Molecular Genetics
and
Molecular Pathology
External Quality Assessment

Schemes 2016

Laboratories are invited
to apply for participation in the
UK NEQAS for Molecular Genetics
2016-17 EQA schemes
UK NEQAS for Molecular Genetics invites your laboratory to participate in the External Quality Assessment (EQA) schemes provided for 2016. The EQAs available to your laboratory are detailed in this document however if you have any queries or require further information then please don’t hesitate to contact the Scheme on info@ukneqas-molgen.org.uk

Online registration for 2016 is now open and laboratories are invited to apply for participation. Registration for the molecular preimplantation genetic diagnosis EQA scheme closes on 31st January, 2016. Registration for some of the molecular pathology EQA schemes will close on 29th February, 2016. Registration for all other EQA schemes will close on 31st March, 2016.

Registration details
• Log into your account on the UK NEQAS for Molecular Genetics website (www.ukneqas-molgen.org.uk).
• Click the “Registration” button and select the scheme(s) in which you wish to participate.
• Please check that your laboratory details are up to date including the laboratory delivery address, primary contact email address and telephone number to ensure the smooth running of the scheme.
• Each laboratory will be charged an annual registration fee of £150 (regardless of the number of schemes applied for). This will be invoiced automatically. If your laboratory participates only in pilot schemes then this fee will be waived.
• If you do not have a website account then please visit the website (www.ukneqas-molgen.org.uk) and follow the instructions by clicking on “Join Now”. Once you have had your account processed then you will be able to register to participate in the EQAs.

Please note: for EQAs with multiple distributions throughout the year, you only have to register once to participate in all EQA rounds.

UK NEQAS for Molecular Genetics is hosted within the Department of Laboratory Medicine, The Royal Infirmary of Edinburgh, NHS Lothian, Edinburgh, UK therefore the Scheme invoicing will be performed by NHS Lothian. If you require a purchase order to register for the EQA scheme then please contact the Scheme for account details. Please ensure the annual registration fee is included in the total amount stated on the purchase order.

Many EQAs provided by UK NEQAS for Molecular Genetics are accredited to ISO 17043. These are indicated by * in this document. The full accreditation scope can be accessed at http://www.ukas.com/wp-content/uploads/schedule_uploads/00013/8505Proficiency%20Testing%20Multiple.pdf

UK NEQAS for Molecular Genetics follows the UK Joint Working Group for Quality Assurance Conditions of Participation. By registering for an EQA scheme you are agreeing to abide by these conditions of participation. Details can be found in the Participant Manual available on the UK NEQAS for Molecular Genetics website. Please note, all EQA reports must be submitted in English.

UK NEQAS for Molecular Genetics is a not-for-profit, self-funding organisation. The Scheme recognises the financial constraints being imposed upon many laboratories and therefore has minimised the fee increase as much as possible. So far in that many participation fees along with the annual registration fee have been frozen, otherwise a small increase has been applied. EQA fees are zero rated for VAT/Tax except for non-Scottish UK participants where VAT will be charged at standard rate. This VAT can be recovered by the NHS authority. If you require any further information then please contact the Scheme.

Participation fees for each EQA are detailed in the appropriate sections throughout this document.
**GENOTYPING & INTERPRETATION SCHEMES**

Twenty-four genotyping and interpretation disease EQAs are provided during 2016 (listed in panel on the left). These EQAs require DNA samples to be genotyped and full interpretative reports submitted. Genotyping, Interpretation and Clerical Accuracy will be assessed. If any dates change then you will be notified by the Scheme.

**MOLECULAR CORE DISEASES**

**SAMPLES**

DNA samples will be distributed for three clinical case scenarios for each disease scheme. The samples will be dispatched in two distributions. The first distribution comprises of the following disease EQAs; BRCA (including one sample for a full gene screen), Cancer panel pilot, C9orf72, CMT1A & HMSN, DM, D/BMD, FH, FRAX, LMNA, LongQT, Lynch syn, mtDNA, POLG, PWS&AS and Rett syn. The testing period is 12 weeks and samples will be dispatched on Monday 13th June, 2016.

The second distribution provides 6 weeks for testing and will take place on Monday 25th July, 2016. This distribution includes samples for the following EQAs; ACH, CF, DRPLA, Fabry disease, HD, MCADD, MCC & sexing and SCAs.

