

**CLINICAL BIOCHEMISTRY**

**HANDBOOK**

**Revision 4.1**

**September 2022**

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[**CLINICAL BIOCHEMISTRY**](http://intranetapps/pathwebfiles/CLINBIO_HEAD.html)

**Personnel**

|  |  |
| --- | --- |
| Dr C Street  Consultant Head of Department  Tel: 01206 742415  [mail](mailto:catherinestreet@nhs.net) | Dr T Likhari Consultant Chemical Pathologist  Tel: 01473 703708 Internal extension: 5708 or [mail](mailto:taruna.likhari@ipswichhospital.nhs.uk) |
| Consultant Clinical Scientist  Vacant | Mrs S Stalley Head of Operations  Tel: 01473 703707  Internal extension: 5707 or [mail](mailto:sarah.stalley@ipswichhospital.nhs.uk) |
| Ms Thelma Masukwedza  Laboratory Manager for Clinical Biochemistry Tel: 01473 703704  Internal extension: 5704 or [mail](mailto:Thelma.Masukwedza@esneft.nhs.uk) | Departmental Secretary  Tel: 01473 703708 Internal extension: 5708 |
| Mr O Pandya  Service Lead for Clinical Biochemistry Tel: 01473 703704  Internal extension: 5704 or [mail](mailto:Opindra.Pandya@ipswichhospital.nhs.uk) | Results/Enquiries  Tel: 01473 703705  Internal extension: 5705 |

**Consultations/Advice**

When considering unusual investigations or requesting interpretative advice, please contact the Duty Biochemist via (lab or Switchboard) during core hours or the consultant on-call via switchboard out of hours’.

**Data Protection and Patient Confidentiality**

The EU General Data Protection Regulation (GDPR) is a pan-European data protection law, which superseded the EU’s 1995 Data Protection Directive and all member state law based on it, including the UK’s DPA 1998 (Data Protection Act 1998), on 25 May 2018.

The GDPR extends the data rights of individuals (data subjects), and places a range of new obligations on organisations that process EU residents’ personal data.

**Complaints**

To make a complaint contact the Patient and Liaison Service (PALS) as follows:

**By Phone**

PALS can be contacted by telephone from 9am to 4pm, Monday to Friday

(Confidential answerphone out of hours)

Free phone 0800 783 7328   
Direct line 01206 742683 or 746448

If your call is urgent and you require assistance outside these hours please dial 01206 747474 and ask to speak to the Duty Matron.

**In Writing**

Patient Advice and Liaison Service

East Suffolk North Essex NHS Foundation Trust  
Colchester General Hospital   
Turner Road   
Colchester, Essex   
CO4 5JL

**By email**

[PALS@esneft.nhs.uk](mailto:PALS@esneft.nhs.uk)

**Location of the Biochemistry Department to which all Samples and Enquiries are referred**

Biochemistry Department,   
Pathology Directorate,   
East Suffolk North Essex NHS Foundation Trust

Ipswich Hospital,   
Heath Road,   
Ipswich,   
IP5 4PD.   
Telephone: 01473 703703/5 or internal extension 5703 or 5705   
Fax: 01473 703259

The Laboratory is situated at the back of the Hospital, off the main street. From the Main outpatient entrance at the front of the Hospital, follow the signs to Pathology reception and ask the reception staff for the Biochemistry Department.

Full service for analysis of samples and for consultation is available from 09:00 - 17:00. A reduced service is available 17:00 - 09:00. Details of this service are provided in the "Out of Hours" area of this document.

**Emergency Investigations**

The Laboratory operates a 24-hour service. Full routine and emergency services operate between 09:00 and 17:00. Outside these hours and at weekends an emergency service is provided.

**Urgent Requests**

Urgent requests requiring immediate analysis, all blood gases must be notified by telephone to the department or, after 17:00 and weekends to bleep 906.

All Emergency Department (A&E) requests where there is a likelihood of 4 hour breach must be notified by telephone.

**The Following Tests are guaranteed out of Hours**

U/E - sodium, potassium, creatinine, urea  
Bone - calcium, phosphate, alkaline phosphatase, albumin, total protein  
LFT - ALT, ALP, bilirubin, albumin, total protein  
CK  
Glucose  
Bicarbonate - request specifically, it is not part of the U&E profile  
Blood Gases - always BLEEP staff  
Amylase  
AST  
Lipids - cholesterol, HDL cholesterol, triglyceride   
Salicylate  
Paracetamol  
CRP  
Magnesium  
Bilirubin  
Ammonia  
COHb  
Gamma GT  
LD  
Lactate  
Lithium  
Osmolality

Digoxin  
Gentamicin  
Phenytoin  
Theophylline  
Vancomycin

TNT   
Serum HCG

Urine sodium

CSF glucose and protein

Requests for tests other than these will be subject to discussion between the requesting clinician and the Consultant Chemical Pathologist or Consultant Clinical Scientist on-call, obtainable via the Hospital switchboard.

**Concerning Out of Hours Work**

It is the responsibility of Health Care Professionals to make sure the samples reach the laboratory, either using the portering services or air tube, where available.

With urgent or emergency work, please do not telephone the laboratory to ask if the results are available.   If the request has been generated by ICE, results will be available on ICE. In addition, results for all requests, however generated, are available via 'Pathology Results' (click on pink heart icon). Very abnormal results, if not previously abnormal will be telephoned in accordance with departmental guidelines. All glucose ≤2.5 & ≥25.0 mmol/L, potassium ≤2.6 mmol/L or ≥6.5 mmol/L, magnesium ≤0.40 mmol/L, and sodium ≤120 mmol/L and ≥150 mmol/L will be telephoned.

Passwords for 'Pathology Results' are available from [Path.admin@ipswichhospital.nhs.uk.](mailto:Path.admin@ipswichhospital.nhs.uk.)

Where tests other than those above are required and it is ESSENTIAL that they are taken during the out of hours period, send them to the laboratory and they will be handled and preserved to ensure that valid analytical results will be obtained when the sample is analysed.  
If it is not essential to collect samples at this time, please wait until the laboratory is operating normal working hours.

**Specimen Collections**

Each specimen must be clearly labelled by hand or using ordercomms label printed at the time of specimen collection, blood gas specimens may also be identified by sticky label but must have some form of identification which meets the following criteria.

The patient details on the specimen must match those on the provided request form. A minimum of 4 matching identifiers are required to enable positive identification of patient.

The following ***must*** be present on the sample, with matching identifiers on both the sample ***and*** the request form:

* Surname
* Forename ( Abbreviated names are not acceptable)
* DOB

Plus one of the following:

* NHS Number
* Hospital Number
* 1st line of address where there is no NHS number or Hospital number – Antenatal partner testing samples and refugee samples only.

**FAILURE TO MEET THESE LABELLING CRITERIA WILL RESULT IN THE SPECIMEN BEING REJECTED**

Fasting status, medications and reasons for investigation should also be stated on the request card.

Each specimen must be appropriately placed in a sealed transport bag for transport to the laboratory.

Specimens which have leaked or are not adequately identified will not be analysed.

High Risk samples from patients with (or suspected to have) Creutzfeldt-Jacob Disease (CJD), Transmissible Spongiform Encephalitis (TSE), Ebola, Viral Haemorrhagic Fever, or Rabies must be labelled clearly with ‘Danger of Infection’ on the request form and sample bags. The Laboratory must be notified that the samples are coming prior to collection and the request must be discussed with the relevant Consultant. All samples from these patients must be sent in the Red High Risk Transporter Boxes available from Pathology.

**Request Form**

Electronic request forms (ICE) should be used whenever possible. It is essential that the request form bears - as an absolute minimum - the above mandatory identifiers, time of sampling, date, the signature of the person making the request and the location to which the report is to be sent.

All requests sent to the laboratory are considered a service agreement between the requester and the laboratory to undertake the analysis of the sample for the tests requested, the laboratory may refer the sample to another laboratory for specialised or confirmatory analysis in order to provide the results.

**Blood Gas Specimens**

Collect minimum 1.0 mL of arterial blood into an electrolyte balanced heparinised commercial syringe or capillary with NO air bubbles present. Blood gases **must** only be collected by trained personnel. Blood gas samples can be sent to the laboratory via the air tube system.

**Blood Specimens**

Sample collection is standardised on the Sarstedt Monovette system and samples should be collected only in the following containers:

|  |  |
| --- | --- |
| 7.5 mL brown top (B) | no anticoagulant |
| 9.0 mL orange (O) | lithium heparin |
| 2.7 mL yellow (Y) | potassium EDTA/sodium fluoride |
| 3.4 mL red (R) | potassium EDTA |

For paediatric use, small volume tubes of the same range are available.  
The appropriate container for most tests is listed. Please note however that in most cases, one Monovette will suffice for a variety of tests, e.g. a single brown topped 7.5 mL tube will allow assay of U & E, LFT, Bone profile, CK etc. There is no need to collect a separate tube for each request unless different types of sample (e.g. both plasma and serum) are required.

**Urine Specimens**

Some compounds are unstable in urine and require special collection containers with added preservatives. Do not discard any such preservative. Refer to collection and preservation list for details.

**24 Hour Urine Specimens**

NOTE:   Complete 24 hour urine collections are essential for quantitative analysis. If any urine passed during the 24 hour period is missed from the collection, the collection should be discarded and the collection procedure recommenced using a fresh bottle.

**The Collection**

**Please ensure that the bottle is labelled with the patient’s full name and date of birth.**  
The collection can be started at any time of day, but must finish at the same time on the next day.

At the start time, empty the bladder and discard the urine.  Do not save any of the urine passed at this point.  The date and time at this point are written onto the bottle.  All urine produced during the next 24 hours is transferred to the bottle.   The collection period is completed when the start time is reached the next day.   At this time the bladder is emptied and the urine saved in the bottle.   The time and date of the completion is written on the label.

The collection is now finished and no further urine should be added to the container.

**Storage**

During the collection, the bottle should be kept in a cool place. It should be sent to the laboratory as soon as possible after completion of collection.

**Cerebrospinal Fluid - CSF**

CSF samples for measurement of glucose and protein should be collected into yellow (fluoride / EDTA) tubes and sent to the laboratory together with a blood sample for the measurement of plasma glucose.  
CSF specimens contaminated with blood will not be analysed for total protein.

Xanthochromia can be detected spectrophotometrically and is available for those patients with a clinical history strongly suggestive of sub-arachnoid haemorrhage but with a negative CT scan. The lumbar puncture should be performed **at least 12 hours** after the onset of symptoms. A minimum of 1.0 mL of fluid is required and should be the final (third or fourth) sample collected from the tap. Additionally a concurrent clotted blood sample (brown cap) should be sent as this may be needed for the interpretation of the findings.

The procedure is requested on the computer using the code XANY, and the specimen should be sent in a plain bottle and despatched directly to biochemistry protected from light. The estimation will be performed Monday-Friday 09:00-16:00 only - it is not available on Bank Holidays.  
CSF samples should not be sent via the air-tube because there is evidence that in vitro haemolysis occurs when such systems are used. This will severely compromise subsequent analysis.

Fluid samples sent to Biochemistry for ?CSF leak e.g. nasal, ear, wound, subretinal (also known as Beta trace protein, Prostaglandin D2 synthetase) **MUST** be accompanied by a serum sample so that we can send both samples to the referral laboratory in London.

The test may also be referred to as ‘TAU’, but this is not the same biomarker as tau-protein, the dementia biomarker. To avoid confusion please do NOT use the term ‘TAU’ when requesting this test.

The use of CSF packs are available from the laboratory.

**Miscellaneous Specimens**

|  |  |  |
| --- | --- | --- |
| For example: |  |  |
| Calculi | } |  |
| Ascitic fluid | } | No special requirements |
| Gastric or duodenal juices | } |  |

Pleural fluid for pH (on non-purulent samples only) requires the standard blood gas syringe and, for glucose the yellow-capped tube.

