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Title: Primary Sample Manual – Clinical Genetics

Changes made since previous version: *Updated sample rejection criteria for samples received from women undergoing immunotherapy treatment. Updated details on fetal sex results for PNS3 test.*

Note*: Please refer to the document record on Ideagen Quality Management (IQM) / Q-Pulse for the revision history of this document.*

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INTRODUCTION

This document contains the details about the requirements for processing patient samples for the Prenatal Safe Non-invasive Prenatal Test (NIPT) in the Eurofins Clinical Genetics Laboratory, Trinity Translational Medicine Institute (TTMI), St. James's Hospital Dublin.

Eurofins Genoma Italy provide additional genetic testing. Please contact our Client Services department on Free Phone 1800 252 966 or 01 295 8545, or e-mail clientservices@ctie.eurofinseu.com for more information.

If you cannot find details of a test you require, please contact our Client Services department on Free Phone 1800 252 966 or 01 295 8545, or e-mail clientservices@ctie.eurofinseu.com.

For sample collection, please contact our Logistics department on Free Phone 1800 252 967, or e-mail logistics@ctie.eurofinseu.com.

TEST INFORMATION TEMPLATE	
Brief information on clinical background, indications for test and interpretation of test results.	
Preparation of Patient: any special preparation required, such as fasting. **Precautions:** any special circumstances, conditions etc. to be aware of.	
Accredited	Whether or not the test is accredited by INAB to ISO 15189. If the test is accredited (Yes), this section is colour-coded in green; if the test is not accredited (No), this section is colour-coded in orange.
Method	Test method. Standard Operating Procedure reference for this test.
Test Options	What each test option offers.
Sample Requirements	Type of tube required, transport temperature and other information.
Turnaround Time	The maximum turnaround time in working days from receipt of the sample in the Eurofins Clinical Genetics Laboratory (TTMI, St James's Hospital Dublin) to the authorisation of the result. **Working days are Monday to Friday 09:00 am to 17:00 pm.**
Stability	Sample stability under various conditions. Please see **SAMPLE STABILITY** notes below.
Storage	Length of time samples are to be stored for.
Units - Reference Ranges and Source	Units and reference range(s) for the test. Source of the reference ranges: 1. Test manufacturer's instructions for use (IFU).

NOTES ON SAMPLE STABILITY

Most incorrect laboratory test results are due to improper sample collection and transport. For details regarding correct phlebotomy technique and our patient identification requirements, please click here.

In order to organise and properly time phlebotomy and sample collection, we have indicated, for each test, its stability after collection.

Stability data are taken from the kit insert for the test, referenced below.

REFERENCE

1. Illumina VeriSeq NIPT Solution v2 Document # 1000000078751 v09.

REASONS FOR REJECTION OF SAMPLES / NON-REPORTING OF TESTS

Every effort will be made to process the samples received in the Eurofins Clinical Genetics laboratory; however, samples may be rejected based on the following criteria:

1. Samples received beyond the stability limits and/or not at the correct temperature indicated in the table below for each test.
2. Samples received in expired Streck Blood Collection tubes.
3. Samples received in leaking Streck Blood Collection tubes.
4. Samples with inconsistent or insufficient patient identifiers. At least 2 unique patient identifiers should match on the test request form and the sample tube. The following criteria will be used to reject specimens as applicable:
    a. The patient identifiers on the sample tube do not match those on the request form.
    b. No patient identifiers on the sample tube or illegible patient identifiers.
    c. Required patient details have not been entered on the request form.
5. Samples received in the incorrect tube (for NIPT, only Streck Blood Collection tubes should be used. Please refer to table below.
6. Samples received with less than the minimum volume (7 ml) as indicated in table below.
7. Lipaemic samples.

8. Heavily haemolysed samples.

9. Clotted samples.

10. Samples taken from women receiving immunotherapy treatment will be rejected if the sample is taken less than 48 hours after the immunotherapy drug treatment (for any queries please contact client services).

11. Further factors that may become apparent only after sample processing begins:

    a. Plasma layer not more than 1.5 ml above the buffy coat layer.

    b. No clear separation of plasma and red blood cells.

