INFORMATION FOR USERS OF THE BIOCHEMICAL GENETICS SERVICE

Web: http://www.nhsggc.org.uk/medicalgenetics
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Biochemical Genetics

Location and Organisation

The Biochemical Genetics Department is located within the Duncan Guthrie Institute of Medical Genetics at Yorkhill. It is one of the three laboratory Departments within Medical Genetics which together with the Clinical Genetics Service comprise the West of Scotland Regional Genetics Service:

Clinical Lead
Director of West of Scotland Regional Genetics Service

Professor J. M. Connor (J.M.Connor@clinmed.gla.ac.uk)
Duncan Guthrie Institute of Medical Genetics
Tel: 0141 201 0363
FAX: 0141 357 4277

Laboratory Departments- Duncan Guthrie Institute of Medical Genetics

Laboratories Reception and general enquiries: 0141 201 0365

Biochemical Genetics

Head of Department: Dr David A Aitken (david.aitken@ggc.scot.nhs.uk)
Consultant Clinical Scientist
Honorary Senior Lecturer
Tel: 0141 201 0370

Cytogenetics:

Head of Department: Mr Gordon Lowther (gordon.lowther@ggc.scot.nhs.uk)
Consultant Clinical Scientist
Honorary Senior Lecturer
Tel: 0141 201 0669

Molecular Genetics:

Head of Department: Ms Su Stenhouse (su.stenhouse@ggc.scot.nhs.uk)
Consultant Clinical Scientist
Honorary Senior Lecturer
Tel: 0141 201 0360
Clinical Genetics Service

The Medical Genetics Laboratories work in close liaison with the Clinical Genetics Service which is located close by in the Ferguson-Smith Centre for Clinical Genetics at Yorkhill:

Ferguson-Smith Centre for Clinical Genetics - Reception  Tel 0141 201 0808

Consultant Clinical Geneticists (Ferguson-Smith Centre for Clinical Genetics):

Dr John Tolmie - Honorary Clinical Senior Lecturer  
(John.Tolmie@ggc.scot.nhs.uk)  Tel 0141 201 0379

Dr Margo Whiteford - Honorary Clinical Senior Lecturer  
(Margo.Whiteford@ggc.scot.nhs.uk)  Tel 0141 201 0357

Dr Rosemarie Davidson - Honorary Clinical Senior Lecturer  
(Rosemarie.Davidson@ggc.scot.nhs.uk)  Tel 0141 201 0701

Dr Vicki Murday - Honorary Clinical Senior Lecturer  
(Vicki.Murday@ggc.scot.nhs.uk)  Tel 0141 201 0451

Honorary Consultant Clinical Geneticists:

Professor J M Connor - Professor of Medical Genetics  
Director west of Scotland Regional Genetics Service  
(J.M.Connor@clinmed.gla.ac.uk)  Tel 0141 201 0363

Dr Douglas Wilcox – Clinical Senior Lecturer  
Director Scottish Muscle Centre  
(D.E.Wilcox@clinmed.gla.ac.uk)  Tel 0141 201 0483

Dr Edward Tobias – Clinical Senior Lecturer  
(e.tobias@clinmed.gla.ac.uk)  Tel 0141 201 0375
Biochemical Genetics Service

The Biochemical Genetics Department is sub-divided into three sections:

1. Prenatal Screening,
2. Newborn Screening
3. Inborn Errors of Metabolism.

Head of Department: Dr David A Aitken BSc, MIBiol, PhD, FRCPath
Consultant Clinical Scientist
Honorary Senior Lecturer: david.aitken@ggc.scot.nhs.uk
Tel: 0141 201 0370

Enquiries regarding testing, results and advice should be directed to the head of the appropriate section.

1. Prenatal Screening Service For Neural Tube Defects and Down's Syndrome.

Head of Section
Deputy Head of Department: Dr Jennifer Crossley BSc, PhD, FRCPath
Consultant Clinical Scientist
Honorary Lecturer (jenny.crossley@ggc.scot.nhs.uk)
Tel: 0141 201 0373

This service is provided for 60% of the Scottish pregnant population in seven Health Boards in the West of Scotland: Ayrshire and Arran, Dumfries and Galloway, Forth valley, Greater Glasgow and Clyde, Lanarkshire, Highland and the Western Isles. Samples are also accepted from further afield. In the west of Scotland, over 20,000 women choose to have the prenatal screening test for neural tube defects and Down's syndrome each year.