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**NEW for 2016** - CANCER PANEL PILOT 2016

To reflect the introduction of panel testing, a pilot EQA will be provided for cancer testing. This will include testing for mutations in the most commonly tested genes e.g. PTEN, APC etc and will not overlap with the clinical cases assessed in the familial breast and ovarian cancer or Lynch syndrome EQAs. Laboratories performing exome and whole genome sequencing are encouraged to participate. There will be a participation fee of £100 charged for this pilot.

**REPORT SUBMISSION, SCORING & SCHEME REPORTS**

The deadline for return of all submissions for these EQAs is Friday 2nd September, 2016.

Individual laboratory scores and the UK NEQAS for Molecular Genetics annual report (including the individual disease scheme reports) will be published in November, 2016. An appeals process will be available and the final report will be published before the end of 2016.

The cost of participation in each disease EQA is £360.

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**MOLECULAR RAPID ANEUPLOIDY SCHEME**

This EQA is offered in collaboration with CEQAS (Cytogenetic European Quality Assessment Service). Full interpretative reports are required and Genotyping, Interpretation and Clerical Accuracy will be marked by both Molecular Genetic and Cytogenetic assessors. Participant appeals and poor performance are ratified by both EQA schemes.

**SAMPLES**

DNA samples will be distributed for three clinical case scenarios on Monday 5th September, 2016. The samples are available at two different concentrations and will be distributed according to the method each laboratory analyses the samples i.e. QF-PCR, BoBs or MLPA. Please inform the Scheme of your laboratory’s preferred method of analysis and the appropriate samples will be dispatched.

**REPORT SUBMISSION, SCORING & SCHEME REPORTS**

Four weeks is given for testing and Individual laboratory scores and the EQA scheme report (pre-appeals process) will be published in December 2016. The appeals process will take place during December 2016/January 2017 with the finalised scheme report being published early 2017. The cost of participation in this EQA is £355.
INTERPRETATION ONLY SCHEMES
These disease EQAs are web-based interpretation only schemes (listed in panel on right). Clinical case scenarios and corresponding genotyping results will be provided on Monday 25th July, 2016 (along with the second distribution of samples for the full genotyping/interpretation schemes).
**No samples will be distributed for these schemes.**
Participants are required to submit full interpretative reports via their laboratory website account. Reports can be submitted during a six week period with the final report submission deadline on Friday 2nd September, 2016. Interpretation and Clerical Accuracy will be assessed. Individual laboratory scores and the disease scheme reports will be published in November 2016. The appeals process will take place during November and early December, with the final report being published before the end of 2016.

The cost of participation will be £180 for each interpretation only disease EQA.

PATHOGENICITY OF SEQUENCE VARIANTS EQA
We are offering the pathogenicity of sequence variants interpretative EQA as a full scheme for 2016, therefore performance standards will be applied. Participants will be provided with three cases with sequence variants from lesser known genes for assessment of pathogenicity in their usual manner. The scheme aims to assess whether ACGS best practice guidelines are being applied, which tools are being applied to variant assessment and whether classification is consistent across different laboratories. The format of the EQA is to assess the process of determining pathogenicity rather than being based on prior knowledge of the disease. The documentation will be provided on Monday 13th June, 2016 (along with the first core disease sample distribution). **No samples will be distributed for this scheme.**

The cost of participation will be £180.

BLOOD SPOT SCHEMES
**CF TESTING ON BLOOD SPOTS**
This EQA will be offered in the same format as in previous years with four rounds of EQA throughout the period April 2016 to March 2017. Three blood spot cards per EQA round will be distributed during April 2016, September 2016, November 2016 and February 2017. Each round will have a testing period of 4 weeks. Genotyping results should be submitted using a results proforma (provided on website). Full interpretative reports are not required. Laboratory genotyping results will be assessed by two independent assessors and a scheme report plus individual laboratory scores will be published following each round of EQA.

**MCADD TESTING ON BLOOD SPOTS - c.985A>G testing**
We are pleased to offer EQA for the MCADD molecular testing for the common mutation (c.985A>G) on blood spots in the same format as the CF blood spot scheme. There will be four rounds of EQA throughout period April 2016 to March 2017 with distributions taking place in April 2016, September 2016, November 2016 and February 2017.

