UK NEQAS
Molecular Genetics

8505

www.ukneqas-molgen.org.uk

Ensuring high quality NIPT

Dr Sandi Deans

Consultant Clinical Scientist

Scheme Director, UK NEQAS for Molecular Genetics and Molecular Pathology, Edinburgh, UK

National Scientific Lead, Genomics Implementation Unit, NHS England, UK

UK NEQAS

International Quality Expertise

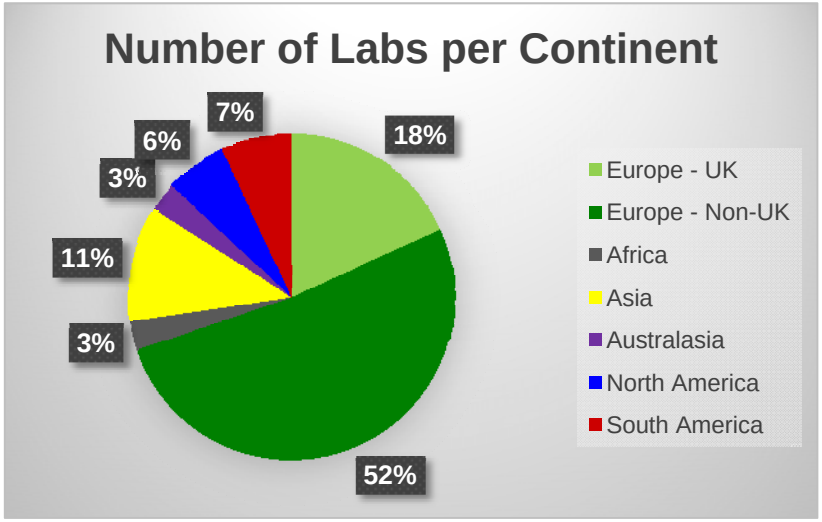
UK National External Quality Assessment Services

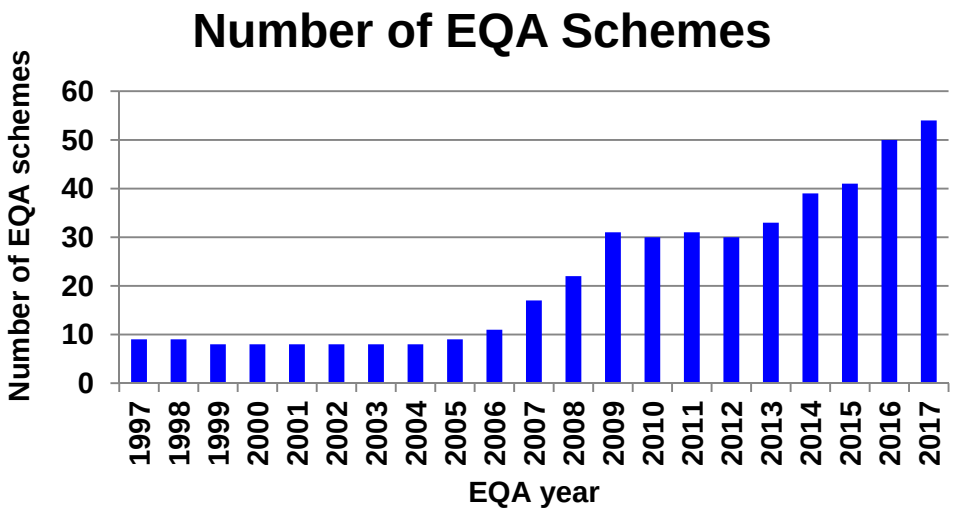
"helping to ensure clinical laboratory test results are accurate, reliable and comparable wherever they are produced"

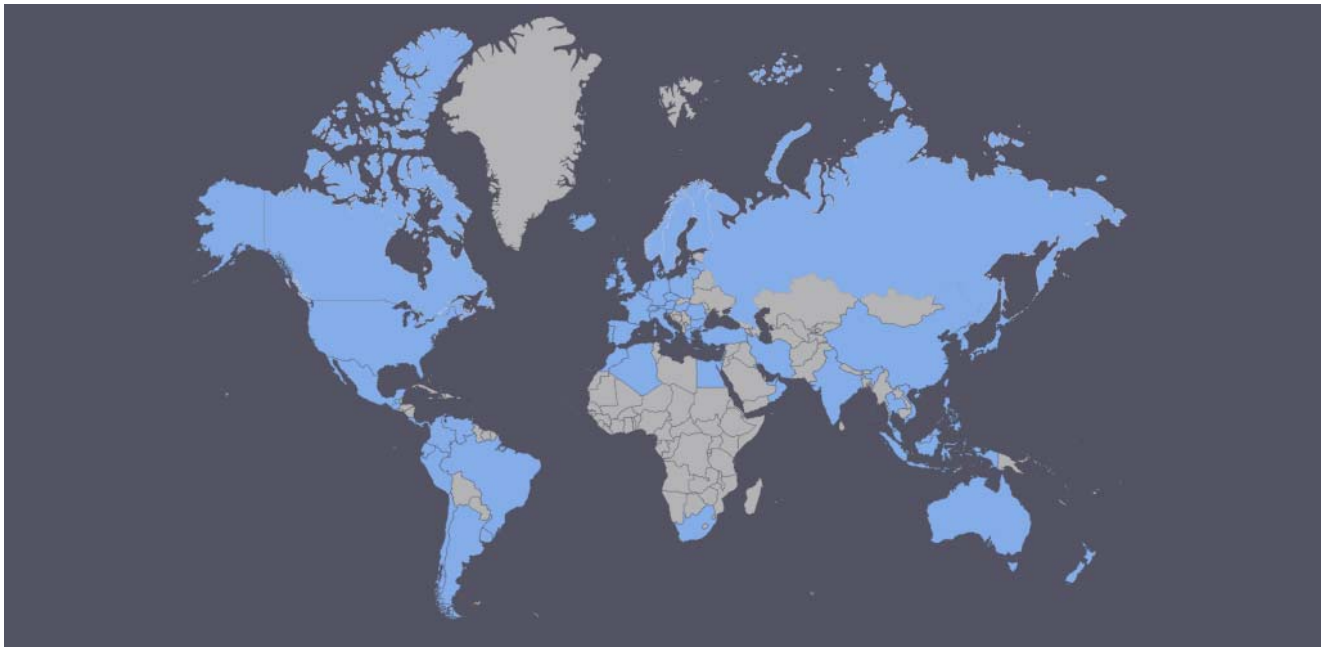
- Charity organisation
- Self funding, non-profit making schemes
- 25 specialist centres
- Over 45 years experience providing EQA
- ISO 17043 accredited EQA schemes
- Participants worldwide