Screening for neural tube defects started in 1975, and screening for Down's syndrome in 1991, making this one of the largest prenatal screening programmes in Europe.

Test principle:

Second Trimester Screening: The concentrations of two maternal serum markers, alpha-fetoprotein (AFP) and human chorionic gonadotrophin (hCG) are measured in maternal blood at 15-20 weeks of gestation.

In pregnancies with open neural tube defects (spina bifida, anencephaly) AFP levels are generally elevated. Around 3% of women will have AFP levels $\geq 2.0$ multiples of the normal median level and should be offered detailed ultrasound scanning.

In Down's syndrome pregnancies the level of AFP is generally reduced and the level of hCG increased. A probability (risk) that each analyte level is associated with a Down's syndrome pregnancy is derived and combined with the prior probability of an affected pregnancy based on maternal age. Women with a combined risk of
≥1 in 250 are considered to be at increased risk and are offered diagnostic amniocentesis for fetal karyotyping. A separate risk protocol is used to provide information about the risks of trisomy 18. Please use the white request form for combined second trimester screening.

**First Trimester Screening:** A first trimester combined ultrasound and biochemical protocol for chromosome abnormalities (CUB screening) is provided. This uses a combination of ultrasound nuchal translucency measurements at 11-13 weeks of gestation and the serum biochemical markers, free beta hCG and Pregnancy Associated Plasma Protein A, (PAPP-A) in maternal blood taken at 9 -13 weeks of gestation. For further information on the availability of CUB screening, request forms and patient information leaflets, contact the Prenatal Screening Office (0141 201 0371/0372). Please use the green request form for CUB screening.

**Second Trimester Screening for Spina Bifida:** This is available for women who have had CUB screening and wish a screening test for spina bifida or for women who wish to have spina bifida screening only. Please use the yellow request form for spina bifida only screening.

**Sample requirements:** For second trimester screening for Down's syndrome and Neural Tube Defects, a clotted venous blood sample (5-10 mls) obtained at 15-21 weeks of gestation is required. Note: Blood should be collected in plain (serum) tubes as some anticoagulants e.g. EDTA interfere with the biochemical test.

For first trimester CUB screening for Down's syndrome, clotted venous blood samples taken at 9 -13 weeks are required. Note that the AFP test for neural tube defects is available only from 15-20 weeks of gestation. NT measurement should be carried out at between 11-13 weeks.

All sample tubes must be labelled and dated, and sealed in the bag attached to the appropriate completed request form bearing the CHI number. This information is essential for interpretation of test results. Request forms can be obtained from the prenatal screening office (0141 201 0371/0372). Patient information leaflets are available from the local Health Board Health Promotion Department. Leaflets in languages other than English can be obtained via the internet: [http://www.healthscotland.com/](http://www.healthscotland.com/)

**Sending samples to the Laboratory:** Samples should be sent to the laboratory with minimum delay after being taken. Samples received more than 7 days (5 days for first trimester CUB screening) after the sample date are no longer suitable for analysis and a repeat sample will be requested. Sample tubes and request forms must be kept separate, with one sample only per sample bag. Samples must be contained in rigid packaging for transportation. High risk samples must be clearly labelled as such. Samples forwarded by post must be packaged to conform with current postal regulations and should be sent first class/guaranteed next day delivery.

**Results:** Over 95% of results are available within 3 working days of sample receipt. Results which require diagnostic follow-up testing for neural tube defects and Down's syndrome are FAXed or telephoned as soon as the result is available. Hard copies of reports are generated for all samples and sent to the referring centre by mail.
For further information on prenatal screening see the Biochemical Genetics pages at:  
http://www.nhsggc.org.uk/medicalgenetics

All Enquiries to Prenatal Screening Office: 0141 201 0371/0372  
Laboratory Hours: 09.00 - 17.00, Monday - Friday.
2. National Newborn Screening Service

Head of section: Mrs Joan Mackenzie FIBMS, MSc
Head Biomedical Scientist
(joan.MacKenzie@ggc.scot.nhs.uk)
Tel. 0141 201 0465

This service, originally based at Stobhill Hospital, relocated to the Institute of Medical Genetics at Yorkhill in February 2001 as a component of the Biochemical Genetics Service.