**FEES FOR 2016**
There is no increase in participation fees for the CF and MCADD blood spot schemes from previous years. In order to maintain this freeze in fees, the samples for both schemes will be distributed together when possible. The cost of participation will be £800 per disease scheme. This includes four distributions for each scheme.
Preimplantation Genetic Diagnosis schemes 2016

- PGD Molecular (PGD 2016 DMD) *
- PGD Array/Next Generation Sequencing for polar body testing pilot EQA (PGD array/NGS pilot – polar body)
- PGD Array/Next Generation Sequencing for trophectoderm/blastomere testing pilot EQA (PGD array/NGS pilot – troph/blasto)

PREIMPLANTATION GENETIC DIAGNOSIS MOLECULAR EQA *Registration closes 31st January 2016*

The 2016 PGD molecular EQA scheme will assess testing for Duchenne muscular dystrophy (DMD). The scheme follows the PGD process as closely as possible and provides samples and information on a “couple” wishing PGD for and their “embryos” for analysis.

There are two stages to this EQA as detailed below:

Stage 1 – Feasibility Study
DNA samples are distributed from a family with a history of DMD who wish to undergo PGD. Participants are required to perform a feasibility study to determine if PGD could be offered to this couple. Six weeks will be given for testing and samples will be distributed in February 2016.

Stage 2 – PGD Case Study
Participants who are able to offer PGD to the couple in Stage 1 will automatically be entered into Stage 2 of the EQA. This stage involves the distribution of lymphocyte cells as biopsied “embryo” cells. Laboratories can request either single cells, a batch of six cells or both, for testing. Participants are required to perform PGD for DMD and report using their usual laboratory format stating whether or not the embryos should be transferred.

The submitted reports will be assigned genotyping, interpretation and clerical accuracy scores anonymously by a panel of expert advisors against peer ratified marking criteria. Samples will be distributed following stage 1.

The cost of participation for this EQA will be £115 (stage 1) and £245 (stage 2).

PREIMPLANTATION GENETIC DIAGNOSIS FOR TESTING OF:
- POLAR BODIES
- BLASTOMERE
- TROPHECTODERM

BY ARRAY/NGS PILOT EQA

This pilot EQA is provided by in collaboration with CEQAS (Cytogenetic European Quality Assessment Service) and has been developed to cover polar body testing, blastomere testing and trophectoderm testing by arrays and/or next generation sequencing. Laboratories can choose which aspect of this pilot EQA they would like to participate in.

Please register for either (or both) the polar body pilot EQA or the trophectoderm/blastomere pilot EQA. Once you have registered then the Scheme will contact you for further information.

The cost of participation in each of these pilot EQAs will be £200 (subsidised).
NEXT GENERATION SEQUENCING PILOT EQA

The 2016 pilot EQA scheme for Next Generation DNA sequencing is a collaboration between the EMQN and UK NEQAS for Molecular Genetics. The project is co-ordinated by a Specialist Advisory Group (SAG) set up by both organisations. We have reviewed our experiences from previous NGS schemes and consequently will be providing the scheme as two parts covering both somatic and germline testing. The format of the EQA will be reviewed following the end of the 2015 pilot and we invite you to register to your interest in participating the germline and or somatic NGS pilot 2016.

The cost of participation in each of these pilot EQAs is £100 (subsidised).

NON-INVASIVE PRENATAL TESTING FOR ANEUPLOIDIES PILOT EQA

A new pilot will be provided during 2016 for the non-invasive prenatal testing for aneuploidies. This is currently in development but full details will be emailed to all laboratories as soon as they are available. All interested laboratories will be invited to complete an online survey. If you would like to be contacted then please email the Scheme on info@ukneqas-molgen.org.uk.

*NEW for 2016*
DNA QUALITY MEASUREMENT EQA

Whilst working with NHS England and Genomics England as part of the UK 100,000 Genomes Project during 2014 and 2015, the Scheme has become aware of the variation in the accuracy of measuring the quantity and quality of DNA between laboratories. Therefore to help standardise and promote accurate measurements an EQA run will be offered during 2016 to compare such measurements reported by laboratories.

Three DNA samples will be distributed for participants to assess the quality and quantity of each sample and return the data to the Scheme for analysis.