Scheme Activity 2016

68 Countries





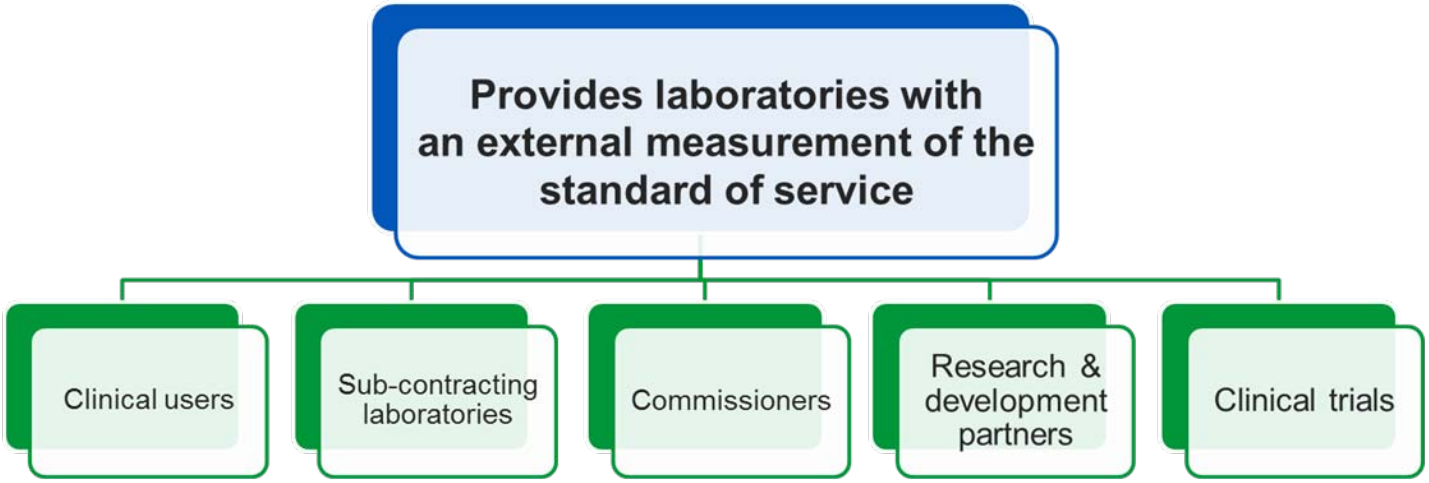


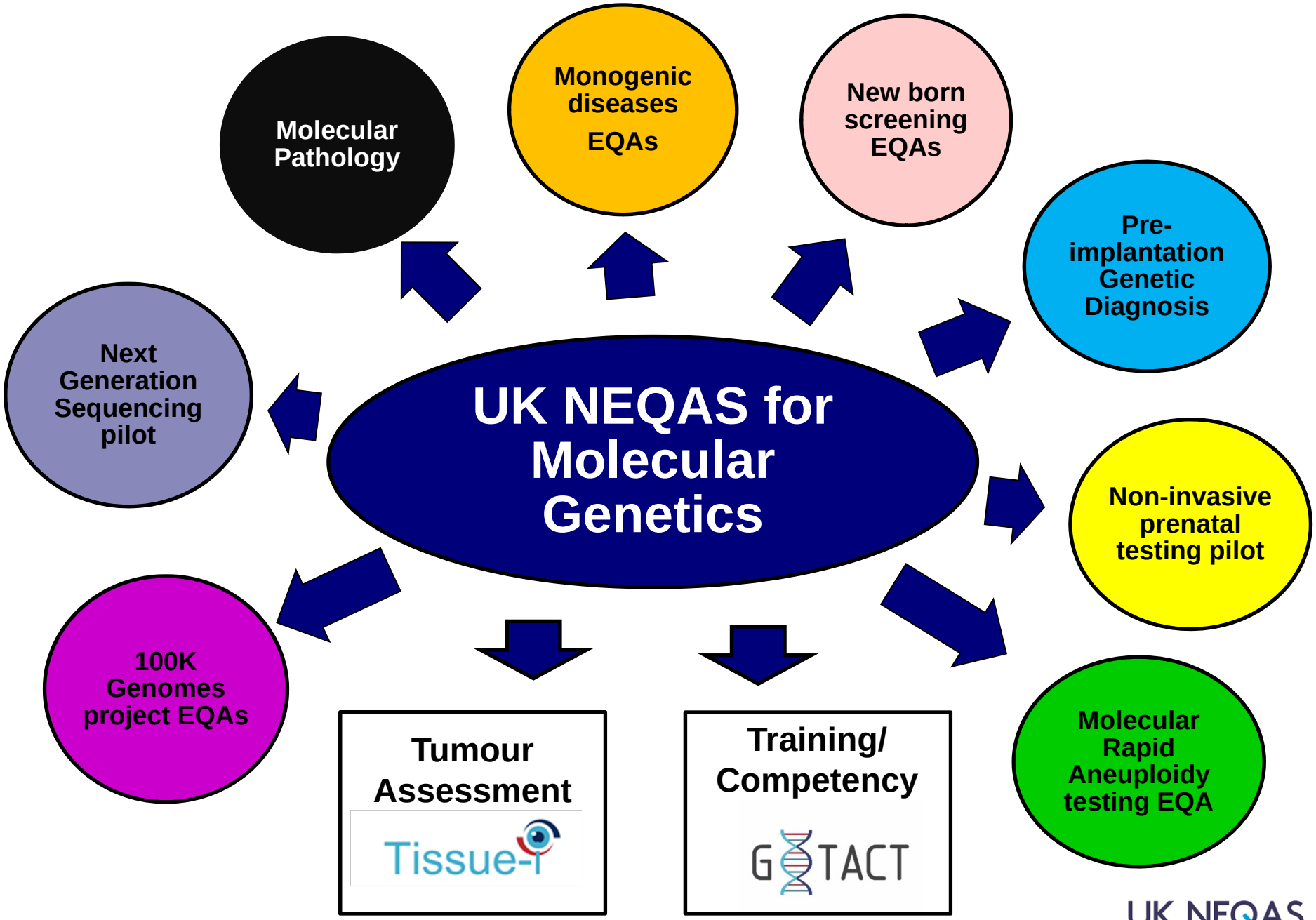
UK NEQAS
Molecular Genetics

Principles of EQA

