SW Thames Regional Genetics Laboratory

# Information for Users of the Cytogenetic Services

SW Thames Regional Genetics Service
At St. George's University Hospital NHS Foundation Trust

| | |
|---|---|
| Document No | MIA04.13 |
| Issue date | 06/10/15 |
| Review date | 06/10/16 |
| Author | Karen Marks |
| Authorised by | Will King |

## 1 Introduction

The Genetics Laboratory is situated in the Jenner Wing of St. George's Hospital and is one of the four London based laboratories providing a comprehensive range of cytogenetic and molecular genetic tests to patients in London, Bedfordshire, Essex, Hertfordshire, Kent, Surrey, Middlesex and Sussex.

The Postal Address is:
S.W.Thames Regional Genetics Laboratory
Lower Ground Floor, Jenner Wing
St. George's, University of London
Cranmer Terrace
London
SW17 0RE

Phone: 020 8725 5332
FAX: 020 8725 0570
Website: www.southwestthamesgenetics.nhs.uk
Email: swtrgl@stgeorges.nhs.uk

The Head of Laboratory is

**Rohan Taylor**
**Phone: 020 8725 1098**
**Email: rohan.taylor@stgeorges.nhs.uk**

Comprehensive cytogenetics services are offered for the investigation of constitutional chromosome abnormalities utilising whole genome screening techniques (karyotyping and Array CGH) and other specialised testing including culturing fibroblasts for biochemical and/or DNA studies.

The Genetics Laboratory is open Monday to Friday between the hours of 9.00 am and 5.00 pm. There will not normally be anyone in the department outside these hours or over Bank Holiday periods. Advice may be sought about any urgent samples arriving outside these hours by contacting the laboratory during its usual opening hours.

## 2 Request Forms

Requests for cytogenetic tests should be accompanied by the laboratory request form whenever possible. (The laboratory request form is available for download from our website) All request forms should be completely and clearly filled out including:

- Correct patient identification including full name, date of birth, address and patient's sex.
- Sample type with date and time of collection.
- NHS number, and where applicable local patient identifier (e.g. hospital number)
- Name and address of referring doctor.
- Patient's GP and the Practice Code
- Clear indication as to why a test has been requested.
- If a full karyotype/Array CGH and/or FISH test is required. (Please specify what specific FISH test if any).
- Consent for testing and possible storage of material
- Status: NHS or Private

***Failure to fill in the request form clearly and correctly may result in inappropriate tests and delays in reporting. Serious problems in patient information may result in the sample not being processed.***

## 3 Sample requirements

All samples should be sent to the Genetics laboratory as soon as possible after collection.

Prenatal samples should arrive in the department by 4.00 pm on the day of sampling whenever possible. First class post is acceptable but will add to the reporting time. Posting over a weekend should be avoided if at all possible.

Peripheral blood samples can usually be sent by first class post. Avoid posting over a weekend whenever possible as samples may deteriorate if delays occur in transit.

All samples must be clearly labelled. When sending samples, please make sure that the containers are tightly capped and well packed with a separate pocket for the request form. All samples must be packaged to comply with current legislation. The address for specimen reception is as printed above.

If you have any queries concerning samples for chromosome analysis please contact the laboratory on 020 8725 5332.

**NO SAMPLE FOR CHROMOSOME ANALYSIS SHOULD BE FROZEN OR PLACED IN ANY PRESERVATIVES OR FIXATIVE**

### 3.1 Venous Blood

For **Array CGH** 4-8ml in EDTA is required.

**For chromosome analysis (karyotyping or FISH**) 4ml is required (1ml will suffice for small babies) unclotted in a Lithium Heparin container (green top vacutainer or orange screw cap)

### 3.2 Fetal Blood

For **Array CGH** at least 1ml in EDTA is required.
For **chromosome analysis (karyotyping or FISH**) at least 1ml is required, unclotted, in a Lithium heparin container

***Please note that Microtainer brand 0.6ml Lithium heparin bottles do not give good results so please avoid using these where possible.***

### 3.3 Amniotic Fluid

10 –20 ml is required in a sterile universal container.

### 3.4 CVS Biopsies

10-15mg is required (more if the sample needs to be split for other tests) in CVS transport medium, (samples less than 5mg may not provide a sufficient amount of material depending on the tests required, the referring clinician will be informed if this is the case).
Transport medium will be supplied by the laboratory on request.