Overall comparisons between laboratories, approach followed and methods performed will be provided.

The cost of participation will be £300 for participation in this EQA run.

Laboratories linked to a NHS Genomic Medicine Centre do not need to register for participation in this EQA scheme. Registration is automatic and the participation fees will be covered by NHS England.
*NEW for 2016*

**G-TACT**

*Genetics Training and Competence Tool*

Individuals working in genetic laboratories are increasingly becoming required to demonstrate training and competency for different roles they fulfil within the laboratory. Therefore UK NEQAS for Molecular Genetics has developed an online training and competency tool to provide a source of external review and evidence continual professional development for the individual.

The **G-TACT** scheme involves entering a virtual genetics laboratory where the participant can navigate between four workstations; sample reception, duty scientist, data analysis and report authorisation. Each station will have tasks to complete appropriate to that role and a participation certificate will be issued once the station EQA module is complete.

Registration will enable annual access to three mainstream workstations; sample reception, duty scientist and data analysis, for up to eight individual users. This registration will cost £480. Additional users can be added in batches of four users at a reduced fee of £200 per batch. Please contact the Scheme if you would like to include additional users.

Registration for participation in the report authorisation assessment workstation can be purchased for a fee of £180 per user (up to 5 individuals) with a sixth included at no extra charge. Additional users can be added for a subsidised fee of £150 per person.

Please register to participate in the G-TACT 2016 scheme and you will be contacted by the Scheme to determine which user option your laboratory would like to opt for.

Please be assured that the Scheme is aware of the financial constraints on laboratories and therefore has endeavoured to keep fees as low as possible to enable maximum participation across laboratories.

*NEW for 2016*

**EDUCATIONAL FRAXA REFERENCE SAMPLES**

In collaboration with the National Institute of Biological Standards and Controls (NIBCS) UK NEQAS for Molecular Genetics is pleased to provide laboratories with two Fragile X syndrome reference samples to enable the assessment of the accuracy of in-house FRAXA allele sizing.

Laboratories are invited to test the two DNA samples and submit their FRAXA allele sizing results to the Scheme. This is an educational voluntary exercise, no scores will be assigned but feedback comments and allele sizes will be provided by the Scheme to laboratories that submit results.

Participants of the FRAX 2016 EQA will receive this sample along with their 2016 distribution. No charge will be applied to laboratories.
*NEW for 2016*

**DNA EXTRACTION EQA SCHEMES**

The implementation of whole genome sequencing and next generation sequencing panels requires high quality DNA to allow good sequencing data to be obtained.

UK NEQAS for Molecular Genetics has been delivering bespoke EQA schemes for the UK 100,000 Genomes Project to assess the standard of DNA samples extracted by laboratories supplying samples for inclusion into the project.

The Scheme is delighted to now offer participation in these EQAs to all interested laboratories. These EQAs provide laboratories with a measure of the quality of the DNA extracted in-house and enables benchmarking with other centres using similar extraction methods.

Laboratories linked to a NHS Genomic Medicine Centre do not need to register for participation in the Blood or Tissue DNA extraction schemes. Registration is automatic and the participation fees will be covered by NHS England.

**DNA EXTRACTION FROM BLOOD SAMPLES**

There will be two EQA runs provided during 2016 to assess the quality and quantity of DNA extracted from blood samples. For each run, three peripheral blood samples anticoagulated in EDTA (ranging from 1ml to 5ml) will be distributed to participants. Laboratories will be required to extract DNA from the total volume of blood and return the DNA to the Scheme for quality assessment.

The DNA samples will be assessed for total volume, total amount of DNA, level of fragmentation and library preparations made which will be assessed using a bioanalyser. Each sample will be given a quality rating. Overall comparisons between methods will be provided.

The cost of participation will be £1,000 for participation in one EQA run.

**DNA EXTRACTION FROM TISSUE SAMPLES**

During 2016 there will be two EQA runs to assess the quality and quantity of DNA extracted from tissue samples. This will include formalin fixed paraffin embedded (FFPE) tissue and fresh frozen tissue. Laboratories will be required to extract DNA from the total sample supplied and return the DNA to the Scheme for quality assessment.

The DNA samples will be assessed for total volume, total amount of DNA, level of fragmentation using commercial kits. Each sample will be given a quality rating. Overall comparisons between methods will be provided.