**Addition of Further Tests to a Request**

Serum samples are stored for three days and glucose and urine samples for two days. Further assays may be added to a request by telephone, subject to the stability of the analyte in the stored sample, and to sufficient sample being available.

**Transport of Samples**

**Transport of Samples from GP Surgeries**

The Pathology Department provides a daily collection service from all GP surgeries. Samples for collection should be individually bagged then placed in a large sealed plastic bag with sufficient wadding to absorb spills.

**Transport of Samples from Wards**

Samples for collection should be individually bagged then placed in a large sealed plastic bag with sufficient wadding to absorb spills. The large bag should be delivered to the pathology reception by the portering staff.

**Transport of Samples via Air Tube**

Samples transported via the air tube must be placed in a sealed plastic sample bag with the request form in a separate pocket to the sample. Samples should be sent to Blood Science stations. All Biochemistry blood samples (including blood gases) may be sent via the air tube system (except High Risk samples or CSF which should be delivered by hand).

**Extremes of Temperature**

Avoid extremes of temperature when transporting samples to avoid sample deterioration.

**Storage of Samples**

If possible deliver samples to the laboratory the same day; all ward samples should reach the laboratory within a few hours of collection. Samples from outside the hospital that cannot be delivered that day should be centrifuged and stored in a refrigerator overnight and delivered the next morning. The refrigerator should maintain a temperature between 2oC and 8oC, it is especially important that the samples do not freeze.

Incorrectly stored samples may result in sample deterioration as seen by raised potassium and phosphate results for example – samples more than 8 hours old that have not been centrifuged will be classed as ‘Old’ and affected analytes will not be assayed.

**Point of Care (POCT)**

**Point of Care Coordinator: Alison Czarnota, Ext 5229 (01473 703229)**

Ipswich Hospital users:

**Blood glucose meters:** Faulty Abbott FPP meters should be brought to the POCT office for troubleshooting.

QC solutions, batteries and workstations (when in stock) are available from the POCT office.

**POCT equipment training:** Training is available for blood gas analysers, urine test strip meters, pregnancy test kits, Coaguchek Pro 2 INR meters, Foetal fibronectin, PROM tests, Hemocue, Hemochron, DCA Vantage HbA1c analysers and urinary drugs of abuse screening kits.

Please call the POCT office for details.

**E-Learning:** Online competency assessments are available for many of the POCT devices in use. Please call the POCT coordinator for details.

**External Quality Assurance schemes (EQA):** We are working towards accreditation under ISO 15189 and ISO 22870 which require all POCT medical devices be enrolled on EQA schemes.

Samples are sent regularly and participation by all staff involved in testing patient samples is mandatory. For details please contact the POCT coordinator.

**Blood gas analysers:** Emergency Department blood gas analyser is available for use by trained staff only.   
For training and barcodes please contact the POCT coordinator.

**Blood gas samples should be collected into electrolyte balanced heparinised syringes.**

**Samples MUST be free from air bubbles, capped and mixed for at least 1 minute before analysis. A minimum of 1ml of blood is required for accurate results.**

**Samples not analysed immediately should be mixed for longer.**

**New equipment:** Any department considering introducing new POCT equipment must seek the advice of the POCT committee.

For details please contact the POCT coordinator.

**Document Review History**

The Biochemistry Department will review the User Guide as and when new information is available or when new test methodologies are introduced.

1. First issued on the intranet September 2003
2. Reviewed 09/04 - location of Biochemistry added.
3. Reviewed 02/11/04 - on call information updated.
4. Reviewed 03/11/04 - expected turnaround times added.
5. Reviewed 10/11/04 - document review history added.
6. Reviewed 01/01/2006 - Quality Standards reviewed by S. Gee
7. Reviewed 01/09/06 - troponin information updated.
8. Reviewed 01/10/06 - occult blood information removed.
9. Removed 01/03/2007 - On call Investigations amended by S. Gee
10. Reviewed 16/05/07 - sample requirements updated.
11. Reviewed 01/06/07 - ascorbic acid information removed & 5HIAA information updated.
12. Reviewed 01/01/08 - Quality Standards updated by S.Gee
13. Reviewed 01/02/08 - On call information updated by S.Gee
14. Reviewed 01/03/08 - On call information updated by S.Gee
15. Reviewed 27/01/09 - magnesium reference range amended by S. Gee.
16. Reviewed 01/04/2011 - Biochemistry personnel amended by S.Stalley
17. Reviewed 01/04/2001 - On call investigations (HCG limits) amended by S.Gee
18. Reviewed 09/06/2011 - Location changed times and added telephone/fax number by S. Stalley
19. Reviewed 09/06/2011 Specimen collection updated for NHS number and fasting status by S. Stalley
20. Reviewed 15/08/2011 5HIAA reference interval and referral lab updated by S.Stalley
21. Reviewed 15/08/2011 Specimen collection updated to include blood gasses, amend CSF XANY times, and labelling criteria by S. Stalley
22. Reviewed 15/08/2011 Quality standard dates corrected by S. Stalley
23. Reviewed 15/02/2012 Specimen collection 24 hr urine patient details updated by S. Stalley
24. Reviewed 15/02/2012Drug interference effects update by S.Stalley
25. Reviewed 27/03/2012 Test Directory A-z updated by S. Stalley
26. Reviewed 27/03/2012 Urine Specimen Collection and Preservation by S.Stalley
27. Reviewed 27/03/12 Acyl Carnitine and AMH pages added to tests by S. Stalley
28. Reviewed 04/04/12 Pages added B12, Ferritin, Folate, Gentamycin and Vancomycin pages added by S.Stalley
29. Reviewed 10/04/12 Thyroglobulin sendaway address updated by S.Stalley
30. Reviewed 12/04/12 Cobalt, Chromium, Complement pages added by S. Stalley
31. Reviewed 25/04/12 PTHrP, Lamotrigine, Inhibin, Lipase pages added by S.Stalley
32. Reviewed 26/04/12 CSF Xanthochromia, POCT, Ethambutol, Isoniazid and Rifampicin added by S.Stalley
33. Reviewed 26/04/12 Biochemistry Quality Standards updated by S.Stalley
34. Reviewed 10/06/14 Biochemistry personnel updated by S.Stalley
35. Reviewed 11/06/14 Emergency and On call investigations expanded to more tests by S.Stalley
36. Reviewed 11/06/14 POCT - QC supply amended by S. Stalley
37. Reviewed 11/06/14 Quality Standards updated by S.Stalley
38. Reviewed 11/06/14 Drug Interference, Interpretation of results and Lipids reviewed with no changes by S.Stalley
39. Reviewed 11/06/14 Endocrinology repertoire updated by S.Stalley
40. Reviewed 11/06/14 Trace metals SAS labs removed updated to referral labs by S.Stalley
41. Reviewed 11/06/14 Updated Therapeutic drug monitoring in mass units
42. Reviewed 13/06/14 Sample requirements ref ranges by S.Stalley
43. Reviewed 16/06/14 Urine preservatives updated by S.Stalley
44. Dynamic Function Test protocols checked 16/06/14 procedurally by S.Stalley require Clinical update by consultant
45. Tests A-M reviewed on 16/06/14 by S.Stalley
46. Tests m-Z reviewed on 18/06/14 by S.Stalley
47. Pages added Methanol, Ethylene Glycol, CTX, Fructosamine, TMA, USteroid, SPSA, LIPO EP, CSF AA, 7 dehydrocholesterol, dihydrotestosterone, MPS1, gauche, fabry, pompe, CDT on 19/09/14 by S.Stalley
48. Clinical review of Troponin T guidelines by Dr T. Likhari, updated by S. Stalley 24/06/14
49. Reviewed by Stephen Gee 14/09/17
50. Following UKAS Assessment – the following have been added preparation of the patient; instructions for transportation of samples; requirements for patient consent; policy on protection of personal information; complaint procedure. Stephen Gee 14/12/17
51. Reviewed post UKAS surveillance visit. Multiple existing change requests incorporated into new draft by R.Nevin 14/12/18
52. Reviewed post NEESPS Dissolution Multiple existing change requests incorporated into new draft by Stephen Gee 05/2021
53. Reviewed post UKAS surveillance visit. Multiple existing change requests incorporated into new draft by Stephen Gee 23/12/2021
54. Reviewed post UKAS surveillance visit. Multiple existing change requests incorporated into new draft by Stephen Gee 16/08/2022

**Quality Standards**

The Clinical Biochemistry Department has attained accreditation from the following professional bodies:

**UKAS (United Kingdom Accreditation Service)**    (<https://www.ukas.com/>)

Reference Number 9318   
Current Date Accredited June 2022   
  
UKAS Clinical Biochemistry - [Accreditation Certificate](https://www.ukas.com/wp-content/uploads/schedule_uploads/00007/9318-Medical-Single.pdf)

**IBMS (Institute of Biomedical Sciences)**    [(http://www.ibms.org/)](http://www.ibms.org/)

This accreditation covers the training of Biomedical Scientists within the department. Date of renewal for this accreditation is October 2026.

Quality Standards

Turnaround Times

Turnaround time for the laboratory is the difference between time of receipt of the sample in the laboratory to the time the result(s) are available.

All urgent requests are time-stamped on receipt but for routine requests the time of booking in the request is the only parameter currently available as a measure of receipt of the sample. The time the result(s) are availableis defined as the time the results are telephoned to the clinician or electronically sent or paper report(s) despatched. This measure does not include such factors as delays from time of delivery to time of booking in however the measured time will show the earliest time at which samples could be analysed and the earliest time results are available.

**Urgent requests,** where prior arrangement has been made with the laboratory

|  |  |  |
| --- | --- | --- |
| Assay | Turnaround Time | Target |
| Blood Gas, Carboxyhaemoglobin (COHB) | 30 Minutes | 95% |
| U/E, Creatinine, Bone, LFT, Lipids, CK, Amylase, Glucose, Gentamicin, Lactate, CRP,  Vancomycin, Troponin T (TNT),  Paracetamol, Salicylate | 60 Minutes | 95% |
| BHCG (inpatient, ?pregnant) | 180 Minutes | 95% |
| Digoxin, Theophylline, Phenytoin, Carbamazepine, Bile Acids, Lithium, Osmolality | 240 Minutes | 95% |

**Urgent requests,** where prior arrangement has been made with a Consultant Chemical Pathologist or Consultant Clinical Scientist

|  |  |  |
| --- | --- | --- |
| Assay | Turnaround Time | Target |
| Lactate Dehydrogenase (LD), Cortisol | 240 Minutes | 95% |

**Ward Routine requests**

|  |  |  |
| --- | --- | --- |
| Assay | Turnaround Time | Target |
| U/E, Bone, LFT, Lipids, CK, Amylase, Glucose | 4 Hours | 95% |
| Digoxin, Theophylline, Phenytoin, Carbamazepine, Bile Acids, Lactate Dehydrogenase (LD), Urine tests, Iron, ACE, Lithium, Osmolality, B12, Folate | 24 Hours | 95% |
| Cortisol, FSH, LH, FT4, TSH, Prolactin, Progesterone, PSA, Testosterone, AFP, TPO, PTH, CA markers, Hba1c, Vitamin D | 3 Days | 95% |
| Specific Proteins, Protein electrophoresis, Paraprotein quantitation, Immunofixation of M band, Calprotectin, IGF-1, Free Light Chains | 7 Days | 95% |
| Renal Calculi | 14 Days | 95% |
| Tests referred to other laboratories | 21 Days | 95% |

**Drug Interference with Test Results**

Many drugs and some natural metabolites may affect results. This normally occurs in one of two ways:

* By direct interference with the laboratory test procedure yielding a higher or lower apparent result for the requested analyte - the in vitro effect.
* By provoking a change in the analyte to be measured through physiological, pharmacological or toxicological processes in the patient - the in vivo effect.

**Direct Interference**

This type of interference is method specific and most are well known and readily recognisable.