### 3.5 Solid Tissues

Samples should be sent in a sterile container in transport medium available on request from the laboratory. If medium is not available **sterile isotonic saline** may be used. Dry samples, if received the same day, are also acceptable.

### 3.6 Any other samples

Please phone the laboratory to discuss requirements.

## 4 Acceptance criteria for samples

Samples are subject to the acceptance criteria adhered to by this laboratory, and the sample will not be processed if it does not meet these criteria. If a clinician feels that there is a good clinical reason to send a sample which does not meet our acceptance criteria they should discuss this with the laboratory staff prior to sending the sample so that agreement can be reached.

**Private Patients**

Patients undergoing investigations as part of a private consultation fall outside the scope of the specification and are regarded as private referrals and test costs will be invoiced to the requesting clinician/patient/other as appropriate. If the referral falls outside the usual laboratory criteria for NHS samples please phone the laboratory to discuss requirements before sending the sample for processing.

### 4.1 Array CGH

Array CGH as offered as the **first line test** for paediatric and adult samples with the following referral reasons.

- Developmental delay, learning difficulties, speech/language delay (with or without autism).
- Dysmorphic facial features
- Multiple congenital abnormalities with or without pre/postnatal growth abnormalities.
- Apparently balanced chromosome rearrangements with an abnormal phenotype.

A blood sample in EDTA is required for this test (see sample requirements).

**Please note that for a postnatal Array CGH test the rear of the referral form should also be completed.**

Family follow up may be requested to clarify the significance of any finding where the association with the phenotype is unknown. Parental samples and any further patient sample required will be requested on the initial report and follow up will be done using a variety of techniques such as FISH (fluorescent in situ hybridization), parental arrays.or other appropriate molecular tests)

Please note that if Array CGH **and** karyotyping / FISH are requested, the karyotyping will not normally be done as any possible imbalance in the patient genome will be detected more accurately by array CGH. Karyotyping of the patient may be done at the laboratory's discretion to give additional information when this would be useful for the interpretation of the array result.

Array CGH is also offered as the first line test for prenatal samples where the nuchal translucency is equal to, or greater than, 3mm and in cases where there is an abnormal ultrasound scan.

**Please note that for a prenatal Array CGH test there is a specific form, obtainable from the Genetics laboratory.**

### 4.2 Indications for Prenatal Diagnosis

**Sample types**: amniotic fluid, chorionic villous, fetal blood.

#### 4.2.1 Referrals that will have routine rapid screening for common trisomies using QFPCR only

- High risk of aneuploidy from the result of a screening programme as defined by the UK National Steering Committee (NSC) current standards. **These samples will have QFPCR testing for trisomy 13, 18, and 21 only. Material will be kept short term to check any abnormal or equivocal QFPCR results with a full karyotype as necessary.**

- Pregnancies at a prior risk of a single gene disorder. **These samples will not be routinely karyotyped, but will be screened for trisomies 13, 18, and 21 using QFPCR <u>in addition</u> to testing for the single gene disorder in question. Cultures will be set up to provide further material for testing as required and used to check any abnormal or equivocal QFPCR results with a full karyotype as necessary.**

- **Samples obtained while undergoing other invasive procedures, e.g. transfusion for RhD, twin to twin transfusion syndrome.**

**Notes**

- *Risks based on Maternal Age alone should not be used.*
- *QFPCR testing will include screening for sex chromosome aneuploidy where:*
  - *The Nuchal Translucency is 4mm or greater*
  - *There is a risk of an X linked disorder*
  - *There is ultrasound detection of abnormalities such as cystic hygroma or cardiac abnormalities indicative of Turner syndrome*
  - *Ultrasound evidence of ambiguous genitalia*
  - *Knowledge of the sex may facilitate counselling and/or allow for targeted investigations*

### 4.2.2 Referrals that will have QFPCR and Array CGH / karyotyping (the most appropriate screening method will be used depending on the referral reason)

- Ultrasound detection of any major structural abnormality including nuchal translucency (NT) >3mm before 14 weeks gestation or a nuchal fold measuring 6mm or greater between 14 and 20 weeks gestation.
- Ultrasound detection of two or more minor markers of aneuploidy. Consistent with NSC guidelines, further testing (other than QFPCR) should not be offered on a single marker in women who have a low risk following aneuploidy screening.
- History of chromosome abnormality indicative of increased risk for future pregnancies. Chromosome abnormality may be present in:
  - ❖ the woman or her partner
  - ❖ a previous pregnancy (excluding non-viable aneuploidy)
  - ❖ If there is a family history, karyotyping of the woman or her partner should be undertaken first in order to establish whether prenatal diagnosis is indicated.