The cost of participation will be as follows:
- £400 for one EQA run for DNA extraction from FFPE tissue
- £600 for one EQA run for DNA extraction from FFPE and fresh frozen tissue

**DNA EXTRACTION FROM SALIVA SAMPLES**

During 2016 there will be two EQA runs to assess the quality and quantity of DNA extracted from saliva samples. For each run, three saliva samples (ranging from 0.5ml to 2ml) will be distributed to participants. Laboratories will be required to extract DNA from the total sample supplied and return the DNA to the Scheme for quality assessment.

The DNA samples will be assessed for total volume, total amount of DNA, level of fragmentation and library preparations made which will be assessed using a bioanalyser. Each sample will be given a quality rating. Overall comparisons between methods will be provided.

The cost of participation will be £200 for participation in one EQA run (subsidised).
COLORECTAL CANCER TESTING EQA
NON-SMALL CELL LUNG CANCER TESTING EQA
MELANOMA TESTING EQA

*Registration closes 29th February 2016*

- These EQA schemes will comprise of 2 rounds of assessment (total of 9 EQA samples) during the period April 2016 to March 2017 and will involve the testing of tumour samples for sequence changes in the genes listed below.

<table>
<thead>
<tr>
<th>EQA</th>
<th>Required testing (assessed)</th>
<th>Validated genes (assessed if tested)</th>
<th>Validated genes (not assessed)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Colorectal</td>
<td>KRAS and NRAS</td>
<td>BRAF and PIK3CA</td>
<td></td>
</tr>
<tr>
<td>Lung</td>
<td>EGFR</td>
<td>ALK rearrangement (by any method)</td>
<td>KRAS, BRAF and PIK3CA</td>
</tr>
<tr>
<td>Melanoma</td>
<td>BRAF</td>
<td>KIT</td>
<td>NRAS</td>
</tr>
</tbody>
</table>

- The samples will be supplied along with clinical case scenarios and participants are required to extract DNA/RNA and test for mutations according to normal laboratory procedures. The findings should be reported in the usual report format with reference to the clinical background supplied.
- Run 1 = April 2016 (5 EQA samples)
- Run 2 = October 2016 (4 EQA samples)
- The genotyping results will be scored according to the tests performed and poor performance criteria (see website for criteria) will be applied as follows:
  - **Colorectal cancer EQA** – KRAS and NRAS test results
  - **Lung cancer EQA** - EGFR test result and the ALK fusion gene test
  - **Melanoma EQA** – BRAF and KIT test result
- The reports will also be assessed for interpretation of the results and clerical accuracy by at least two independent assessors against peer reviewed marking criteria.
- All EQA returns will be submitted via your laboratory UK NEQAS for Molecular Genetics website account.
- On completion of each round of EQA participants will receive a scheme report and a full summary of their laboratory results.

**SAMPLES PROVIDED**

Once you have registered for the EQA(s) you will be contacted by the Scheme to request your sample type preference. Participants will be provided with either:
- Rolled sections of formalin fixed paraffin embedded (FFPE) tumour tissue
- Rolled sections of FFPE tumour tissue plus one slide mounted paraffin section
- Mounted slides of FFPE tumour tissue for each case.

**FEES FOR 2016-17**

There is no increase in participation fees for these EQAs. The fee of £600 covers annual participation (both EQA rounds) per disease EQA.
ADDITIONAL LUNG BIOMARKERS PILOT EQA

Following the success of the first additional lung biomarkers pilot EQA scheme, another pilot scheme will be provided during 2016. This will consist of two distributions with samples being sent in April and October. Laboratories will be supplied with formalin fixed paraffin embedded samples for ROS1 and RET testing by RT-PCR of FISH and MET testing by FISH. Reports will be assessed for genotyping accuracy.

The fee for this EQA is £100 which covers participation in both runs of the EQA.

GASTRO-INTESTINAL STROMAL TUMOUR TESTING (GIST) EQA

This EQA will be provided in the same format as the other Molecular Pathology EQAs but with only one distribution taking place in September 2016.

Participants will be required to test the samples for sequence changes in the following genes: KIT and PDGFRA.

The reports will be assessed for genotyping accuracy by two independent assessors against ratified validated results.