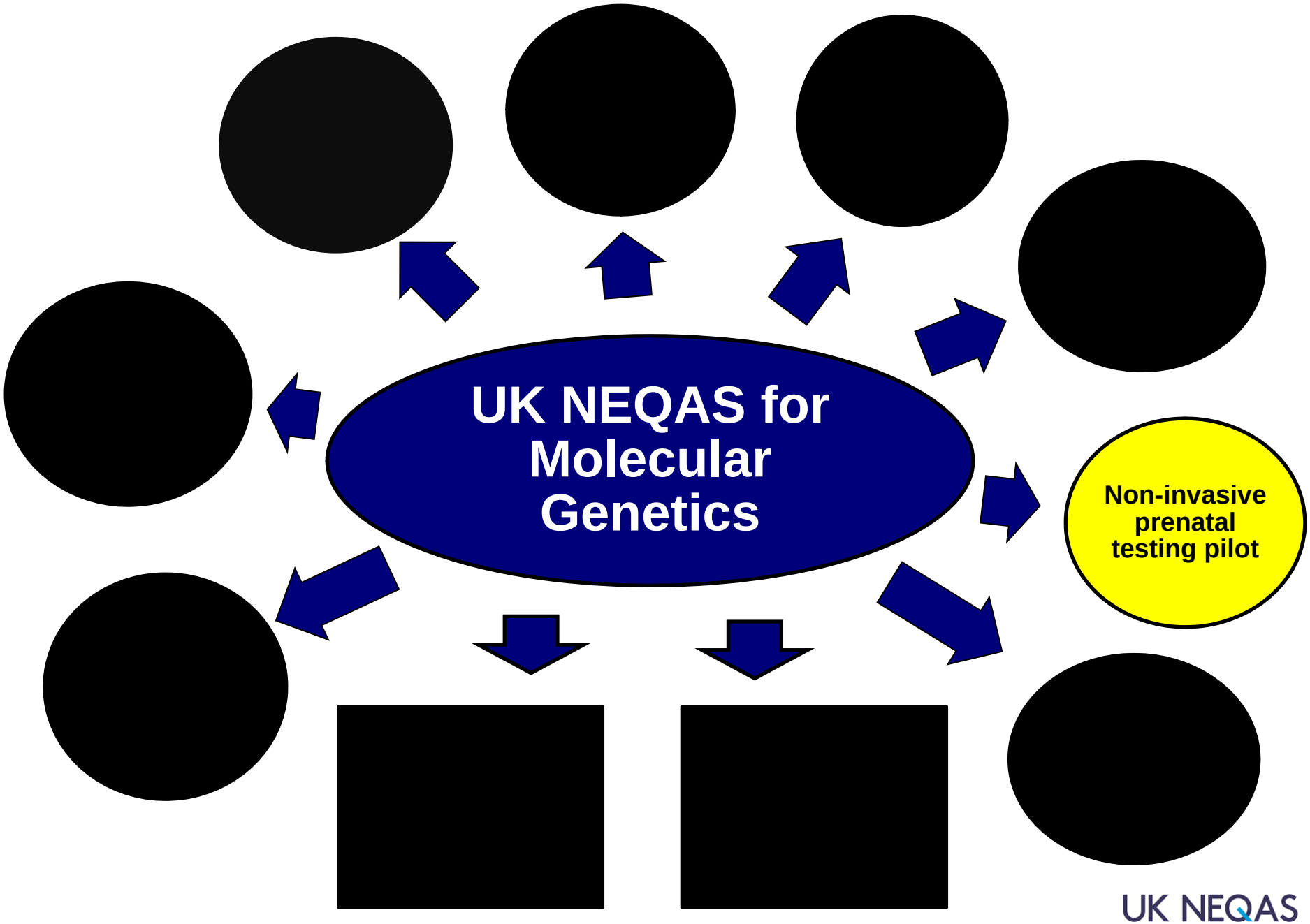
- Same samples sent to all participating laboratories
- Assessed for - genotyping accuracy
  - interpretation of the result
  - clerical accuracy of the report