**Indirect Interference**

The indirect mode of interference is more important as it accounts for approximately 75% of all known cases of interference and is more difficult to identify.

If there is any doubt about the validity of an analytical result, please check with the laboratory (ext.: 5705) to discuss possible drug interference.

**Interpretation of Results**

**General Considerations**

It is only possible to indicate on reports leaving the laboratory, the briefest of information on how the result presented may be interpreted. Ideally to interpret any single result on a patient, the result should be compared to the results expected in the reference population of which the patient forms a part. To identify the reference population, it may be necessary to know the patient’s:

* Fasting Status
* Age
* Sex
* Posture
* Timing of menstrual cycle

Please note that a reference interval will only appear on the report if age and sex of patient are stated on the request (except for babies <1 month of age). Such abnormal results will be **printed in bold type**.

In addition to these physiological and related factors, it is also necessary to consider these other major potential causes of change in analyte concentration:

* Iatrogenic influences
* The handling of the sample from the moment of removal from the patient
* Analytical precision

Iatrogenic effects on the analyses do not just include effects of the drugs which may have been prescribed for the patient’s present illness, but must also take into account e.g. the contraceptive pill, hormone replacement therapy for post-menopausal females, long-term diuretics, anticoagulants etc.   The effect of “foodstuffs” such as caffeine, alcohol, tobacco etc. may also be relevant. A fuller account of drug interference with test results is given in the following section.

Biotin interference: Please note that a high biotin intake can cause interference with immunoassay results. This includes the following tests: AFP, CA 125, CA 15-3, CA19-9, CEA, cortisol, ciclosporin, DHEAS, digoxin, ferritin, folate, FSH, FT3, FT4, hCG, LH, oestradiol, PTH, procalcitonin, progesterone, prolactin, P1NP, PSA, SHBG, testosterone, TSH, TPO antibodies, troponin and vitamin B12. Patients taking biotin doses >5 mg/day should wait at least 8 hours before a sample is taken. If in doubt, please contact the laboratory.

Sample collection and handling variables include tourniquet application time, ease of flow of blood into the syringe, correct specimen collection bottle, transport delays, centrifugation technique, and storage time and temperature before analysis.

Analytical precision varies from assay to assay, and normally consists of three elements: - chemical, instrumental and human. Whilst the human element is normally thought of in terms of manual dexterity and degree of excellence of manual procedures, human fallibility must never be forgotten - errors can occasionally occur. Quality control sets out to quantify and minimise laboratory imprecision, but does not eliminate it entirely.

**SPECIALISED SERVICES GUIDE (BIO)**

**Acute Kidney Injury**

**No AKI Warning:** No evidence of Injury

**AKI Warning Stage 1:** An increase in serum creatinine of more than or equal to 50% to 100%

(1.5 to 2 times the reference value RV1 or RV2) or an absolute increase of 26 moles/L in the serum creatinine within 48 hours.

**AKI Warning Stage 2:** An increase in serum creatinine of more than or equal to 101% to 200%

(2 to 3 times the reference value RV1 or RV2)

**AKI Warning Stage 3:**  An increase in serum creatinine of more than 200% (More than 3 times the reference value RV1 or RV2) or serum creatinine ≥355μmol/l with an acute rise of at least 45μmol/l

**Proteins**

Assays available:

* Immunoglobulins: IgG, IgA, IgM
* Carrier proteins: Albumin, Caeruloplasmin
* Acute phase proteins: C-reactive protein (CRP)
* Serum and urine protein electrophoresis, including Bence Jones protein
* Kappa and Lambda free light chains
* Identification and quantitation of paraproteins in serum and urine using immunofixation
* Detection of cryoglobulins
* Urine microalbumin

NOTE:   Protein electrophoresis is only of use for the diagnosis of patients suspected of having B-cell malignancy or in the monitoring of such patients. More specific assays should be requested in other clinical situations.

**Principal Applications**

|  |  |
| --- | --- |
| Immunoglobulins: | monitoring immune competence |
| Albumin: | hypoalbuminaemia is a reliable indication of illness but has little diagnostic specificity. It is, however, important in assessing calcium and magnesium status. |
| Caeruloplasmin: | Wilson's disease |
| C-reactive protein: | acute and chronic inflammation |
| Protein electrophoresis: | diagnosis/monitoring of B-cell tumours |

**Sample Requirements**

* 7.5 mL clotted blood (B) for any serum measurement.
* Two clotted blood samples collected in a 7.5 ml non gel tube (white top) and two EDTA samples collected in a 3.4 ml red top tube. An appointment should be made with the phlebotomy clinic at Ipswich Hospital
* Urine Bence Jones protein assays and microalbumin/creatinine ratio require an early morning urine (10ml) collected in a yellow top universal container.
* Many other protein assays can be provided via referral laboratories.  We are able to advise on appropriate tests and sample requirements.

**Blood Lipid Studies**

**Assays Available On Site**

* Cholesterol
* HDL cholesterol
* Non HDL Cholesterol
* LDL cholesterol
* Triglycerides

**Principal Applications**

* Coronary risk assessment
* Dyslipoproteinaemias

**Sample Requirement**

* For cholesterol / HDL cholesterol - fasting is NOT required 7.5 mL blood (B)
* Other assays - fast for 12-14 (not less, not more) hours 7.5 mL blood (B)

**Notes**

Serum triglycerides are subject to major increases following meals and may also be released (as VLDL) after prolonged fasting: the 12-14 hour fast for meaningful triglyceride measurements is therefore critical.  Cholesterol levels can exhibit a seasonal variation and there may be marked day-to-day variations in certain individuals.  
Serum cholesterol measurements during admission for myocardial infarction can be misleading due to marked but variable decreases in the circulating cholesterol level.  Assessment/re-assessment of cholesterol status should be postponed to 3 months post-infarct.

**Endocrinology**

**Assays Available On Site**

* IGF-1
* Free T4
* TSH
* Free T3
* FSH
* LH
* Progesterone
* Prolactin
* Cortisol
* Testosterone
* SHBG
* Estradiol
* Parathyroid Hormone (PTH)
* DHEAS

Some of these assays may be carried out as part of dynamic function tests. Further information can be obtained from that section of this handbook or from the Clinical Biochemistry Department - Ext. 5708.

**Sample Requirement(s)**

* 7.5 mL clotted blood (B) is sufficient for any desired combination of assays
* PTH assays require a blood 3.4 mL (R) as well as 7.5 mL clotted blood (B)

**Trace Metals**

**Assays Available:**

* Lead
* Magnesium
* Zinc
* Copper
* Selenium
* Cobalt
* Chromium

**Principal Applications**

**Copper and Zinc:** Suspected deficiency due to inadequate dietary intake, particularly in patients receiving parenteral nutrition, with malabsorption, or as a result of excessive losses.  Measurements of serum copper together with caeruloplasmin are indicated for the diagnosis of suspected Menke’s syndrome (rare) and Wilson’s disease. Urinary copper excretion is only of value in the investigation of Wilson’s disease.

**Lead:** Blood lead measurement is required for assessment of exposure to inorganic lead as 95% of blood lead is bound to erythrocytes.

**Magnesium:** Suspected deficiency due to poor intake, decreased absorption, or increased loss.

**Sample Requirements**

|  |  |
| --- | --- |
| Copper, Magnesium, Zinc | 7.5 mL blood(B) |
| Copper (urine) | 24 hour collection (no preservative) |
| Lead | 3.4 mL blood (R) |
| Urine Lead | 20 mL urine (no preservative) |
| Cobalt, Chromium, Selenium | 9 mL blood (O) (Special trace metal - no gel tube) |

**Please note the above tests, other than magnesium, are sent away to referral laboratories.**

Other trace metal assays may be available at other referral labs.  
Please note that for some of these assays, a special blood collection tube available from the laboratory is required.   Please contact Ext. 5705.

**Therapeutic Drug Monitoring**

**Assays available on site**

* Carbamazepine
* Digoxin
* Lithium
* Phenytoin
* Theophylline
* Gentamicin
* Vancomycin
* Ciclosporin

**Please note Phenobarbitone and Valproate are not measured on site but are sent to referral laboratories**.

**Principal Applications**

The monitoring of serum levels of those drugs for which there is an established therapeutic window range of serum concentrations between levels that are ineffective and levels which are toxic.   
  
Please note that valproate assay is only of value in assessing patient compliance.

**Sample Requirement**

7.5 mL blood (B) collected at least 6 hours post-dose or pre-dose for Digoxin, and immediately before a dose for all the other drugs listed.  
  
Please state dosage details (dose, time of last dose and time sample collected) on the request form.

**Optimal Therapeutic Serum Concentrations:**

|  |  |
| --- | --- |
| Carbamazepine | 4.0 - 12.0 mg/L |
| Phenytoin | 5.0 - 20.0 mg/L |
| Theophylline | 10.0 - 20.0 mmol/L |
| Digoxin | 0.9 - 2.0 mmol/L |
| Lithium | 0.4 - 1.0 mmol/L |

**NOTES**

The serum concentration may not necessarily reflect the pharmacological effects in any one particular patient as the pharmacokinetics depend upon many factors, including hepatic and renal function, concurrent drug therapy, and the nature of the bio-active drug.  In consequence, the TDM service is supported by the Pharmacy Department who will advise on the interpretation and action in relation to any specific result.

It is advisable that a serum creatinine estimation be requested at the same time as digoxin, lithium and gentamicin levels, so that current renal function can be assessed if it is not already known.

The availability of a TDM service does not imply that every patient on the drug concerned should be monitored. It is particularly important that requests are limited to the comparatively small number of occasions when they are of clinical value.

**Toxicology and Drug Screening**

**Assays Available On Site**

* Salicylate
* Paracetamol
* Ethanol  (clinical, not medico-legal)
* Carboxyhaemoglobin

**Sample Requirement**

|  |  |
| --- | --- |
| **Salicylate and/or paracetamol:** | 7.5 mL clotted blood (B) |
| Note that for assessing the likelihood of liver damage by paracetamol, the blood sample for the paracetamol assay must be taken at least four hours after ingestion of the drug, and the time since ingestion carefully recorded. | |
| **Alcohol:** | 7.5 mL blood in plain tube (B) |
| **Carboxyhaemoglobin:** | 3.4 mL blood in EDTA (R) or heparinised (O) tube or blood gas syringe |

**Interpretation**

**Salicylate:** Poisoning can produce profound metabolic disturbances  
Toxic Levels: Child >300 mg/L  
Toxic Levels: Adult >500 mg/L  
Potentially Lethal: >700 mg/L  
Salicylate may be released from the tissues sometime after ingestion.   Check the blood level after 4 hours if it is thought that toxic levels might be reached.  
  
**Alcohol:** Toxic >150 mg/dL

|  |  |  |  |
| --- | --- | --- | --- |
| **Carboxyhaemoglobin** | | **% of total haemoglobin** | |
| Suburban non-smokers | | 1.5 | |
| Smokers | | 1.5 - 5.0 | |
| Heavy smokers | | 5.0 - 9.0 | |
| Severe poisoning | | >50 | |

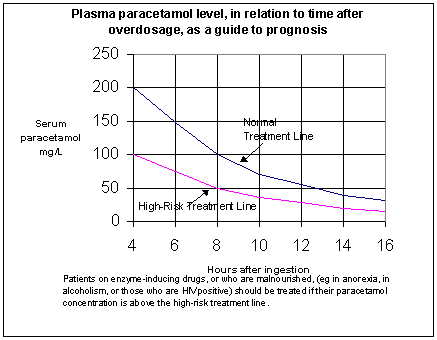
**Paracetamol:**   
Patients whose plasma paracetamol concentrations are above the normal treatment line on the [nomogram](#nomogram) should be treated with acetylcysteine by intravenous infusion (or, provided the overdose has been taken within 10-12 hours, with methionine by mouth).