- Non-routine cases not fulfilling the above criteria after discussion and agreement between the referring clinician and a senior staff member from Cytogenetic Unit.

**NOTES**

***Maternal anxiety is not an indication for prenatal karyotyping unless considered by the referring clinician to be important for the management of the pregnancy.***

***All prenatal samples of amniotic fluid or CVS are routinely sent for QFPCR for rapid diagnosis of common trisomies 21, 13 and 18.***

***QFPCR testing is sent to another currently CPA accredited laboratory at Guy's hospital who issue their results directly to the referrer.***

Refs:
1) Ultrasound Screening Supplement - to Ultrasound Screening for Fetal Abnormalities RCOG 2000. (http://www.rcog.org.uk/womens-health/clinical-guidance/ultrasound-screening.
2) Normal variant screening in pregnancy NHS Screening Programmes – Fetal Anomaly – Programme Statement on: 'Normal Variant ('previously known as soft markers').
This statement contains a note that the information therein supersedes *'Ultrasound for Screening for Aneuploidy: Guidance for the professional'* (RCOG 2000).
3) QF-PCR Health Professional Information Leaflet '*Information for Health Care Professionals* – Guidelines for QF-PCR testing alone for women at increased risk for Down's syndrome' 2007.
UK National Screening Committee current guidance for Testing for Down's Syndrome in pregnancy

### 4.3 Indications for paediatric/adolescent karyotyping

**Sample type**: Mostly Peripheral blood but may be Solid Tissue (skin biopsies).

- Ambiguous genitalia/indeterminate gender.
- Delayed puberty, or inappropriate secondary sexual development.
- Family history of a chromosome abnormality
- Short stature, amenorrhoea in females.
- Microdeletion syndromes (where commercial FISH probes are available, although these will normally be screened using Array CGH).
- Cases not fitting within these categories after discussion and agreement between the referring clinician and senior laboratory staff.
- Confirmation of a diagnosis made prenatally

### 4.4 Indications for adult karyotyping

**Sample type:** Mostly Peripheral Blood but may be Solid Tissue (skin biopsies).

- Any of the paediatric/adolescent referral categories.
- Oligozoospermia or azoospermia in males.
- Premature ovarian failure.
- Family history of a known chromosome abnormality other than simple aneuploidy due to non-disjunction.
- Suspected family history of chromosome abnormality where the karyotype of the affected individual is not known.
- Sperm and egg donors for NHS funded patients.
- Couples undergoing assisted conception funded by the NHS.

**NOTE:** *Parental karyotyping for a family history of a chromosome abnormality where karyotyping has shown aneuploidy from non disjunction (typical primary trisomy) is not an appropriate referral request.*

### 4.5 Indications for post mortem perinatal, fetal or placental whole genome screening

For the majority of our post mortem, perinatal, fetal or placental screening referrals, we will perform QFPCR for the common aneuploidies encountered in this referral group (13, 15, 16, 18, 21, 22, X and Y) and then ArrayCGH will be performed on those with a normal aneuploidy result. In a few instances it may be more appropriate to karyotype the sample depending on the referral reason.

**Sample types**: Solid tissues such as: products of conception, fetal/neonatal tissue, placental tissue, or fetal/neonatal blood.

- Fetuses/stillbirths/neonatal deaths with congenital abnormality suggestive of a chromosomal anomaly or with neural tube defect or with IUGR.
- Follow up of prenatal ultrasound findings suggestive of a chromosome anomaly.
- Follow up confirmation of prenatal cytogenetic findings on post termination tissue.
- Unexplained stillbirth or neonatal death (≥ 24 weeks).
- Unexplained IUD/spontaneous abortion (≥ 16 weeks).
- Spontaneous abortion where the couple has a known chromosome rearrangement.
- Spontaneous abortion less than 16 weeks where the couple were undergoing assisted conception funded by the NHS.
- Products of conception / spontaneous early miscarriage (<16 weeks) from the **3rd or subsequent** miscarriage.
- Cases of perinatal death under investigation by the coroner.