NON-INVASIVE PRENATAL TESTING (NIPT)

Fetal chromosome abnormalities, specifically aneuploidy, are a common cause of reproductive failure, congenital anomalies, developmental delay, and intellectual disabilities. Aneuploidy affects approximately 1 in 300 live births, with much higher rates associated with miscarriage and stillbirth. There are two types of prenatal tests available for identifying these disorders: diagnostic testing or screening. Diagnostic testing involves invasive procedures such as amniocentesis or chorionic villus sampling. These testing methods are considered the gold standard for detection of fetal aneuploidy. However, they are associated with a risk of pregnancy loss between 0.11% and 0.22%.

VeriSeq NIPT Solution v2 is an *in vitro* diagnostic test intended for use as a screening test for the detection of fetal genetic anomalies in maternal peripheral whole blood specimens from pregnant women of at least 10 weeks gestation. This test is a screening test and does not replace diagnostic confirmatory tests, e.g. fetal karyotyping.

A negative result does not fully exclude the possibility for the foetus to be affected. The Limit of Detection of the method is at a fetal fraction greater than or equal to 2% (Pertile *et al.*, 2021 PMID: 34077512). If the fetal fraction is not sufficient or the data obtained do not allow a univocal interpretation, a new sample will be requested to repeat the analysis.

VeriSeq NIPT Solution v2 employs whole genome sequencing of cell-free DNA (cfDNA) to detect aneuploidy status of chromosomes 21, 18, 13 and the sex chromosomes with a high degree of accuracy. A recent meta-analysis of multiple clinical studies reported the weighted pooled detection rates and specificities for trisomy 21 and trisomy 18 in singleton pregnancies as follows: trisomy 21 99.7% and 99.96% and trisomy 18 97.9% and 99.96%, respectively. One study indicated that use of NIPT as a primary screen across all pregnancies could result in an 89% reduction in the number of confirmatory invasive procedures.

Preparation of patient: There is no physical preparation for NIPT.

Precautions: The VeriSeq NIPT Solution v2 is a non-invasive in vitro diagnostic test that utilizes whole-genome sequencing of cfDNA fragments derived from maternal peripheral whole blood samples from pregnant women of at least ***10 weeks gestation.***

Accredited	Yes
Method	Next Generation Sequencing. SOPs: NGS02, NGS03, NGS05, NGS08
Test Options	**Prenatalsafe 3:** Trisomy 21 (Down Syndrome), Trisomy 18 (Edwards Syndrome), Trisomy 13 (Patau Syndrome); Fetal Sex (as requested). **Prenatalsafe 5:** Trisomy 21, Trisomy 18, Trisomy 13; Sex Chromosome Aneuploidies XXY, XXX, XYY, XO. Fetal Sex (as requested). **NB: Prenatalsafe 5 cannot be performed for twin pregnancies and vanishing twins. Prenatalsafe3 will be offered instead. For PNS3 test with fetal sex result required, biological sex result as determined by presence or absence of Y chromosome will be provided.**

Sample Requirements	7–10 ml of maternal peripheral whole blood in a Streck cell-free DNA Blood Collection Tube (BCT), which prevents cell lysis and genomic contamination and stabilizes whole blood. **NB:** A minimum volume of 7 ml of sample is required. Please check for expiry date on Streck tubes before blood draw.
Turnaround Time	5 working days from reception of samples at the Eurofins Clinical Genetics Laboratory, TTMI, St James Hospital Dublin.
Stability	Transport: store at temperatures between 4°C and 30°C. Sample storage: stored at 2-8°C prior to testing for up to 10 days. Post-processing, samples stored at 2-8°C (up to a total of 10 days after blood collection). **NB: SAMPLES MUST BE RECEIVED AT THE CLINICAL GENETICS LABORATORY WITHIN 5 DAYS OF THE BLOOD DRAW. IDEALLY SAMPLES SHOULD BE SENT TO THE LABORATORY ON THE DAY OF BLOOD DRAW.**
Units - Reference Ranges	**Autosomal Aneuploidies:** Trisomy 13: No Anomaly Detected, Anomaly Detected Trisomy 18: No Anomaly Detected, Anomaly Detected Trisomy 21: No Anomaly Detected, Anomaly Detected **Sex Chromosome aneuploidies**: No Anomaly Detected Anomaly Detected: XXY Anomaly Detected: XXX Anomaly Detected: XYY Anomaly Detected: XO **Fetal Sex** (singleton pregnancy): Male / Female **Fetal Sex** (Twin): CHR Y Present / CHR Y Not Present

Consultant Responsible: Dr Laura Gigante

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