Patients on enzyme-inducing drugs (e.g. carbamazepine, phenobarbitone, phenytoin, rifampicin, and alcohol) or who are malnourished (e.g. in anorexia, in alcoholism, or those who are HIV-positive) should be treated if their plasma-paracetamol concentrations are above the high-risk treatment line.

**Other Drug Overdoses**

It is not practical for the laboratory to screen for all poisons.  
Further treatment, advice and analytical services can be obtained by contacting the POISONS UNIT (New Cross) at GUY’S HOSPITAL (020 7771 5370 or #6 358) through the hospital switchboard operator.  Bear in mind, however, that not all assays suggested by this unit are available (or indeed necessary.)  
In all suspected overdose cases, the following samples should be collected: Urine in a 30 mL white container, 10 mL clotted blood, and gastric aspirate (where available) to be sent to the laboratory for storage (this will be for 8 weeks) and possible later analysis.

The Biochemistry Department does not undertake monitoring of patients on **restricted drugs or suspicion of addiction**.



**Covid -19 Antibodies**

The Anti-SARS-CoV-2, Nucleocapsid, total antibody assay is available for the qualitative detection of antibodies to SARS-CoV-2. It aids in identifying individuals with an adaptive immune response to SARS-CoV-2, indicating recent or prior infection. This test is recommended for individuals at greater than or equal to 14 days post-symptom onset or following exposure to individuals with confirmed COVID-19. Results from antibody testing should not be used as the sole basis to diagnose or exclude SARS-CoV-2 infection or to inform infection status.

Note: This assay will not detect antibodies induced by currently available SARS-CoV-2 vaccines.

**Interpretation:**

**Negative:** No antibodies to SARS-CoV-2 detected. Negative results may occur in serum collected too soon following infection, in patients who are immunosuppressed, or in patients with mild or asymptomatic infection. Follow-up testing with a molecular test is recommended in symptomatic patients.

**Positive:** SARS-CoV-2 antibodies to the Nucleocapsid protein detected. Results suggest recent or prior infection with SARS-CoV-2. Correlation with epidemiologic risk factors and other clinical and laboratory findings is recommended.

**Covid -19 Spike Antibodies**

The Anti-SARS-CoV-2, Spike, total antibody assay is available for the qualitative detection of antibodies to SARS-CoV-2 spike antigen. It aids in identifying individuals with an adaptive immune response to SARS-CoV-2, indicating recent or prior infection or vaccination. This test is recommended for individuals at greater than or equal to 14 days post-symptom onset or following exposure to individuals with confirmed COVID-19. Results from antibody testing should not be used as the sole basis to diagnose or exclude SARS-CoV-2 infection or to inform infection status or vaccine response.

**Interpretation:**

Negative: SARS-CoV-2 antibodies to Spike protein not detected. Negative results may occur in serum collected too soon following infection or vaccination, in patients who are immunosuppressed, or in patients with mild or asymptomatic infection. Follow-up testing with a molecular test is recommended in symptomatic patients.

Positive: SARS-CoV-2 antibodies to the Spike protein detected. Results suggest recent or prior infection with SARS-CoV-or vaccination. Correlation with epidemiologic risk factors, vaccine history and other clinical and laboratory findings is recommended.

For further information use the following:

[https://www.gov.uk/government/publications/antibody-testing-for-sars-cov-2-key-information/antibody-testing-for-sars-cov-2-information-for-general-practitioners#result-interpretation-and-sars-cov-2-antibody-mechanics](https://www.gov.uk/government/publications/antibody-testing-for-sars-cov-2-key-information/antibody-testing-for-sars-cov-2-information-for-general-practitioners%23result-interpretation-and-sars-cov-2-antibody-mechanics)

**Referral Assay Services**

**Assays Available by Referral**

* Immunologically important proteins and tumour markers
* Metals
* Vitamins
* Therapeutic Drugs
* Genetic enzymes

It is neither possible nor appropriate to give a full list of individual analytes.  If you believe that the assay you require might be referred, contact the laboratory.

**Sample Requirements**

Many of the substances to be measured are labile and special collection/transport arrangements are needed.  Contact the laboratory before you collect any such sample.  
A request form must be fully completed. In all cases give adequate information and indicate current medication. If you think that it might be helpful, write a brief covering letter (in certain instances you may be asked for such a letter anyway).  
Reference intervals are always stated on reports.

**Notes**

Referred services can only be contacted through the laboratory - requests are not accepted directly from individual clinicians.  
In addition to referred services, the laboratory is often able to arrange for other special investigations to be undertaken by colleagues in other clinical biochemistry laboratories. We are always interested to hear of unusual biochemical problems and may well be able to assist with their investigation - telephone ext. 5708 to discuss the problem.

**Troponin Guidelines**

Troponin T (TnT) is a component of the contractile apparatus of the striated musculature. Although the function of TNT is the same in all striated muscles, TNT originating exclusively from the myocardium (cardiac TNT, molecular weight 39.7 kDa) clearly differs from skeletal muscle TNT. As a result of its high tissue specificity, cardiac troponin T (c TNT) is a cardio-specific, highly sensitive marker for myocardial damage.

Cardiac troponin T (cTNT) is an independent prognostic marker which can predict the near, mid and even long term outcome of patients with acute coronary syndrome (ACS).

Cardiac troponin T is also useful to identify patients that benefit from anti-thrombotic therapy (GPIIb /IIIa inhibitors, low molecular weight heparin).

Low concentrations of troponin T can be detected in clinically stable patients such as patients with ischaemic or non-ischaemic heart failure, patients with different forms of cardiomyopathy, renal failure, sepsis and diabetes.

Elevated levels of troponin T correlate with the severity of coronary artery disease and to poor outcome independent of natriuretic peptide (BNP or NT-pro BNP) levels.

Myocardial cell injury leading to elevated cTNT concentrations in the blood can also occur in other clinical conditions such as myocarditis, heart contusion, atrial fibrillation, pulmonary embolism and drug induced cardiotoxicity.

**Current Criteria of Interpretation of Results:**

1. All results to be considered with all other criteria necessary to diagnose myocardial infarction (MI)   
2. 2 samples are required, 6 hours apart  
3. Peak sample:  
a) <14 ng/L – Negative, b) 14 - 100 ng/L - Intermediate (an increment between the samples of > 7 is suggestive of MI), c) >100 ng/L - High probability.

**SAMPLE REQUIREMENTS**

**Sample Requirements and Reference Intervals**

Sample requirements are listed showing the recommended collection tube. In some instances alternative tubes may be used, e.g. the full range of tubes is available for paediatric samples. Contact the laboratory if in doubt. The volumes indicated are to identify the correct tube only and do not imply that more than one Monovette is required for a selection of tests. If in doubt about minimum volumes for given assays, please contact the laboratory to discuss (Ext. 5705).

|  |  |  |
| --- | --- | --- |
| (B) | Brown top | Clotted sample |
| (W) | White top | Clotted sample - no separating gel |
| (R) | Red top | EDTA |
| (O) | Orange top | Lithium heparin |
| (Y) | Yellow top | Fluoride |

**PLEASE NOTE**:  The intervals given are for adults. Note that some intervals vary with age and sex. The intervals below are for guidance only as all printed reports contain the appropriate age/sex related intervals.