Dried blood spots (Guthrie cards) are received from new-born babies from all over Scotland (about 60,000 / year) and tested for three conditions: Phenylketonuria (PKU), Congenital Hypothyroidism (CHT) and Cystic Fibrosis (CF).

The aim of the service is to diagnose these disorders as early as possible to allow affected infants to be placed on the appropriate treatment.

Test Principle:
Dried blood spots from neonates are tested by mass spectrometry for high levels of Phenylalanine (as an indicator of PKU) and for high levels of Thyroid Stimulating Hormone (TSH) by Delfia immunoassay (as an indicator of Hypothyroidism). Screening for CF is based on measurement of immunoreactive trypsinogen (IRT) by Delfia immunoassay. DNA, extracted from blood spots from neonates with elevated concentrations of IRT, is analysed for 29 different mutations.

Sample requirements: The routine dried blood spot specimen (Guthrie specimen) should be collected when the baby is at least 96 hours old and before the 8th day of life. Instructions for collecting the blood are printed on the reverse of the blood spot card. Briefly, the heel is swabbed, warmed and punctured with a suitable bloodletting device (e.g. Tenderfoot) and the blood droplets applied to the filter paper portion of the blood collection card. The card should be air-dried for approximately 3 hours, placed in a glassine envelope and sent by first class post using the pre-printed pre-paid envelopes provided by the laboratory. Cards should be dispatched immediately. All details requested on the card should be filled in at the time of collection. Blood spot cards can be obtained on request from the laboratory.

Patient information leaflets are available from the local Health Board Health Promotion Department. An electronic version, including leaflets in languages other than English, can be obtained via the internet: http://www.healthscotland.com/

Results: Negative results and requests for repeat specimens are sent by post to the department of Child Health for each area. These are generally issued within 48 hours. Positive results, requiring the child to be further investigated, are telephoned to the appropriate Paediatrician/Child Health Department. Written confirmation is issued within 24 hours of the telephone call.

More detailed information on newborn screening including action limits and reporting and recall procedures can be found at:
http://www.nhsggc.org.uk/medicalgenetics

All enquiries to Newborn Screening Office: 0141 201 0870.
Laboratory Hours: 08.30 - 16.30, Monday – Friday
3. Diagnosis and Prenatal Diagnosis of Inborn Errors of Metabolism

Head of Section: Dr Gordon Graham BSc PhD
Principal Clinical Scientist
Honorary Senior Lecturer
(gordon.graham@ggc.scot.nhs.uk)
Tel: 0141 201 0375

The Department has extensive experience in diagnosis and prenatal diagnosis of inborn errors of purine metabolism. Diagnostic and carrier status testing for Steroid Sulphatase deficiency is also offered as a service. Measurements include both STS enzyme activity and gene deletion studies for X-linked Ichthyosis.

**Reporting Times:** The target turnaround time for analyses (i.e. the interval between receipt of request and the reporting of results to the requesting health provider) is within 1 week. Some more complex investigations involving further analyses may involve liaison with, or referral to, other Departments and may cause the reporting times to be longer. These will be discussed with the user on a case by case basis.

**Request Forms:**
Samples for investigation of these Inborn Errors are often sent with request forms designed for other disciplines. We would recommend that our own request form be used where possible.
See Appendix 1 on page 14 of this handbook for the request form which may be printed as necessary.

**A) Purine Metabolic Disorders**

Testing is offered for:

- Adenosine Deaminase deficiency (ADA). Autosomal recessive.
- Nucleoside phosphorylase deficiency (PNP). Autosomal recessive.

**Test principle:** Specific enzyme analyses are carried out on red cells or cultured fibroblasts using radiolabelled assays (HGPRT and ADA) or kinetic spectrophotometric assay (PNP). Complete deficiency or low levels of residual enzyme activity are indicative of an affected individual. Carrier detection is possible by identification of intermediate levels of enzyme activity in cases of ADA and PNP carriers. Female carriers of the Lesch Nyhan syndrome have normal HGPRT activity and cannot be identified by enzyme analysis. In some families with Lesch Nyhan syndrome where the mutation is known, diagnosis and carrier detection may be possible by DNA analysis.
**Sample requirements:** For routine diagnoses, 2-5 ml heparinised blood is required. In cases of neonates or small children, diagnoses can be attempted on 1.0 ml. Samples should be transported to the laboratory at ambient temperature (Do not freeze) to arrive within 24 hours. For DNA studies 2-5 ml of blood in EDTA is required. For cultured fibroblasts, one small (25cm ) flask approaching confluency should be filled completely with culture medium and sent to the laboratory at ambient temperature to arrive within 24 hours.