### 4.6 Supplementary and/or specialised tests

Cytogenetic testing involves whole genome screening utilising Array CGH and karyotyping (G-banded chromosome analysis). However, in some circumstances more specialised techniques are required. The following list indicates the supplementary tests that will routinely be applied when necessary for full interpretation of results.

- Extra specialised conventional staining techniques (e.g. C-banding)
- Chromosome breakage analysis for diagnosis of chromosome instability disorders (These will be sent to another accredited laboratory for testing).
- Routine application of commercially available FISH probes including whole chromosome painting probes, centromeric probes, sub-telomeric probes and locus specific probes (for microdeletion syndromes).
- Rapid testing by interphase FISH for urgent neonatal referrals for sex ascertainment and to test for trisomy 13, 18 and 21**.**
- Mosaicism screening of skin samples from patients with a normal blood karyotype / Array CGH result.

**Please phone and ask about any tests which may have recently become available**.

## 5 Storage of clinical material following analyses

Storage will meet the minimum guidance in 'The retention and storage of pathological records and specimens (5th Edition, 2015). Guidance from The Royal College of Pathologists and the Institute of Biomedical Science'.
Storage of tissue will be in accordance with the requirements of The Human Tissue Act (2004).
There will routinely be storage of slides / digitised images. Storage of viable fibroblast cultures at -80$^0$C can be undertaken on request.
Individual requests for non routine storage will be considered depending on facilities and material available.

## 6 Responsibility for obtaining consent

It is the responsibility of the clinician who requests the test to ensure that the patient has given informed consent for the test to be carried out and for material to be stored where necessary.

## 7 Responsibility for transport of samples

It is the responsibility of the referring centre to ensure that samples arrive at the laboratory in suitable condition i.e. promptly, in the correct container, packaged and transported according

to current legislation, labelled with the patient's name and a second identifier e.g. date of birth or hospital/NHS number and accompanied by a completed request form.

## 8 Unsuitable samples

- The laboratory has clear procedures for dealing with unsuitable samples which include liaison with the referring clinician and issuing of a written report.
- Incorrect labelling of sample (i.e. discrepancy between name on sample and name on referral form or no name on sample or no name on referral form). Following discussion with the referring clinician if the labelling discrepancy cannot be resolved, the sample may not be processed.
- Sub-standard samples. Genetic testing may be attempted if repeat sampling would not be appropriate or possible as in the case of a prenatal diagnosis sample or death of the patient e.g.
    - Sample in wrong tube.
    - Sample clotted.
    - Sample delayed in transit for more than 5 days.
    - Non-viable sample (lysed, frozen, fixed).
    - Broken sample tube.
- High-risk samples requiring containment level 3 facilities e.g. known cases of CJD/vCJD, TB cannot be processed.
- Samples with inappropriate referral reasons may not be processed.

## 9 Disposal of samples

Samples will be disposed of according to national guidelines.

## 10 Reports

- Final reports will be issued when chromosome or ArrayCGH analysis is complete. Reports will include a karyotype or array result using current ISCN where appropriate, and a clinically relevant interpretative comment and reference to the inclusion of relevant literature where appropriate.
- All prenatal and neonatal reports will be promptly communicated to the referring centre by telephone and/or fax.

## 11 Turnaround Time

The laboratory will aim to provide reports within the current reporting guidelines indicated by ACGS Best Practice Guidelines, or agreed with the Genetics Consortium. These are given on our website (http://www.southwestthamesgenetics.nhs.uk/cyto_default.shtml)

We aim to report 90% of our cases within the above timeframes, but unfortunately this is not always possible. In order to keep the users of our service informed, current median reporting times and target achievement are displayed on our website and update monthly.

## 12 Audit of Outcome

Data will be provided for data monitoring and/or chromosome anomaly registers as required.

## 13 Quality Assurance

This Laboratory is CPA accredited and participates in external quality assessment schemes relevant to the test repertoires (CEQAS in Clinical Cytogenetics). The laboratory also complies with Best Practise Guidelines issued by the ACGS. A copy of our quality policy is available on our website.