Please click on blue hyperlinks on each test for more information

|  |
| --- |
|  |

|  |  |  |
| --- | --- | --- |
| **ANALYTE** | **SAMPLE** | **REFERENCE** |
| Acyl Carnitine | Guthrie card blood spots |  |
| Adrenocorticotrophic hormone (ACTH) | Blood 2 x 3.44 mL (R) Send to laboratory immediately on ice | 5-50 ng/L |
| Alanine transaminase (ALT) | Blood 7.5 mL (B) | Males: 0-41 U/L  Females: 0-33 U/L |
| Albumin | Blood 7.5 mL (B) | 35-50 g/L |
| Albumin / Creatinine ratio (ACR) | Random urine | 0 – 2.9 mg/L |
| Alcohol (ethanol) Post-mortem/clinical samples only | Blood 7.5 mL (B) | Toxic >150 mg/dL  Potentially Lethal >350 mg/dL |
| Aldosterone | Blood 1 x 3.4 mL (R) or Blood 7.5 mL (B) | Recumbent 100-450 nmol/L Ambulant 100-800 nmol/L **Separate tube for renin if required** |
| Alkaline phosphatase | Blood 7.5 mL (B) | 30-130 IU/L (age/sex related) |
| Alkaline Phosphatase Isoenzymes | Blood 7.5 mL (B) | Descriptive report |
| Alkaline Phosphatase - BONE | Blood 7.5 mL (B) | Males: >20yrs 15-41 U/L, Females: >15yrs 11-31 U/L |
| Alpha-1 antitrypsin | Blood 7.5 mL (B) | 0.9-2.0 g/L |
| Alpha-1 antitrypsin phenotype | Blood 7.5 mL (B) | Descriptive report |
| Alpha fetoprotein (AFP) | Blood 7.5 mL (B) | <5.8 kU/L |
| Aluminium | Blood 9 mL. Special tube required. Contact laboratory. | <0.6 µmol/L |
| Amino acids | Blood 2.0 mL (O) | Numeric report and description |
| CSF Amino Acids | CSF |  |
| Amino acids (urine) | Random urine | Descriptive report |
| Amiodarone | Blood 3.4 mL (R) | 0.5-2.0 mg/L (pre- dose) |
| Ammonia | Blood 1.3 mL (screwtop), special collection bag required, please contact laboratory. | 0-80 µmol/L (<5 weeks old)  0-40 µmol/L (>5 weeks old) |
| Amylase | Blood 7.5 mL (B) | 28-100 U/L |
| Androstenedione | Blood 7.5 mL (B) | Males: 0-13 yrs <3.6 nmol/L,  Males:14-99yrs 4.4-10.9 nmol/L, Females: 0-13yrs <3.6 nmol/L,  Females: 14-99yrs 4.0-10.2 nmol/L |
| Angiotensin converting enzyme (ACE) | Blood 7.5 mL (B)  Samples MUST be separated from cells or processed within 4 hours of venepuncture. | 8 - 65 U/L |
| Anti Mullerian Hormone (AMH) | Blood 7.5mL (B) |  |
| Anti-SARS-CoV-2 (Nucleocapsid) | Blood 7.5mL (B) |  |
| Anti-SARS-CoV-2 (Spike) | Blood 7.5mL (B) |  |
| Aspartate transaminase (AST) | Blood 7.5 mL (B) | Males: 0 - 40 U/L Females: 0-32 U/L |
| Beta-2 microglobulin | Blood 7.5 mL (B) | 1.2-2.4 mg/L |
| Beta-2 microglobulin (urine) | Random urine | No longer available |
| Bicarbonate (serum) | Blood 7.5 mL (B) | 22-29 μmol/L |
| Bile acids | Blood 7.5 mL (B) | <10 µmol/L |
| Bilirubin (conjugated) | Blood 7.5 mL (B) | 0-3 μmol/L |
| Bilirubin (total) | Blood 7.5 mL (B) | 0-20 μmol/L |
| Bilirubin (unconjugated) | Blood 7.5 mL (B) | 0-15 μmol/L |
| Bismuth | Blood 3.4 mL (R) |  |
| NT-proBNP | Blood 7.5 mL (B) | Normal levels (<400 pg/mL), Elevated levels (400-2000), Very high levels (>2000) |
| CA125 | Blood 7.5 mL (B) | 0-35 U/mL |
| CA15-3 | Blood 7.5 mL (B) | <28.5 kU/L |
| CA19-9 | Blood 7.5 mL (B) | 0-34 U/mL |
| Cadmium | Blood 3.4 mL (R) |  |
| Caeruloplasmin | Blood 7.5 mL (B) | 0.20- 0.50 g/L |
| Calcitonin | Blood 9 mL (B). Send to lab immediately on ice. | Females: <5.5 ng/L Males: <18.9 ng/L |
| Calcium | Blood 7.5 mL (B) | >17 years 2.20-2.60 mmol/L |
| Calcium (adjusted for albumin) | Blood 7.5 mL (B) | >17 years 2.20 - 2.60 mmol/L |
| Calcium (urine) | Random urine | 2.50 - 7.50 mmol/L |
| Calcium (24 hr urine) | 24 hr urine | 2.5-7.5 mmol/d |
| Calculus |  |  |
| Calprotectin | Random faeces | 0-50 µg/g faeces |
| Carbamazepine | Blood 7.5 mL (B) | 4.0 - 12.0 mg/L |
| Cocaine and Amphetamine Regulator Transcript (CART) | Blood 2 x 3.4 mL (R) EDTA. Send to laboratory immediately on ice. | No longer available |
| Carbohydrate Deficient Transferrin (CDT) | Blood 7.5 mL (B) |  |
| Carbon dioxide (CO2) serum | Blood 7.5 mL (B) | 22-29 mmol/L |
| Carboxyhaemoglobin | Blood 7.5 mL (O) | Non Smoker 0-1.5% total Hb Smoker 1.5-5.0 % total Hb |
| Carcino-embryonic antigen (CEA) | Blood 7.5 mL (B) | 0 - 4.7 ng/mL |
| Chloride | Blood 7.5 mL (B) | 95-108 mmol/L |
| Chloride (sweat) | Sweat | Sweat chloride < 30 mmol/L is normal - low probability of Cystic fibrosis |
| Chloride (urine) | 24 hr urine | Adults: 110-250 mmol/d |
| Cholesterol | Blood 7.5 mL (B) |  |
| Cholesterol (HDL) | Blood 7.5 mL (B) | Moderate risk male 0.9-1.45 mmol/L Moderate risk female 1.15-1.68 mmol/L |
| Cholesterol (LDL) | Blood 7.5 mL (B) | <4.0 mmol/L |
| Cholinesterase (organo-phosphate poisoning) | Blood 3.4 mL (R) | Descriptive report |
| Cholinesterase phenotype (dibucaine number etc.) | Blood 7.5 mL (B) | Descriptive report |
| Chromium | Blood 7.5 mL (O) - Orange Top. Trace metal tube. | 0 - 20 nmol/L |
| Chromogranin A | Blood 2 x 3.4 EDTA. Send to laboratory immediately on ice. | 0-60 pmol/L |
| Chromogranin B (GAWK) | Blood 2 x 3.4 mL (R) EDTA. Send to laboratory immediately on ice. | 0-150 pmol/L |
| Citrate (urine) | 24 hr urine with hydrochloric acid preservative. | 1.60-4.50 mmol/d |
| CK (total) | Blood 7.5 mL (B) | Males: 40-320 U/L Females: 25-200 U/L |
| CO2 (Serum) | Blood 7.5 mL (B) | 22-29 mmol/L |
| Cobalt | Blood 7.5mL (O) - Orange Top. Trace metal tube. | 0-17 nmol/L |
| Complement C3 | Blood 7.5 mL (B) | 0.9 – 1.8 g/L |
| Complement C4 | Blood 7.5 mL (B) | 0.1 – 0.4 g/L |
| Copper | Blood 7.5 mL (B) | 3.0-11.0 mmol/L (0-6 weeks) 12.0–26.0 mmol/L (>6 weeks) |
| Copper (urine) | 24 hr urine | 0-1.0 mmol/d |
| Cortisol | Blood 7.5 mL (B) | 9am Cortisol 155-605 nmol/L Midnight Cortisol 40-210 nmol/L |
| Cortisol (urine free cortisol) | 24 hr urine | 50-280 nmol/d |
| Covid-19 (Nucleocapsid) Antibodies | Blood 7.5 mL (B) | Cut Off Index < 1.0 Non-reactive Negative for anti‑SARS‑CoV‑2 antibodies. Cut Off Index ≥ 1.0 Reactive Positive for anti‑SARS‑CoV‑2 antibodies |
| Covid-19 Spike Antibodies | Blood 7.5 mL (B) | <0.80 U/mL |
| C peptide | Blood 7.5 mL(B) and Blood 2.7 mL (Y) | Send to laboratory immediately |
| C Reactive Protein (CRP) | Blood 7.5 mL (B) | 0-5 mg/L |
| Creatine kinase (CK) | Blood 7.5 mL (B) | Males: 40-320 U/L Females: 25-200 U/L |
| Creatinine | Blood 7.5 mL (B) | 59-104 µmol/L (Adult males) 45-84 μmol/L (Adult females) |
| Creatinine Clearance | 24 hr urine + concurrent blood sample | 70-120 mL/min |
| Creatinine (urine) | 24 hr urine | Males: 3.54-24.6 mmol/d  Females: 2.55-20.0 mmol/d |
| Cryoglobulin | 2X Blood 7.5 mL (W) in non-gel tube and 2X Blood 3.4 mL (R), appointments to be made with the phlebotomy clinic at Ipswich hospital. | Positive / Negative |
| Ciclosporin | Blood 3.4 mL (R) | Samples should be taken >12 hrs post dose |
| CTX Beta Crosslaps | Blood 3.4 mL (R) |  |
| Cystine | Random or 24 hr urine | Positive / Negative |
| Dehydroepiandrosterone sulphate (DHEAS) | Blood 7.5 mL (B) | 19-24 yrs = 1.77-9.99 µmol/L 24-34 yrs = 4.02-11 µmol/L 34-44 yrs = 2.68-9.23 µmol/L 44-54 yrs = 1.65-9.15 µmol/L 54-64 yrs = 0.96-6.95 µmol/L 64-74 yrs = 0.51-5.56 µmol/L 74-120 yrs = 0.26-6.68 µmol/L |
| 11-deoxycortisol | Blood 7.5 mL (B) | Contact laboratory |
| Digoxin | Blood 7.5 mL (B)  collected pre-dose or >6 hr post dose. | 0.9-2.0 µg/L  If results are unexpectedly high, consider digoxin-like immunoreactive substance (DLIS) cross-reactivity. |
| 7-Dehydrocholesterol | Blood 3.4 mL (R) |  |
| Dihydrotestosterone | Blood 7.5 mL (B) or Blood 3.4 mL (R) | 0.90 - 2.90 nmol/L |
| Drug screen | Random urine | Descriptive report |
| Drugs of abuse | Random urine | Consent required for all <16 yrs |
|  |  |  |
| Ethambutol - for compliance testing | Blood 7.5 mL (B) | 2 -6 µg/L |
| Ethosuximide | Blood 7.5 mL (B) | 40-80 mg/L |
| Faecal elastase | Random stool | >200 µg/g |
| Ethylene Glycol | Blood 3.4 mL (R) | Contact department before requesting |
| FK 506 (Tacrolimus) | Blood 3.4 mL (R) |  |
| Ferritin | Blood 7.5 mL (B) | Males: 20-60yrs 30-400 ng/mL Females: 17-60yrs 13-150 ng/mL |
| Fibroblast Growth Factor (FGF- 23) | Blood 3.4 mL (R) | <100 RU/mL |
| Folate | Blood 7.5mL (B) | <3 µg/L suggests deficiency |
| Follicle Stimulating Hormone (FSH) | Blood 7.5 mL (B) | Female Follicular phase 3.5-12.5 U/L Female Ovulation phase 4.7-21.5 U/L Female Luteal Phase 1.7-7.8 U/L Female Perimenopausal >35 U/L Males: 1.5-12.4 U/L |
| Free Androgen Index | Blood 7.5 mL (B) - Calculated from Testosterone and Sex Hormone Binding Globulin |  |
| Free Light Chains | Blood 7.5 mL (B) | Kappa light chains 3.30-19.40 mg/L Lambda light chains 5.71-26.30 mg/L  Kappa:Lambda ratio 0.26-1.65 |
| Free T3 | Blood 7.5 mL (B) | 3.1 – 6.8 pmol/L |
| Free T4 | Blood 7.5 mL (B) | 12.0 – 22.0 pmol/L |
| Fructosamine | Blood 7.5mL (B) | 205-285 μmol/L |
| Galactosidase | See white cell enzymes |  |
| Galactose-1-Phosphate Uridyl Transferase | Dried Blood Spots | Descriptive report |
| Gamma GT | Blood 7.5 mL (B) | Males: 0-60 U/L  Females: 0-40 U/L |
| [Gamma glutamyl transferase (GGT)](http://intranetapps/pathwebfiles/testinfo_ggt.html) | Blood 7.5 mL (B) | Males: 0-60 U/L  Females: 0-40 U/L |
| Gastrin | Blood 2x 3.4 mL (R) EDTA. Send to laboratory immediately on ice. | 0-40 pmol/L |
| Gentamycin | Blood 7.5 mL (B) |  |
| Globulin | Blood 7.5 mL (B) | 17-35 g/L |
| Glucagon | Blood 2x 3.4 mL (R) EDTA. Send to laboratory on ice. | 0-50 pmol/L |
| Glucose | Blood 2.7 mL (Y) | 3.0-6.0 mmol/L = fasting |
| Glucose (CSF) | CSF 2.7 mL (Y) | Contact laboratory |
| Glycosaminoglycans (mucopolysaccharides) | Random urine |  |
| Growth Hormone | Blood 7.5 mL (B) | Contact laboratory |
| Haemoglobin A1c (HbA1c) | Blood 3.4 mL (R) | Non-Diabetic 20-45 mmol/mol Diabetic ideal control 48-58 mmol/mol |
| Haptoglobin | Blood 7.5 mL (B) | 0.5-2.6 g/L |
| ßhCG (Tumour Marker) | Blood 7.5 mL (B) | 0-5 IU/L |
| HCG (?ectopic pregnancy) | Blood 7.5 mL (B) |  |
| Homocysteine (total) - for investigation of arteriosclerosis risk | Blood 9 mL (O) | Send to laboratory immediately |
| Homocystine (free) - for investigation of inborn errors of metabolism | No longer available |  |