For prenatal diagnoses, chorionic villus samples (CVS) or cultured amniotic fluid cells can be used. CVS should be sent in transport medium at ambient temperature (not frozen). Enzyme analyses are carried out direct on uncultured CVS and, where samples are of adequate size, on cultured cells. Fetal blood (heparinised) may also be used.

Amniotic fluid samples (either whole liquor or growing cells) should be sent to the laboratory at ambient temperature (Do Not Freeze) to arrive within 24 hours.

**Please contact the laboratory before sending samples**

All Enquiries to Biochemical Genetics Laboratory : 0141 201 0375
Laboratory Hours: 09.00 - 17.00, Monday - Friday.

Reference Ranges

Reference Ranges for the purine metabolic disorders are shown in Table 1.
## Table 1: Reference Ranges for Purine Metabolic Disorders

<table>
<thead>
<tr>
<th></th>
<th>HGPRT nmols/hr/mg Hb or nmols/hr/mg Protein</th>
<th>APRT nmols/hr/mg Hb or nmols/hr/mg Protein</th>
<th>HGPRT/APRT Ratio</th>
<th>ADA nmols/hr/mg Hb</th>
<th>PNP μmols/hr/mg Hb</th>
</tr>
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<tbody>
<tr>
<td><strong>Adult Red Cells</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean</td>
<td>94.0</td>
<td>14.7</td>
<td>6.4</td>
<td>56.8</td>
<td>3101</td>
</tr>
<tr>
<td>Range (±2sd)</td>
<td>49.6 – 162.9</td>
<td>6.3 – 48.5</td>
<td>1.9 – 15.5</td>
<td>25.4 – 112.5</td>
<td>1300 – 7836</td>
</tr>
<tr>
<td><strong>Uncultured CVS</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean</td>
<td>63.8</td>
<td>49.5</td>
<td>1.47</td>
<td>497.9</td>
<td></td>
</tr>
<tr>
<td>Range (±2sd)</td>
<td>9.7 – 113.5</td>
<td>3.0 – 121.9</td>
<td>0.32 – 3.90</td>
<td>186 – 480.5</td>
<td></td>
</tr>
<tr>
<td><strong>Cultured CVS</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean</td>
<td>50.2</td>
<td>63.5</td>
<td>0.77</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Range (±2sd)</td>
<td>3.4 – 174.6</td>
<td>4.1 – 192.6</td>
<td>0.13 – 2.6</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Amniotic Fluid Cells</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean</td>
<td>178.9</td>
<td>160.2</td>
<td>1.47</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Range (±2sd)</td>
<td>44.2 – 555.6</td>
<td>14.7 – 308.0</td>
<td>0.12 – 3.57</td>
<td></td>
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<tr>
<td><strong>Fibroblasts</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Mean</td>
<td>56.2</td>
<td>74.7</td>
<td>1.09</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Range (±2sd)</td>
<td>3.8 – 138.0</td>
<td>5.3 – 196</td>
<td>0.12 – 3.57</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Fetal Red Cells</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean</td>
<td>73.6</td>
<td>29.0</td>
<td>2.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Range (±2sd)</td>
<td>51.4 – 105.4</td>
<td>14.5 – 57.8</td>
<td>1.3 – 4.7</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Cord Red Cells</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean</td>
<td>95.2</td>
<td>29.4</td>
<td>3.9</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Range (±2sd)</td>
<td>65.8 – 153.2</td>
<td>10.4 – 53.9</td>
<td>1.9 – 8.6</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
B) X-linked Ichthyosis

1. **Enzyme Analysis for Steroid Sulphatase (STS) deficiency.**

   **Test principle:** STS activity is measured on white cells or cultured fibroblasts using a radiolabelled assay with $^3$H Dehydroepiandrosterone sulphate as substrate. Where possible, enzyme studies are supplemented with gene deletion analysis.

