocolitica is susceptible to cotrimoxazole, tetracycline and fluoroquinolone but antibiotics do not shorten the diarrhoea which usually settles spontaneously in 3–10 days.

Infections tend to be sporadic rather than epidemic and are found worldwide. It can cause terminal ileitis which mimics acute appendicitis presenting as right lower quadrant pain, fever and leucocytosis. Some patients develop a reactive arthritis.

Yersinia has been transmitted by blood donated by asymptomatic donors. Patients who have *Yersinia*–associated diarrhoea should inform the blood collection service if they donate blood over the next six months.

Yellow fever Antibodies

Specimen: Serum – Gel

Reference Range: Supplied with report

Z

Zarontin (Ethosuximide)

Specimen: Plasma – Lithium heparin
Trough level is taken before next oral dose (within one hour).
Peak level is collected 2–4 hours after last oral dose.
Please supply time of last dose.

Reference Range: Supplied with report

Zinc, red cell

Specimen: Whole blood – Trace element tube

Reference Range: Supplied with report

Zinc, serum

Specimen: Serum – Gel

Reference Range: Supplied with report

Zinc, urine

Specimen: 24–hour urine (nil preservative)
or
Spot urine–10 mL early morning.

Reference Range: Supplied with report

Capital Pathology, 2 Makin Place Deakin, ACT 2600 Australia

ABN 49 452 500 422

A subsidiary of Sonic Healthcare Limited
ABN 24 004 196 909

Contact us:

Doctors Service Centre (DSC) 02 6285 9803
www.capitalpath.com.au