| Homovanillic acid | 24 hr urine or plain universal |  |
| 5-Hydroxy-indole-acetic acid (5HIAA) | 24hr urine with glacial acetic acid preservative | <47 µmol /24 hours |
| 17-hydroxyprogesterone | Blood 7.5 mL (B) |  |
| 17-hydroxy progesterone | Blood Spot |  |
| 17-hydroxy progesterone | Saliva |  |
| Hydroxyproline | 24hr urine | 110-370 µmol/d  110-290 μmol/mmol creat |
| Immunoglobulin A (IgA) | Blood 7.5 mL (B) | Age/sex related, contact laboratory |
| Immunoglobulin E (IgE) | Please refer to haematology |  |
| Immunoglobulin G (IgG) | Blood 7.5 mL (B) | Age/sex related, contact laboratory |
| Immunoglobulin G (subclasses 1,2,3,4) | Blood 7.5 mL (B) | Age/sex related, contact laboratory |
| Immunoglobulin M (IgM) | Blood 7.5 mL (B) | Age/sex related, contact laboratory |
| Inhibin A and B | Blood 7.5 mL (B) |  |
| Insulin | Blood 7.5 mL (B) and Blood 2.7 mL (Y) | Send to laboratory immediately |
| Insulin like Growth Factor-1 (IGF-1) | Blood 7.5 mL (B) | Age/sex related, contact laboratory |
| Iron | Blood 7.5 mL (B) | Males: 11.0-28.0 µmol/L Females: 6.6-26.0 μmol/L |
| Isoniazid - for compliance testing | Blood 2.7 mL (Y) | 2 -10 µg/L |
| Lactate | Blood 2.7 mL (Y) | 0.5-2.2 mmol/L |
| Lactate (CSF) | CSF 2.7 mL (Y) | 1.1-2.4 mmol/L |
| Lactate dehydrogenase (LD) | Blood 7.5 mL (B) | 240-480 U/L |
| Lamotrigine | Blood 7.5 mL (B) | 1 -15 mg/L |
| Lead | Blood 3.4 mL (R) | 0-0.24 µmol/L |
| Lipase | Blood 7.5mL (B) | 6-51 IU/L |
| Lipoprotein Electrophoresis | Blood 7.5mL (B) | Free Text |
| Lithium | Blood 7.5 mL (B) | 0.4-1.0 mmol/L |
| Luteinising hormone (LH) | Blood 7.5 mL (B) | Female Follicular phase 2.4-12.6 U/L Female Ovulation phase 14.0-95.6 U/L Female Luteal phase 1.0-11.4 U/L Female Post-Menopausal 7.7-58.5 U/L Male 1.7-8.6 U/L |
| Magnesium | Blood 7.5 mL (B) | 0.7-1.0 mmol/L |
| Magnesium (urine) | 24 hr urine with hydrochloric acid preservative | 2.4 - 6.5 mmol/d |
| Maternal serum screen | Blood 7.5 mL (B) | Descriptive report |
| Manganese | Blood 9.0 mL (O) |  |
|  |  |  |
| Melanin | Random urine. Send to laboratory immediately. | Positive / Negative |
| Mercury (blood) | Blood 7.5 mL (O) or Blood 3.4 mL (R) | Normal <30 nmol/L  Hazardous >250 nmol/L |
| Mercury (urine) | Random early morning urine or 24 hr collection | Normal <50 nmol/L <5 μmol/mol creatinine |
| Metanephrines (urine) | 24 hr urine collection  (Random urine no preservative for children (mainly)) |  |
| Metanephrines (plasma) | EDTA or Citrate Plasma |  |
| Methaemalbumin | Blood 7.5 mL (O) or Blood 3.4 mL (R) freshly collected | 0-6 mg/L |
| Methaemoglobin | Blood 7.5 mL (O) | <1.5 %Hb |
| Methanol | Blood 3.4 mL (R) |  |
| Microalbumin / Creatinine ratio | Random urine | MAU/Creatinine ratio 0 – 2.9 mg/L |
| Mucopolysaccharides (Glycosaminoglycans) | Random urine |  |
| Myoglobin | Random urine | Not routinely available |
| Neurotensin | No longer available - see CART(Cocaine and amphetamine regulator transcript) |  |
| N-Telopeptide X-links | Random urine | Females: <65 μmol/mol Males: <51 μmol/mol |
| Occult blood (FIT) | Random faeces | <10 µg Hb/g faeces |
| Oestradiol (17-Beta) | Blood 7.5 mL (B) | Females: Follicular phase 45 - 854 pmol/L Ovulation phase 151 - 1461 pmol/L Luteal phase 82 - 1251 pmol/L Post menopause 0 - 505 pmol/L 1st trimester pregnancy 563 - 11902 pmol/L Males: 41 - 159 pmol/L |
| Organic acids | Random urine | Descriptive report |
| Osmolality | Blood 7.5 mL (B) | 280-295 mOsm/kg |
| Osmolality (urine) | Random urine | Depends on clinical scenario |
| Oxalate (urine) | 24 hr urine | Males: 0.08-0.49 mmol/d Females: 0.04-0.34 mmol/d |
| Procollagen 1 N-terminal peptide (P1NP) | Blood 7.5 mL (O) | Pre-menopause <58.6 µg/L Post-menopause on HRT <58.8 µg/L Post-menopause no HRT <76.3 µg/L |
| pCO2 (arterial) | Blood (PICO syringes) | Females: 4.3-6.0 kPa Males: 4.7-6.4 kPa |
| pH (arterial) | Blood (PICO syringes) | 7.35-7.45 |
| pO2 (arterial) | Blood (PICO syringes) | 11.0-14.4 kPa |
| Pancreatic polypeptide | Blood 2x 9 mL (O). Contact laboratory BEFORE sample collection. | 0-300 pmol/L |
| Paracetamol | Blood 7.5 mL (B) collected 4-12 hr post-ingestion | See nomogram |
| Parathormone | Blood 3.4 mL (R) and Blood 7.5 mL (O) | 1.6-6.9 pmol/L |
| PTH related peptide (PTH rP) | Blood 3.4 mL (R) send to lab immediately | Contact laboratory before collection |
| Paraprotein | Blood 7.5 mL (B) | Descriptive report |
| Paraprotein (urine) | Random urine | Descriptive report |
| Paraquat | Random urine | Positive / Negative |
| Phenobarbitone | Blood 7.5 mL (B) | 40-170 μmol/L |
| Phenytoin | Blood 7.5 mL (B) | 10 -20 mg/L |
| Phosphate | Blood 7.5 mL (B) | 0.8-1.5 mmol/L |
|  |  |  |
| Phosphate (urine) | 24 hr urine | 15.0-50.0 mmol/d |
| Phytanic acid | Blood 3.4 mL (R) | <1 yr 0-10 μmol/L 1 yr upwards 0-15 μmol/L |
| Porphobilinogen | Random urine or 24 hr collection | Protect sample from light |
| Porphyrins | Blood 3.4 mL (R) | Protect sample from light |
| Porphyrins (faeces) | Random faeces | Protect sample from light |
| [Porphyrins (urine)](http://intranetapps/pathwebfiles/testinfo_porphyrin.html) | Random urine or 24 hr collection | Protect sample from light |
| Porphyrins | For screening for porphyria send random urine, random faeces & EDTA blood | Protect samples from light |
| Potassium | Blood 7.5 mL (B) | 3.5-5.3 mmol/L |
| Potassium (urine) | Random urine | 20-60 mmol/L |
| [Potassium (urine)](http://intranetapps/pathwebfiles/testinfo_potassiumurine.html) | 24 hr urine | 25 -125 mmol/d |
| Pregnancy test | Random urine | Not routinely available |
| Pristanate | Blood 3.4 mL (R) | <1 year 0-1 μmol/L 1-10 years 0-2 μmol/L >10 years 0-2 μmol/L |
| Procalcitonin | Blood 7.5 mL (B) | <0.5 ng/mL |
| Procollagen III N-terminal peptide | Blood 9.0 mL(W) Non-gel tube | Adults: 1.7-4.2 µg/L |
| Progesterone | Blood 7.5 mL (B) | >30 nmol/L indicates ovulation |
| Prolactin | Blood 7.5 mL (B) | 0-500 mU/L |
| Prostate specific antigen (PSA) | Blood 7.5 mL (B) | Males under 48 = <2.0 ng/mL 49-58 = <3.9 ng/mL 59-68 = < 5.4 ng/mL 68-110 = <6.2 ng/mL |
| Sensitive PSA | Blood 7.5mL (B) | 0-4.0 µg/L |
| Protein | Blood 7.5 mL (B) | 60-80 g/L |
| Protein electrophoresis | Blood 7.5 mL(B) or Random urine | Descriptive report |
| Protein (urine) | Random urine | 0-0.14 g/L |
| Protein (urine) | 24 hr urine | 0-0.15 g/d |
| Protein (CSF) | CSF 2.7 mL (Y) | 0.15-0.40 g/L |
| Rapamycin | See Sirolimus |  |
| RAST (specific IgE) | Please refer to haematology |  |
| Reducing substances (faeces) | No longer available |  |
| Reducing substances (urine) | No longer available |  |
| Renin | Blood 2x 3.4 mL (R). Separate tube for aldosterone |  |
| Rifampicin - for compliance testing | Blood 7.5 mL (B) | 8 -24 µg/L |
| Rheumatoid Factor | Blood 7.5 mL (B) |  |
| Salicylate | Blood 7.5 mL (B) |  |
| Selenium | Blood 9 mL (O) (Special trace metal - no gel tube) |  |
| Sex Hormone Binding Globulin (SHBG) | Blood 7.5 mL (B) | Females: 20-49yrs 32.4-128 nmol/L Females: >50yrs 27.1- 128 nmol/L Males: 20-49yrs 18.3-54.1 nmol/L Males: >50yrs 20.6-76.7 nmol/L |
| Sirolimus | Blood 3.4 mL (R) |  |
| Sodium | Blood 7.5 mL (B) | 133-146 mmol/L |
| Sodium (urine) | Random urine | 50-125 mmol/L |
| Sodium (urine) | 24 hr urine | 40-220 mmol/d |
| Urinary Steroid Profile | 24 hr plain urine collection | Descriptive report |
| Tacrolimus (FK506) | Blood 3.4 mL (R) |  |
| Testosterone | Blood 7.5 mL (B) | Females: 20-49yrs 0.29-1.67 nmol/L Females: >50yrs 0.10-1.42 nmol/L Males: 20-49yrs 8.64-29.0 nmol/L Males: > 50yrs 6.68- 25.7 nmol/L |
| Theophylline | Blood 7.5 mL (B) | 10.0-20.0 mg/L |
| Thiopurine methyl transferase (TPMT) | Blood 3.4 mL (R) |  |
| Thyroglobulin | Blood 7.5 mL (B) | 0-35 µg/L |
| Thyroid peroxidase antibody (aTPO) | Blood 7.5 mL (B) | <34 mIU/L = Negative 34 mIU/L = Equivocal >34 mIU/L = Positive |
|  |  |  |
| Thyroid Stimulating Hormone (TSH) | Blood 7.5 mL (B) | 0.27 – 4.20 mIU/L |
| Tryptase(Mast Cell tryptase) | Blood 3.4 mL (R) |  |
| TSH Receptor Antibody | Blood 7.5 mL (B) | Healthy ≤1.22 IU/L  Thyroid disease ≤1.58 IU/L |
| Thyroid Stimulating Immunoglobulin | Blood 7.5 mL (B) | Contact laboratory |
| Thyroxine (Free) | Blood 7.5 mL (B) | 12-22 pmol/L |
| Total CO2 (serum) | Blood 7.5 mL (B) | 22-29 mmol/L |
| TPMT | Blood 3.4 mL (R) |  |
| Transferrin | Blood 7.5 mL (B) | 2.0 – 3.6 g/L |
| Triglyceride | Blood 7.5 mL (B) Fasting | 0.3-2.3 mmol/L |
| Troponin T | Blood 7.5 mL (B) | <14 ng/L |
| Trimethylamine | Random or 24hr collection with HCL | 2.5 - 10.9 μmol/mmol creatinine |
| Urea | Blood 7.5 mL (B) | 2.5-7.8 mmol/L |
| Urea (urine) | Random urine | 125-500 mmol/L |
| Urea (urine) | 24 hr urine | 428-714 mmol/d |
| Uric acid | Blood 7.5 mL (B) | Females: 140-360 μmol/L Males: 200-430 μmol/L |
| Uric acid (urine) | 24 hr urine | 1200 - 5900 μmol/d |
| Valproate | Blood 7.5 mL (B)  FOR  ASSESSMENT OF COMPLIANCE ONLY | 50-100 mg/L |
| Vancomycin | Blood 7.5 mL (B) | Therapeutic vancomycin levels should be between 10 and 15 mg/L |
| Vasoactive Intestinal Peptide (VIP) | Blood 2x 3.4 mL (R) EDTA. Send to laboratory immediately on ice. | 0-30 pmol/L |
| Very long chain fatty acids | Blood 3.4 mL (R) | Descriptive report |
| Vitamin A | Blood 7.5 mL (O)  Protect from light | Age/sex related. Contact laboratory. |
| Vitamin B profile | 2x Blood 9 mL (O) | Protect sample from light |
| Vitamin B12 | Blood 7.5 mL (B) | 197-771 pg/mL |
| Vitamin D   (25-hydroxy cholecalciferol) | Blood 7.5 mL (B) |  |
| 1,25 dihydroxy Vitamin D | Blood 7.5 mL (B) | 48 - 120 pmol/L |
| Vitamin E | Blood 7.5 mL (O)  Protect from light |  |
| White cell enzymes see also MPS1, Gaucher, Pompe, Fabry screen | Blood 3.4 mL (R). Contact laboratory BEFORE sample collection. | Descriptive report |
| CSF Xanthochromia | CSF (plain) and Blood 2.7 mL (Y), Blood 7.5 mL (B) | Protect CSF sample from light |
| Zinc | Blood 7.5 mL (B) | 11.0-24.0 mmol/L |