- Agreed marking criteria ratified by peer Specialist Advisory Group
- Independently marked by team of assessors
- Continual performance monitoring
- On-going records of participation and performance





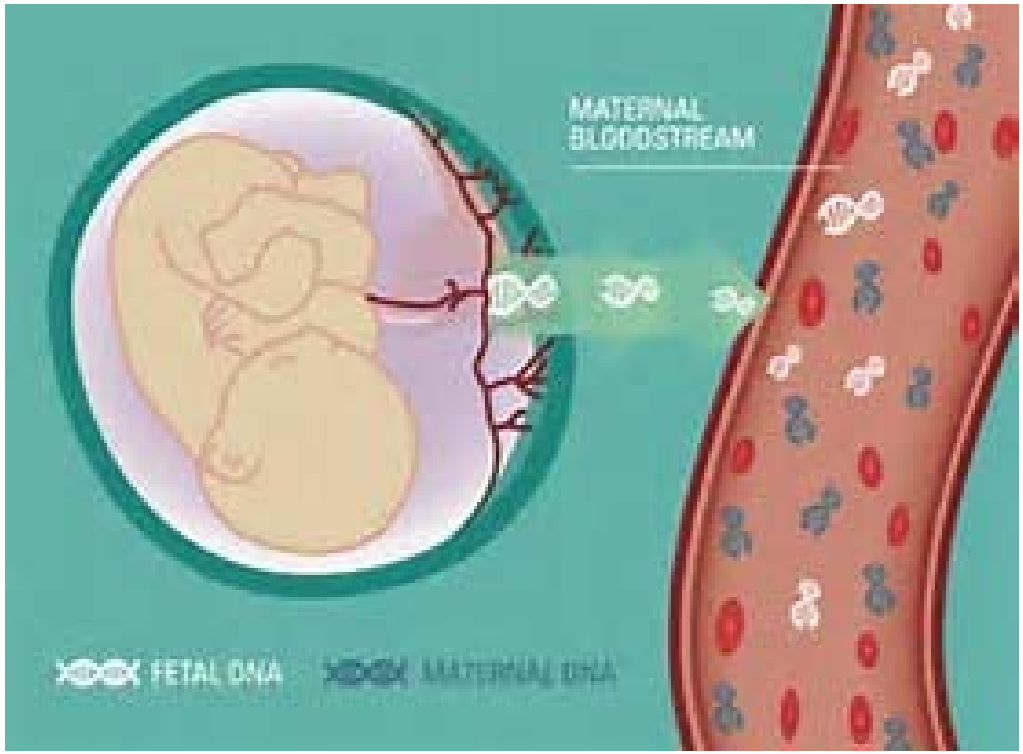
UK NEQAS
Molecular Genetics



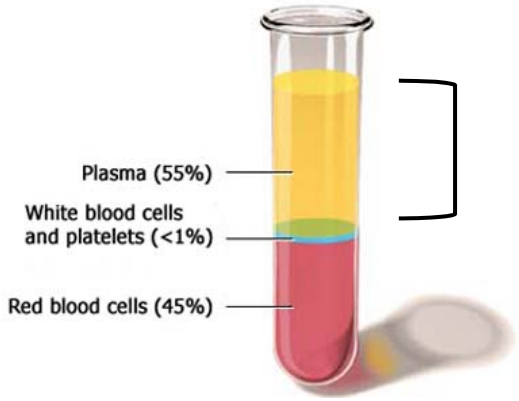
UK NEQAS
Molecular Genetics

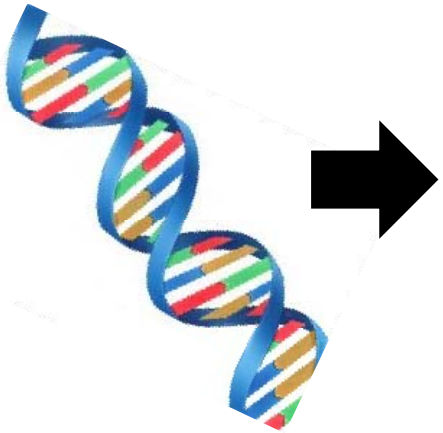
Non-invasive prenatal testing (NIPT) for aneuploidies

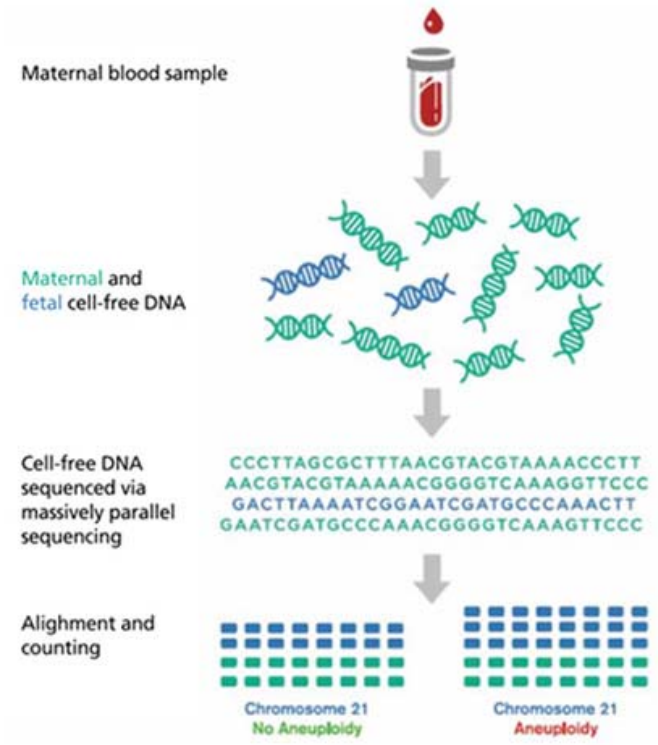
Detection of cell free fetal DNA in maternal blood