   **Sample requirements:** White cells are extracted from 10 mls EDTA blood. For small children, the analyses can be attempted on 5.0 mls. The best yield of WBCs is obtained from freshly drawn blood and samples. The use of EDTA tubes enables both enzyme and DNA studies to be performed on a single sample.

   **Transportation requirements**
   Samples should be transported to the laboratory in a sealed, leak-proof container at ambient temperature with minimum delay. An overnight courier or first class post may be used, with avoidance of a Friday posting where possible.

   **Reference Ranges:**
   The Reference Ranges for white cell Steroid Sulphatase activity are as follow:

   - **Normal Males**
     Mean: 103.7 µmoles/hr/mg Protein
     Range (±2sd): 53.1 – 154.3

   - **Normal Females**
     Mean: 131.7 µmoles/hr/mg Protein
     Range (±2sd): 66.5 – 197

2. **Gene deletion analysis for Steroid Sulphatase (STS) deficiency.**

   **Test principle:**
   Presence or absence of the STS gene by DNA PCR analysis using oligonucleotide primers to amplify two regions of the gene. One is approximately 900bp upstream of the initiation site of STS and the other contains the termination site in exon 10. (Reference: Ballabio et al (1990) Screening for steroid sulphatase (STS) gene deletions by multiplex DNA amplification. *Hum. Genet.* May; 84(6); 571-573)

   **Sample requirements:** DNA analyses are carried out on 2-5mls of EDTA blood. Samples should be sent at ambient temperature by first class post or commercial courier service. If white cell enzyme analysis is also required, a single sample of 5-10mls for this and DNA studies will suffice.

   **Transportation requirements**
Samples should be transported to the laboratory in a sealed, leak-proof container at ambient temperature with minimum delay. An overnight courier or first class post may be used, with avoidance of a Friday posting where possible.

**Reference Ranges**

The analysis of STS by DNA PCR is a qualitative test requiring visualisation of a specific gel banding pattern and as such has no numerical reference range. The result for the non-affected individual shows 2 bands corresponding to each end of the gene, while the affected individual shows neither of these.

**Please contact the laboratory before sending samples**

All Enquiries to Biochemical Genetics Laboratory: 0141 201 0375
Laboratory Hours: 09.00 - 17.00, Monday - Friday.

**C) Other Tests**

**Prenatal Diagnosis of Neural Tube Defects by Amniotic Fluid Biochemistry.**

**Test principle:** Amniotic fluid AFP levels are raised at 15-21 weeks gestation in cases of open neural tube defect and certain other structural abnormalities of the fetus. AFP levels measured by immunoassay are assessed against the normal range and classified as elevated if the levels are ≥2.5 multiples of the normal median at 15 weeks of gestation, ≥3.0 MoM at 16-18 weeks and ≥3.5 MoM at 19-24 weeks. Fetal blood in amniotic fluid can lead to spuriously elevated AFP levels in unaffected pregnancies. Visibly bloodstained samples are analysed by electronic particle counting to determine type and quantity of blood and a corrected AFP value calculated if fetal blood is present.

All samples are analysed for the presence of neuro-specific acetylcholinesterase by polyacrylamide gel electrophoresis. A single band (non-specific cholinesterase) is found in unaffected pregnancies and a double band pattern (non-specific cholinesterase + specific acetylcholinesterase) in pregnancies with open neural tube defects.

**Sample requirements:** 5 ml amniotic fluid supernatant, plus 1.0 ml uncentrifuged liquor if visibly blood stained. Samples should be sent at ambient temperature by first class post.

All Enquiries to Biochemical Genetics Laboratory: 0141 201 0376
Laboratory Hours: 09.00 - 17.00, Monday - Friday.
### Sample requirements

**ADA, PNP, HGPRT deficiencies**

- **Enzyme studies**: 2-5 ml heparinised blood
- **DNA studies**: 2-5 ml blood in EDTA.
- **Prenatal diagnosis**: amniotic fluid cell cultures or chorionic villous samples.
  
  *** Please contact laboratory before sending samples***.

- Transport all samples at ambient temperature, guaranteed next day delivery

**X-linked ichthyosis (STS deficiency)**

- **Enzyme and DNA studies**: 2 -10 ml EDTA blood.

- Transport all samples at ambient temperature, guaranteed next day delivery