**REFERENCE RANGES FOR THE MOST COMMON BLOOD TESTS**

|  |  |  |  |
| --- | --- | --- | --- |
| **ANALYTE** | **REFERENCE** | **UNITS** | **COMMENTS** |
| Alanine aminotransferase | Females: 0-33 Males: 0-41 | U/L |  |
| Albumin | 35-50 | g/L |  |
| Alkaline phosphatase | 30-130 | U/L | Age-related |
| Amylase | 28-100 | U/L |  |
| Bicarbonate | 22-29 | mmol/L |  |
| Bilirubin | <21 | µmol/L |  |
| Calcium | 2.2-2.6 | mmol/L | nb albumin |
| Chloride | 95-108 | mmol/L |  |
| Cholesterol |  | mmol/L | nb desirable range |
| CK | Females: 25-200 Males: 40-320 | U/L |  |
| pCO2 | Females: 4.3-6.0 Males: 4.7-6.4 | kPa | Pulsator syringe |
| Creatinine |  | µmol/L | Age-related |
| CRP | <5 | mg/L |  |
| Gamma GT | Females: 0-40 Males: 0-60 | U/L |  |
| Globulin | 17-35 | g/L |  |
| Glucose | 3.0-6.0 | mmol/L | Fluoride tube (fasting) |
| pH | 7.35-7.45 |  | Pulsator |
| HbA1c | Non-Diabetic 20-45 Diabetic Ideal Control 48-58 | mmol/mol | EDTA tube |
| HDL-cholesterol | Females: 1.15-1.68 Males: 0.9-1.45 | mmol/L |  |
| IgG | Age-related | g/L |  |
| IgA | Age-related | g/L |  |
| IgM | Age-related | g/L |  |
| LD or LDH | 240-480 | U/L |  |
| LDL-cholesterol | 0-4.0 | mmol/L |  |
| Magnesium | 0.7-1.0 | mmol/L |  |
| pO2 | 11-14.4 | kPa | Pulsator |
| Osmolality | 280-295 | mOsm/kg |  |
| Potassium | 3.5-5.3 | mmol/L |  |
| Phosphate | 0.8-1.5 | mmol/L |  |
| Sodium | 133-146 | mmol/L |  |
| Thyroxine (free T4) | 12-22 | pmol/L |  |
| Triiodothyronine (Free T3) | 3.1-6.8 | pmol/L |  |
| Triglyceride | 0.3-2.3 | nmol/L |  |
| TSH | 1-65 yrs 0.27-4.2 >65 yrs 0.2-5.7 | mIU/L |  |
| Urea | 2.5-7.8 | mmol/L |  |
| Uric acid | Females: 140-360 Males: 200-430 | μmol/L |  |

N.B. The intervals quoted are intended as a guide only, and should not be regarded as rigid limits.

The majority of analyses are performed on serum using the brown 7.5 mL Sarstedt tubes, unless otherwise stated.

Reference intervals for the less common tests are available from the laboratory, and are given on the hard copy reports.

For information on assays or for clinical reference use: <http://labtestsonline.org.uk/>

[**DYNAMIC FUNCTION TEST PROTOCOLS**](http://intranetapps/pathwebfiles/BIO_DYNAMICFUNC_HEAD.html)

## Multiple Blood Sampling Technique

Many dynamic function tests require several blood samples, and to spare the patient repeated venepuncture an indwelling cannula is placed in a suitable forearm vein, from which multiple samples may be obtained without discomfort.  Which cannula is used is a matter of personal preference: the “Venflon” type tends to be more secure but probably more traumatic, while the “Butterfly” is perhaps less reliable for prolonged sampling, but is no more painful than a simple venepuncture.  After introduction of the cannula a small volume (0.5 mL) of heparinised saline (“Hepsal”) is injected to prevent clotting.  At each sampling 2 mL of blood is withdrawn and discarded (the dead space), then the appropriate volume for the test obtained.  Again the cannula is flushed through with a small volume of heparinised saline.

## Anterior Pituitary

#### Insulin Tolerance Test

This test is potentially dangerous, and fatalities have been reported. It should only be performed under constant medical supervision. Contraindications include ischaemic heart disease, epilepsy, and a random serum cortisol concentration of <100 nmol/L. The stress of hypoglycaemia is used to stimulate secretion of ACTH and growth hormone. Note that in many cases ACTH/adrenal function may be assessed using the short Synacthen test, thus avoiding the use of insulin.  
Intravenous glucose 50% and hydrocortisone 200 mg for IV injection must be readily available.

#### Procedure

After fasting from midnight, at 08.00 - 09.00 hrs the patient is weighed and an indwelling cannula placed in a forearm vein, kept patent with Hepsal. After 30 minutes (time 0) Actrapid insulin is given as an IV bolus. The standard dose is calculated as 0.15 units insulin per kilogram body weight. If insulin resistance is suspected (Cushing’s syndrome, acromegaly) the dose is 0.3 units per kg; if there is suspicion of adrenal hypofunction the dose is reduced to 0.1 units per kg.

Blood for glucose, cortisol and growth hormone is taken at 0, 30, 60, 90 and 120 minutes. Some workers prefer to check the glucose at 15 minutes. Glucose is collected into the yellow topped fluoride tube; while the brown topped 7.5 mL Sarstedt tube is sufficient for growth hormone and cortisol. Bedside monitoring of the blood glucose is very useful.

Signs and symptoms of hypoglycaemia occur ca. 20 – 30 minutes post injection (sweating, tachycardia, neuroglycopaenia). If these do not occur and the blood glucose has not fallen, a second equivalent dose of insulin is given.  
If symptoms are severe and/or prolonged, hypoglycaemia is reversed with intravenous glucose 25 - 50 mL 50%. This does not invalidate the response of growth hormone and cortisol, and sampling should continue. At the end of the test the cannula is removed and the patient is given a meal, and observed for at least one hour before leaving the ward.

#### Interpretation

Blood glucose should fall below 2.2 mmol/L accompanied by symptoms of hypoglycaemia.  
Cortisol should rise by 200 nmol/L to greater than 550 nmol/L.   
Growth hormone should rise to   >20 mU/L.  
In Cushing’s syndrome, the cortisol does not increase, and this has been used to differentiate these cases from depressed patients with raised cortisol levels.

## Thyrotrophin Releasing Hormone Test

This is performed less frequently nowadays following the introduction of highly sensitive TSH assays. However, the test still has a place in certain situations, to demonstrate the capacity of the pituitary to respond.  No specific patient preparation is required.  TRH may be combined with the ITT/LHRH tests, but recent opinion has cast doubt on the usefulness of this pituitary test (See Pavord et al., Clinical Endocrinology (1992) **36**:135).

#### Procedure

A cannula is placed in a forearm vein, and at time 0 a sample removed for fT4 and TSH assay into a brown capped Sarstedt tube.   200 mg TRH is given as in IV injection SLOWLY (over at least 1 minute, preferably more).  Rapid bolus administration is associated with unpleasant symptoms (nausea, desire to micturate, syncope).  Further blood samples for TSH are taken at 20 and 60 minutes post TRH.

#### Interpretation

Normally TSH rises by >2 mU/L to >5 mU/L, the 20 minute level being higher than the 60 minute.  Some workers simply sample at 0 and 20 minutes.  A reduced or flat response is seen in many situations, typically thyrotoxicosis but also acromegaly, hypopituitarism, patients taking thyroxine, multinodular goitres, the euthyroid sick syndrome and other conditions.  The chief value of the test is that a normal response excludes hyperthyroidism, but similar information is given by a single normal TSH level using the sensitive assays.  Often in hypothalamic diseases the 60 minute level is greater than the 20 minute, but this is by no means specific.

## Gonadotrophin Releasing Hormone Test

As with TRH, this test is less useful than previously thought, but is still performed in certain patients to assess gonadotrophin reserve.  It may be combined with the ITT and TRH tests.

#### Procedure

A cannula is placed in a forearm vein and at time 0 a blood sample removed into a brown capped Sarstedt tube for LH and FSH assay.  100 mg of gonadotrophin releasing hormone (GnRH, LH/FSH-RH) is injected as a bolus and further samples for LH and FSH taken at 20 and 60 minutes.

#### Interpretation

Normally LH and FSH rise at 20 minutes, but there is little agreement on the precise definition of normality. In constitutional delay of puberty the LH peak exceeds that of FSH.  In polycystic ovarian syndrome the LH peak is greatly in excess of the FSH peak.  
In suspected hypogonadism due to pituitary disease the test gives little more information than basal gonadotrophin plus testosterone/oestradiol levels.

**Adrenal**

The hypothalamic/pituitary/adrenal axis may be assessed by the ITT, and this test remains the “gold standard”.  However, adequate information regarding ACTH/adrenal reserve may be obtained using the less stressful Synacthen tests.  In cases of virilisation/hirsutism, measurements of testosterone, SHBG, LH/FSH, 17OH-progesterone and DHEAS may be required.

## Posterior Pituitary

#### Water Deprivation Test

This is used to assess vasopressin reserve. The test is potentially dangerous in diabetes insipidus, and close supervision is required throughout. Patients with hypopituitarism should be adequately treated with glucocorticoids and thyroxine. Ideally the test is performed in a side room where no water is available for surreptitious ingestion.  
Inform the laboratory that the test will be performed well in advance of the date.

#### Procedure

Fluids ad libitum are allowed until the morning of the test. A light breakfast is given at 0800 hours (no tea, coffee), and smoking is forbidden. The patient is weighed accurately and a basal blood sample for serum osmolality is taken into a brown capped tube. A specimen of urine is also required for osmolality, taken into a container with NO preservative (boric acid invalidates the osmolality measurements). If the basal plasma osmolality exceeds 300 mOsm/kg in the presence of a dilute urine the diagnosis of diabetes insipidus is made and ADH is given (vide infra).  
No fluids are allowed, and samples for plasma and urine osmolality are obtained at hourly intervals. The urine volume is measured and charted, as is the patient’s weight. If concentration of the urine has not occurred after 8 hours, 20 microgram Desmospray is given intranasally, and further urine samples collected hourly thereafter for 4 hours. Fluids are allowed after the Desmospray has been administered.  
The test should be terminated if the serum osmolality rises above 300 mOsm/kg, and/or the patient loses > 3% of body weight. Desmopressin is given as above, with free fluids.

#### Interpretation

May be difficult, particularly in patients with primary polydipsia. Normally the serum osmolality remains within the reference range (280-295 mOsm/kg), and the urine: plasma osmolality ratio rises to >2.0, at which point the test is terminated. Partial defects of ADH secretion may give equivocal results.   
Suspicion that the patient is taking fluids surreptitiously is passage of large quantities of dilute urine without weight loss. An accurate balance capable of reading to 50 g is essential.

**Pancreas**

**Oral Glucose Tolerance Test**

**Procedure**

The patient fasts from midnight. At time 0 a cannula is placed in a suitable forearm vein, 75 g glucose is given orally, either in orange juice or as 113 mL “Polycal”. The amount of glucose given is adjusted for children to 1.75 g per kilogram body weight up to the maximum of 75 g. The formal OGTT involves sampling at 30 minute intervals for 2 hours, but if the investigation is being performed to establish/refute a diagnosis of diabetes mellitus, a 0 and a 120 minute sample are sufficient. The samples are taken into fluoride tubes (Sarstedt yellow cap).