Non-invasive prenatal testing (NIPT) for aneuploidies



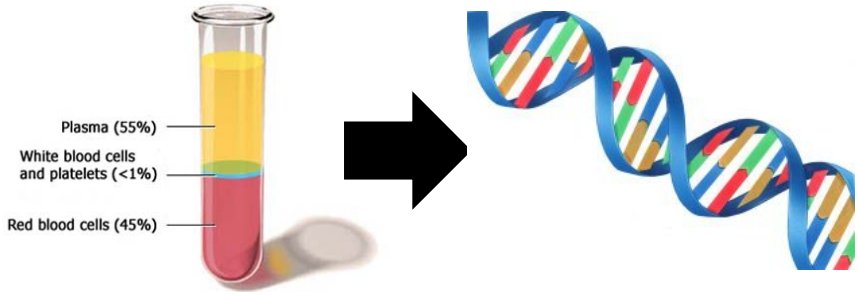




UK NEQAS
Molecular Genetics

Introduction of Non-invasive prenatal testing (NIPT) for aneuploidies EQA

- Participant demand
- Technically challenging
- Logistically challenging
- Difficult to obtain patient material
  – *ethics, availability, disease incidence*
- Collaboration of three International EQA providers



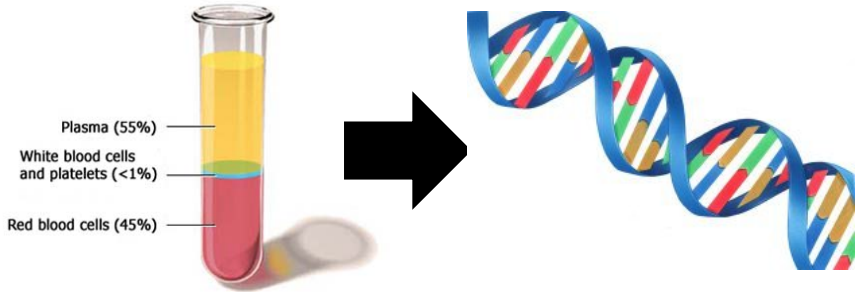
UK NEQAS Molecular Genetics

EMQN The European Molecular Genetics Quality Network

CEQAS Cytogenetic External Quality Assessment Service

Introduction of Non-invasive prenatal testing (NIPT) for aneuploidies EQA

- Participant demand
- Technically challenging
- Logistically challenging
- Difficult to obtain patient material
  – *ethics, availability, disease incidence*
- Collaboration of three International EQA providers



UK NEQAS Molecular Genetics

EMQN The European Molecular Genetics Quality Network

CEQAS Cytogenetic External Quality Assessment Service

STAGE 1 - Need to know what laboratories are actually doing……

2016 plan to implement NIPT EQA pilot

➢ Laboratory survey – January/February 2016

- Distributed to >1,200 laboratories
- NIPT for aneuploidies
- NIPD/NIPT for fetal sexing
- NIPD for single gene disorders

EQA for Non-invasive prenatal testing

Application to participate in 2016 pilot EQA

You are invited to apply to participate in pilot EQA schemes for non-invasive prenatal testing based on analysis of cell free DNA in maternal plasma.

A collaboration between three External Quality Assessment (EQA) providers (CEQAS, EMQN, and UK NEQAS for Molecular Genetics) has been initiated to provide a pilot EQA to assess the standard of testing for the presence of fetal aneuploidies (trisomies 13, 18 and 21), fetal sex testing and diagnostic testing in maternal plasma.

There is currently high demand for the provision of such an assessment and therefore we would like to invite your laboratory to apply to participate in these pilot EQAs which will be delivered during 2016.

The number of participants will be limited and in order for the EQA providers to select the participating laboratories then we require the following information. Please note that all contact information will be treated in confidence and the anonymised data may be published to help promote high quality testing.