As in previous years this is a full EQA provided with performance monitoring of all genes tested (see website for performance criteria).

The fee for this EQA is £200.

*NEW DEVELOPMENT for 2016*

MICROSATELLITE INSTABILITY EQA

The format of this EQA has been expanded from previous EQA runs. As many laboratories routinely test samples for MSI and continue with further testing then this EQA scheme has developed to reflect this practice.

Participants will be required to test three pairs of normal and tumour samples (either DNA or FFPE tissue samples) according to normal procedures, and submit their results as full interpretative reports. The EQA will include MSI testing (as in previous years), then laboratories will have an option to test and report the results of MLH1 promoter methylation testing and BRAF mutation analysis.

The MSI results will be assessed for genotyping accuracy and all other tests will be classed as pilot tests.

Samples will be distributed in September 2016.

The fee for this EQA is £200.

SARCOMA PILOT EQA

Following the success of the pilot EQAs for testing of sarcomas by RT-PCR and FISH during 2014 and 2015, a further pilot EQA will be provided during 2016. This pilot EQA is provided by in collaboration with CEQAS (Cytogenetic European Quality Assessment Service). Laboratories can test the EQA samples by either method or both according to their clinical service offered.

This EQA will comprise of two rounds of assessment during the period April 2016 to March 2017 and will involve the testing of FFPE tumour samples.

The fee for this EQA is £200 which covers participation in the two EQA runs.
**NEW for 2016**

UK NEQAS for Molecular Genetics along with PathXL have developed Tissue-i, an online tissue assessment EQA module to enable individuals to evidence their competency to select appropriate tissue areas for molecular testing.

Molecular pathology testing of solid tumour relies on the pathological review of H&E stained tissue sections to annotate tumour regions for enrichment by macrodissection. This ensures sample quality, sufficiency of tumour DNA for analysis and ultimately the precision of the molecular test result. For the use of sensitive molecular techniques this is critical to deliver high quality testing and allow accurate interpretation of the sequence variants detected. It is known that the annotation of tumour and estimation of neoplastic nuclei can be highly variable thus there is a need to measure the degree of variation between laboratories and standardise approaches to tumour annotation and analysis to promote high quality molecular pathology testing.

This module covers a range of tissue types (breast cancer, colorectal cancer, lung cancer, melanoma, ovarian cancer and prostate cancer). Participants can select which tissue type they wish to assess. They will be required to mark the regions of tumour they deem appropriate for macrodissection for molecular testing, drawing the boundary using the online EQA module. An estimation of the cellularity and percentage of neoplastic nuclei across the slide image and within the annotated region will be requested.

This online competency tool has been designed to provide a source of external review, benchmarking with peers and evidence continual professional development in the field of molecular pathology tissue assessment for the individual.

Registration will enable access to all five tissue types to up to 3 users for an EQA run. This registration will cost £480. Additional users can be added at a reduced fee of £100 per user. Please contact the Scheme if you would like to include additional users.

Please be assured that the Scheme is aware of the financial constraints on laboratories and therefore has endeavoured to keep fees as low as possible to enable maximum participation across laboratories.

Laboratories linked to a NHS Genomic Medicine Centre do not need to register for participation in this assessment. You will be contacted by the Scheme regarding participation covered by NHS England.
MOLECULAR TISSUE IDENTIFICATION PILOT

Laboratories test tissue samples using molecular fingerprinting assays for a variety of reasons e.g. confirmation of origin of tissue, sample switch detection, sample mislabelling. To provide laboratories with assurance of the quality of this testing and accuracy of data analysis, a second pilot EQA run will be offered.

Three EQA cases will be supplied. Participants will be required to extract DNA from FFPE tissue and perform molecular identification assays. Clinical case scenarios will be provided to allow interpretation of the result.

Samples will be distributed in September 2016.
The fee for this pilot EQA is £100.

*NEW for 2016*

BRCA TESTING IN OVARIAN CANCER (somatic and germline) pilot

UK NEQAS for Molecular Genetics in collaboration with EMQN are proud to provide a new pilot EQA scheme for ovarian cancer. This is supported by an educational grant from AstraZeneca.

The 2016 pilot scheme is currently underway but if your laboratory would like to participate in future schemes (germline and somatic testing) then please contact the Scheme on info@ukneqas-molgen.org.uk