**Interpretation**

Glucose in venous plasma, mmol/L:

|  |  |  |
| --- | --- | --- |
| **DIAGNOSIS** | **FASTING** | **2 HR** |
| Normal | <6.1 | <7.8 |  |
| Impaired GT | <7.0 | 7.8 to   <11.1 |  |
| Diabetic | ≥7.0 | ≥11.1 |  |

**Acromegaly**

In active acromegaly growth hormone levels are not suppressed by a glucose load. The “growth hormone suppression test” is simply the 75 g OGTT with samples taken for growth hormone (Sarstedt brown capped tube) at 30 minute intervals. In normal subjects GH levels fall to <2 mU/L, while in acromegaly there is no suppression or even a paradoxical rise.  
Measurement of IGF-1 (a single random blood sample, brown capped tube), may give the same information as the OGTT, and is obviously more convenient.

**Prolonged (5 Hr) Glucose Tolerance Test**

RARELY OF USE IN ANY CLINICAL SITUATION and probably responsible for misdiagnosis of “reactive hypoglycaemia” in many patients. Samples are taken as for the 75 g OGTT and at 3, 4, and 5 hours. In many normal individuals the blood glucose level falls to 2.5 mmol/L, without symptoms. The test is NOT recommended, and is inappropriate for patients in whom a diagnosis of insulinoma is being considered. In the latter, fasting with blood sampling for glucose, insulin and C-peptide if and when symptoms occur is the preferred investigation.

#### Procedure

The following is a summary of the procedure to be followed.  Full details are provided within the test kit used.  These details must be strictly adhered to if a meaningful interpretation of the test result is to be made.   
The patient eats and drinks normally on the evening prior to the test, but no vitamin supplements may be taken until the test is complete.

DAY ONE

06.30 Pass urine and discard. Drink 500 mL of liquid.

07.00 Eat a test meal of 50 g bread with 220 g of butter. Swallow   
 the blue capsules during the meal and drink a cup of liquid.

**FROM THIS POINT, ALL URINE MUST BE COLLECTED INTO A LABELLED BOTTLE. NO PRESERVATIVE IS NECESSARY**

10.00 Drink on litre of liquid between 1000 am and noon.

12.00 Eat and drink as required.

17.00 The bladder is emptied as completely as possible and the urine is added to the collection. The procedure for day one is now complete.

DAY TWO

The procedure is followed exactly as for day ONE excepting that the red capsules are taken with the meal. Both urines are sent to the laboratory.

#### Interpretation

The results are expressed as the **T/K ratio.**  
Ratio less than 20 indicates low pancreatic exocrine function.  
Ratio 20-30 is equivocal.  
Ratio greater than 30 indicates normal pancreatic exocrine function.

## Pentagastrin Acid Output Studies

This test is occasionally employed to exclude a diagnosis of achlorhydria in patients with raised gastrin levels and peptic ulceration.

#### Procedure

The patient fasts for twelve hours without food or drink.   
A nasogastric tube is passed into the stomach, preferably confirmed radiologically.  The stomach contents are emptied with a hand syringe and labelled fasting contents.   
The next two fifteen minute specimens are collected to give basal secretion.   
Pentagastrin 6 micrograms per kilogram body weight is given subcutaneously.   
Subsequently four accurately timed fifteen minute specimens are collected.   
It is helpful to blow air down the nasogastric tube every five minutes or so to prevent blockage. The samples then go straight to the laboratory.

## Pentagastrin Stimulation (Calcitonin)

This test is performed to screen for medullary carcinoma of the thyroid (MCT).  It should NOT be performed in pregnancy.

#### Procedure

After overnight fasting, the patient is weighed and a cannula placed in a forearm vein.  At time 0 a blood sample for calcitonin is taken into a heparinised tube (Sarstedt 7.5 mL orange cap) and pentagastrin (“Peptavlon”) 0.5 micrograms/kg body weight injected intravenously over 10 seconds.  Further samples for calcitonin assay are obtained at 1 and 5 minutes post injection. The samples are placed on ice and taken to the laboratory immediately for separation.  
Side effects include sensations of warmth and/or burning, flushing, nausea and abdominal discomfort, but are transient and disappear after a few minutes.

#### Interpretation

In normal subjects peak calcitonin levels are <0.21 mg/L in males, <0.11 mg/L for females.  Raised levels are highly suggestive of C-cell hyperplasia/medullary carcinoma of the thyroid.  
In patients with a strong family history of MCT the test should be performed at regular intervals, yearly from the age of 5 years to teens, 2 yearly until late twenties, and 3-5 yearly until age 40.  
Recent developments suggest that a specific DNA test for the mutation causing MCT may be more sensitive than the pentagastrin stimulation test, particularly in cases of multiple endocrine neoplasia type II. 10 mL blood in EDTA (Sarstedt red cap) are required with an appropriate letter giving all relevant details and sent first class/van delivery to:  
  
Molecular Genetics  
East Anglian Regional Genetics Laboratory  
Addenbrookes Hospital  
Hills Road  
CAMBRIDGE  
CB2 2QQ  
  
This will be arranged through the laboratory.

## Testis

#### Human Chorionic Gonadotrophin (HCG) Test

This test assesses the presence of functional testicular tissue.

#### Procedure

Blood for testosterone assay (Sarstedt 7.5 mL brown capped tube) is taken at time 0 and 2000 units HCG given by intramuscular injection on day 0 and day 2. Further samples for testosterone are obtained on days 2 and 4.

#### Interpretation

In normal subjects the serum testosterone concentration rises above the reference interval. Failure to rise indicates the absence of functional testicular tissue.  If the testes are not in the scrotum, but a positive response to HCG is seen, the testes may be intra-abdominal.  
In secondary hypogonadism due to pituitary disease, a low basal testosterone level will increase threefold after HCG.

## Ovary

#### CLOMIPHENE TEST

This test is sometimes helpful in distinguishing gonadotrophin deficiency from weight related hypogonadism and constitutional delay of puberty.

#### Procedure

Clomiphene 3 mg/kg body weight is given in three divided doses daily for 7 days (maximum dose 200 mg daily).  Blood samples for serum LH and FSH (7.5 mL Sarstedt brown capped tube) are obtained on day 0, 4, 7 and 10.

#### Interpretation

In normal subjects both LH and FSH rise above the reference interval, or the basal level is doubled.  Some patients with anorexia nervosa show no response until weight has been gained. Pre-pubertal children do not respond to clomiphene.

## Urine Acidification Test

Used to establish/refute a diagnosis of renal tubular acidosis.  The laboratory should be informed well in advance of the test.

#### Procedure

The patient is weighed and may have a normal breakfast and meals thereafter.  At 0800 hours the bladder is emptied and hourly urine collections are begun and put into plain bottles with no preservative.  
At 1000 hours ammonium chloride 100 mg/kg body weight is given orally over a period of 1 hour.  This is most conveniently taken in the form of capsules supplied by Pharmacy to avoid gastric irritation and nausea.  Blood for electrolytes and bicarbonate (specified) is taken at 1000, 1400 and 1800 hours (Sarstedt brown capped tube) and hourly urines for pH measurement throughout the test.

#### Interpretation

In normal subjects the urine pH (determined by a pH meter, NOT using the sticks) should fall to below 5.3 at some point during the test, which may then be terminated.  The test should not be performed if the basal serum bicarbonate level is less than 19 mmol/L. A fall in plasma bicarbonate during the test confirms that the NH4Cl has been ingested and absorbed.

**[PRESERVATIVES FOR URINE COLLECTIONS](http://intranetapps/pathwebfiles/BIO_PRESERVATIVES_HEAD.html" \t "ContentPage)**

**Urine Specimens Collection and Preservation**

Note: Tests highlighted in ***Italics*** are referred to other laboratories

|  |  |  |  |
| --- | --- | --- | --- |
| TEST/TEST CODE | COLLECTION | PRESERVATIVE | NOTES |
| *Amino acids (AAU)* | *Random* | *Nil* | *Freeze until referral* |
| Bence - Jones protein (TPU/PEU) | Random | Nil | Acidified collection NOT acceptable |
| Calcium (CAU/CAD) | Random or24 hr | Hydrochloric acid | Suitable for preservation on receipt (pH must be <3) |
| *Catecholamines (CATD code is inactive) 🡪 use METD code* | *24 hr* |  | *Change request to Metanephrines and treat as such* |
| *Citrate (CITD)* | *24 hr* | *Hydrochloric acid* | *Unsuitable for preservation on receipt* |
| *Copper (CUD)* | *24 hr* | *Nil* | *Acidified collection NOT acceptable* |
| *Cortisol (CORD)* | *24 hr* | *Nil preferred* | *Acidified collections also accepted* |
| Creatinine and Creatinine clearance (CRU/CRD/CRC) | Random or 24 hr | Nil | Acidified collection acceptable |
| *Qualitative Cystine* *(See AAU)* | *Random* | *Nil* | *Write Amino acid/AAU on the form and treat as such* *(See above for information)* |
| *Quantitative Cystine (CYSQ)* | *24 hr* | *Hydrochloric acid (pH <3.0)* | *Unsuitable for preservation on receipt. 1 drop of 5M HCL may be added to aliquot if pH of acidified collection on receipt is >3* |
| *Drugs of abuse (DOAU)* | *Random* | *Nil* | *NOT TO BE ALIQUOTTED! Do not open the original container.* ***Freeze until referred.*** |
| Electrolytes (EU/ED) | Random or 24 hr | Nil | Aliquot and send to the priority bench |
| TEST | COLLECTION | PRESERVATIVE | NOTES |
| *Glycosaminoglycans**(GAGU)* | *Random* | *Nil* | *Freeze until referred* |
| *5HIAA (HIAD)* | *24 hr* | *Nil**(HCL preservative also acceptable)* |  |
| Magnesium (MGU/MGD) | Random or 24 hr | Hydrochloric acid | Suitable for preservation on receipt (pH must be ≤1)Not routinely available.Discuss with Roche Team / Consultant |
| *Mercury (HGU/HGD)* | *Random or 24 hr* | *Nil* | *Acidified collection NOT acceptable* |
| *Methylmalonate creatinine Ratio (MMAC)* | *Random* | *Nil* | *Freeze until referred* |
| *Metanephrines (METD)* | *24 hr* | *Nil**(HCL preservative also acceptable)* |  |
| Microalbumin (MAU) | Random | Nil | Acidified collection NOT acceptable |
| *Myoglobin (MYOU)* |  |  | *No Longer Available* |
| *N-Telopeptide X-links (NTXU)* | *Random* | *Nil* | *No need to aliquot. Send in original container.* |
| *Organic acids (ORGU)* | *Random* | *Nil* | *Freeze until referred* |
| Osmolality (OSMU) | Random | Nil | Aliquot and Send to Priority bench |
| ***Oxalate (OXD)*** | ***24 hr*** | ***Hydrochloric acid*** | ***Unsuitable for preservation on receipt*** |
| Phosphate (PO4U/PO4D) | Random or 24 hr | Hydrochloric acid | Suitable for preservation on receipt (pH must be <3) |
| ***Porphyrin – screen (POR)*** | ***Random*** | ***Nil*** | ***Should be accompanied by an EDTA sample and a faeces sample and protect all samples from light*** |
| Protein (TPU/TPD) | Random or 24 hr | Nil | Acidified collection NOT acceptable |
| Protein Electrophoresis/ Paraprotein (PEU/PPU) | Random or 24 hr | Nil | Acidified collection NOT acceptable |
| TEST | COLLECTION | PRESERVATIVE | NOTES |
| ***Steroid profile (STEU/STED)*** | ***Random or 24 hr*** | ***Nil*** | ***Acidified collection NOT acceptable*** |
| ***Sulphonylurea (SPU)*** | ***Random*** | ***Nil*** | ***Acidified collection NOT acceptable*** |
| Total protein/Creatinine ratio (TPC) | Random | Nil | Acidified collection NOT acceptable |
| Urate/Uric acid (UAU/UAD) | Random or 24 hr | Nil | Acidified collection NOT acceptable |
| Urea (URU/URD) | Random or 24 hr | Nil | Acidified collection NOT acceptable |