*** 1. Please provide your contact details.**

Title	
First name	
Surname	
Email address	

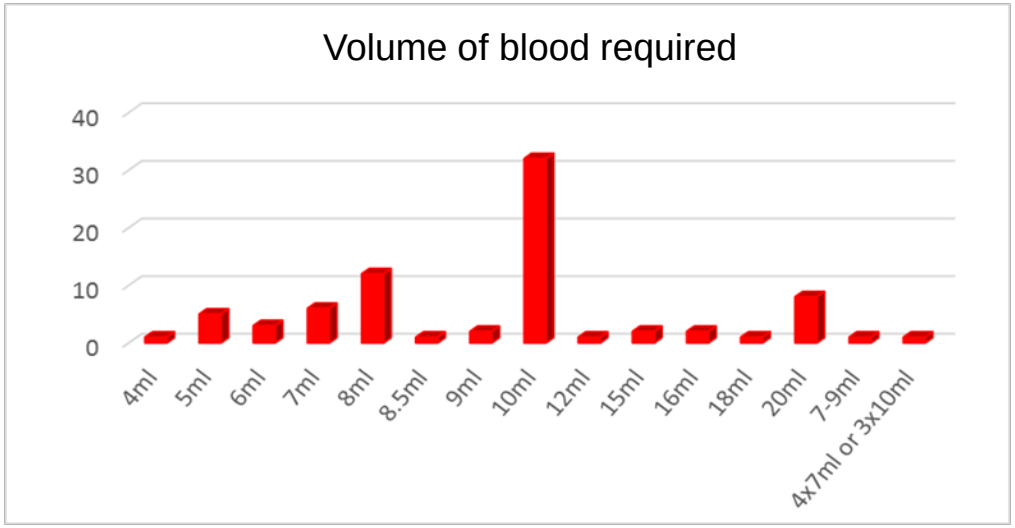
STAGE 1 - Laboratory survey 2016

UK NEQAS
Molecular Genetics

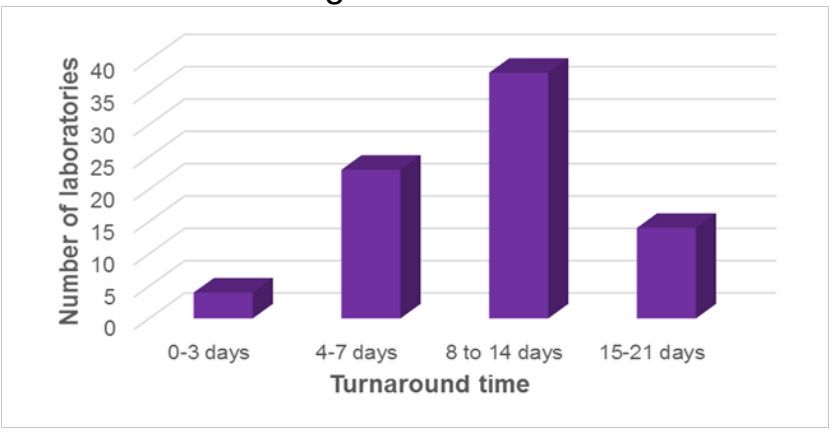
Response from 121 laboratories

61% laboratories provide NIPT for aneuploidies

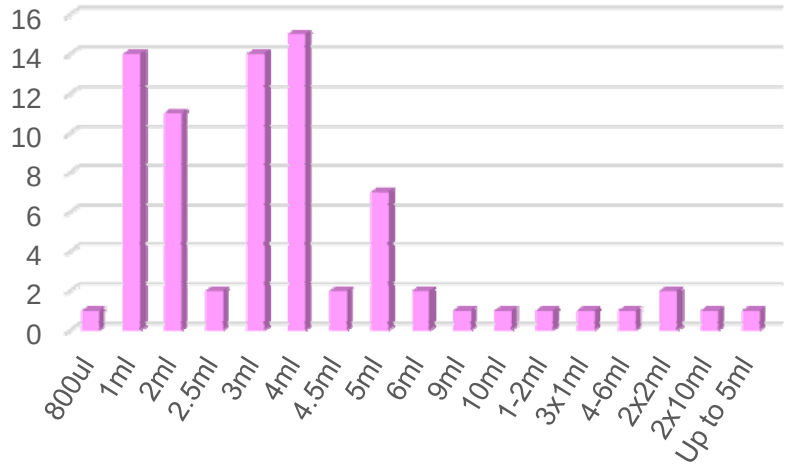
Number of samples tested per month *(mean = 565)*



Testing turnaround time



Volume of plasma required

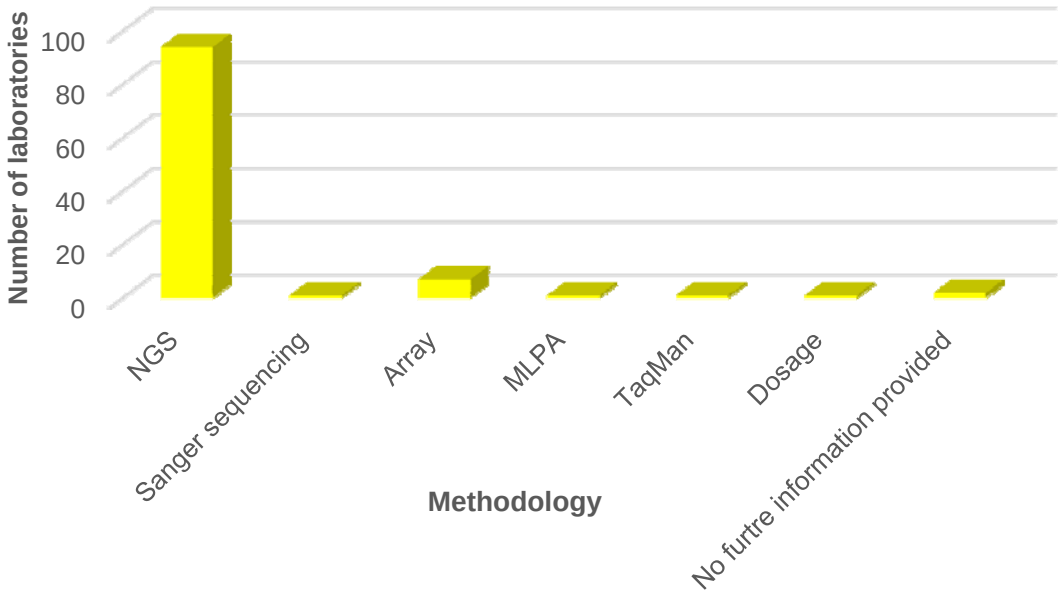


STAGE 1 - Laboratory survey 2016

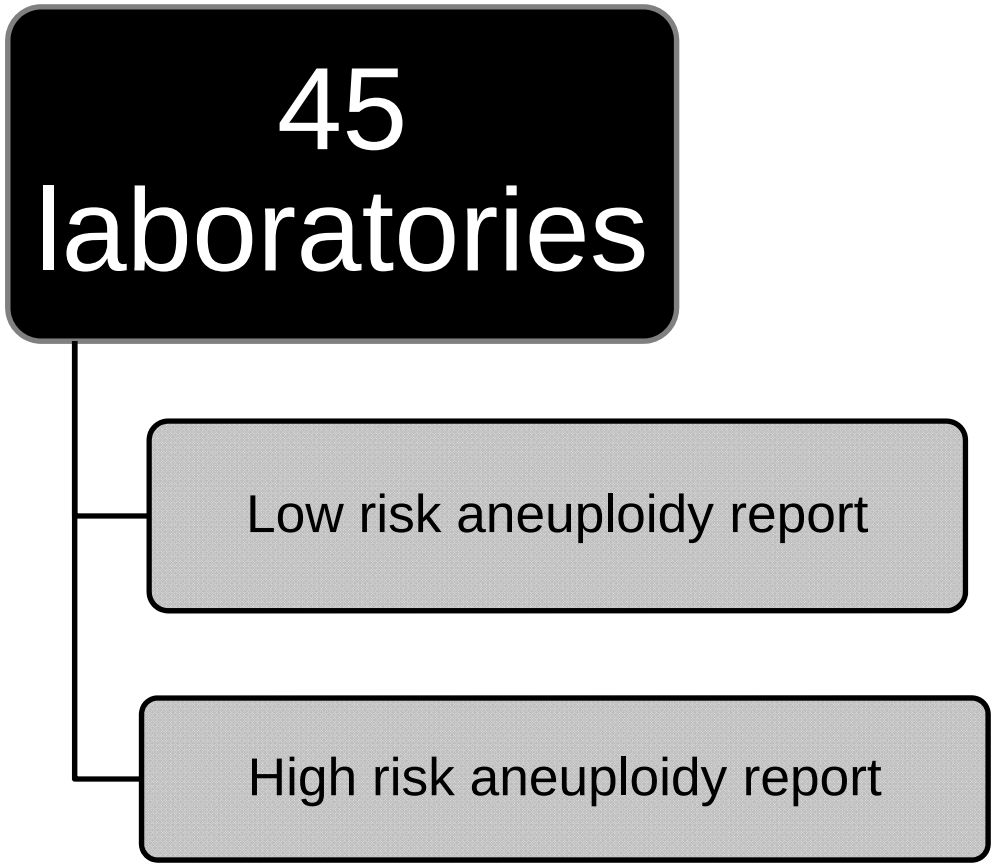
UK NEQAS
Molecular Genetics

Does the laboratory test for sex chromosome abnormalities?
Does the laboratory test for fetal sex?
Does the laboratory report fetal sex?
Do you measure fetal fraction as part of the assay?
Do you report fetal fraction as part of the assay?
What information is required for testing?
Does your laboratory outsource testing?
Future plans for testing 'other' abnormalities?
Future plans for NIPD?

Methodologies



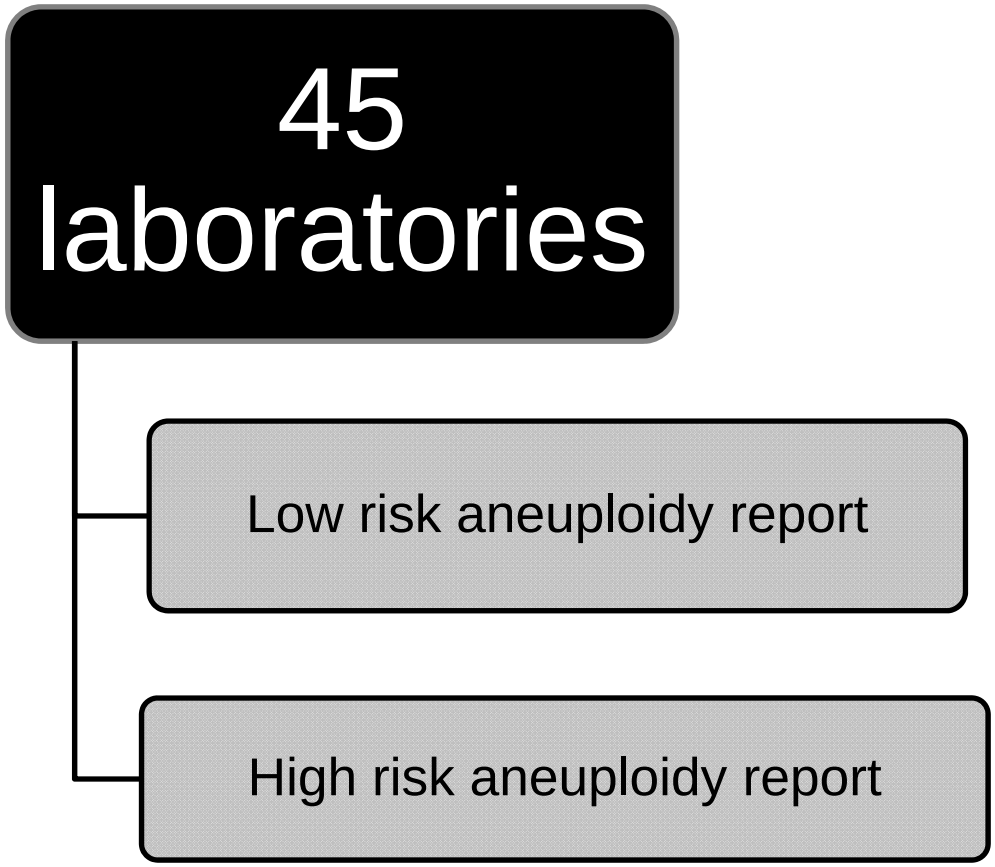
STAGE 2 - Review of reporting



Panel discussion at ISPD 2016 workshop

UK NEQAS
Molecular Genetics

STAGE 2 - Review of reporting



Panel discussion at ISPD 2016 workshop

Submitted to Prenatal Diagnosis
Deans Z.C. *et al.* Recommended practice for laboratory reporting of non-invasive prenatal testing of trisomies 13, 18 and 21: a consensus opinion

UK NEQAS
Molecular Genetics

STAGE 3 – Exploratory pilot (5 clinical laboratories)

Samples	Methods	Testing
UK RAPID Project – patient samples	WGS Illumina (multiple)	Trisomy 13
Artificial reference material A	Ion Proton	Trisomy 18
Artificial reference material B	SNP arrays	Trisomy 21
		Euploidy

CEQAS Cytogenetic External Quality Assessment Service

EMQN The European Molecular Genetics Quality Network

UK NEQAS Molecular Genetics

STAGE 3 – Exploratory pilot (5 clinical laboratories)

Samples	Methods	Testing
UK RAPID Project – patient samples	WGS Illumina (multiple)	Trisomy 18
	Ion Proton	Trisomy 21
	SNP arrays	Euploidy

Validated fetal result	Illumina NGS platform (different protocols)			Ion Proton NGS platform	SNP Array
	Lab 1	Lab 2	Lab 3	Lab 4	Lab 5
Trisomy 21	Trisomy 21, Normal 13 & 18	Trisomy 21	Trisomy 21 (NCV: 21.56)	Elevated risk for trisomy 21	T21 high risk >99%
Trisomy 21	Trisomy 21, Normal 13 & 18	Not tested	Not tested	Not tested	T21 high risk >99%
Trisomy 18	Trisomy 18, Normal 13 & 21	Trisomy 18	Trisomy 18	Elevated risk for trisomy 18	T18 high risk >99%
Euploidy	Not tested	No T21,18 or 13 *(would have requested repeat as on QC boundary)*	No evidence of trisomy 13,18,21	Normal, but low fetal fraction (~3.3%)	Not tested

CEQAS Cytogenetic External Quality Assessment Service

EMQN The European Molecular Genetics Quality Network

UK NEQAS Molecular Genetics

STAGE 3 – Exploratory pilot (5 clinical laboratories)

Samples	Methods	Testing
Artificial reference material A	WGS Illumina (multiple)	Trisomy 13
	Ion Proton	Trisomy 18
		Trisomy 21
		Euploidy

Expected result	Illumina NGS platform (different protocols)			Ion Proton NGS platform	SNP Array
	Lab 1	Lab 2	Lab 3	Lab 4	Lab 5
Trisomy 21	Not tested	Trisomy 21* 38% mapped reads	Trisomy 21	Elevated risk for trisomy 21	Not tested
Trisomy 18	Not tested	Trisomy 18* 22% mapped reads WISECONDOR-insufficient reads	Trisomy 18	Elevated risk for trisomy 18	Not tested
Trisomy 13	Not tested	Trisomy 13* 38% mapped reads	Trisomy 13	Elevated risk for trisomy 13	Not tested
Euploidy	Not tested	No trisomies* 29% mapped reads WISECONDOR-insufficient reads	Trisomy 13	Normal	Not tested

* poor quality, too much adapter

CEQAS Cytogenetic External Quality Assessment Service

EMQN The European Molecular Genetics Quality Network

UK NEQAS Molecular Genetics

STAGE 3 – Exploratory pilot (5 clinical laboratories)

Samples	Methods	Testing
Artificial reference material B	WGS Illumina (multiple) Ion Proton	Trisomy 21 Fetal fraction ranges

Expected result	Illumina NGS platform (different protocols)			Ion Proton NGS platform	SNP Array
	Lab 1	Lab 2	Lab 3	Lab 4	Lab 5
Trisomy 21 8% FF	Not Tested	Trisomy 21 29% mapped reads	Trisomy 21	Elevated risk for trisomy 21	Not Tested
Trisomy 21 4% FF	Not Tested	Trisomy 21 37% mapped reads	Trisomy 21	Elevated risk for trisomy 21 (low FF: 3.7%)	Not Tested
Trisomy 21 2% FF	Not Tested	No trisomies* 32% mapped reads	Trisomy 13 Trisomy 21	Elevated risk for trisomy 21 (low FF: 1.8%)	Not Tested
Trisomy 21 1% FF	Not Tested	No trisomies* 34% mapped reads	Equivocal result chromosomes 13 & 21 No evidence of trisomy 18	Elevated risk for trisomy 21	Not Tested

*WISECONDOR- insufficient reads
Analysis repeated with less adapter gives normal result but insufficient reads

CEQAS Cytogenetic External Quality Assessment Service

EMQN The European Molecular Genetics Quality Network

UK NEQAS Molecular Genetics

STAGE 4 – Delivery of EQA pilot NIPT for aneuploidies

Pilot 1

- Delivered 2017
- Artificial material
- Limited to NGS
- 41 labs participated

- Assessment underway
- Only genotyping/interpretation comments
- 3 genotyping errors
- Results issued mid-June 2017

Pilot 2

- Sourcing patient plasma
- Open to all methodologies

- Registration following close of run 1
- EQA distribution – Autumn 2017

Oral presentation at ISPD 2017, San Diego, USA

Measuring the quality of NIPT for aneuploidies – results from the first pilot EQA

Zandra C Deans, Farrah Khawaja, Ros Hastings, Katrina Rack, Simon Patton, Weronika Gutowska-Ding, Stephanie Allen, Lucy Jenkins, Lyn S Chitty, Erik Sistermans

UK NEQAS
Molecular Genetics

Acknowledgements

- *Rapid Prenatal EQA Specialist Advisory Group*
- *Validating laboratories*
- *Peer assessors*
- *CEQAS, EMQN and UK NEQAS for Molecular Genetics teams*

The laboratories

Contact us on info@ukneqas-molgen.org.uk

sera care™

Reliable Accurate Prenatal non-Invasive Diagnosis RAPID

CEQAS Cytogenetic External Quality Assessment Service

EMQN The European Molecular Genetics Quality Network

UK NEQAS Molecular Genetics