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Testing performed at the above address only

DETAIL OF ACCREDITATION

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS Amniocytes-cultured Amniotic fluid Whole Blood Whole Blood components (Plasma/buffy coat etc) Chorionic villus biopsy Saliva Bone Marrow Products of conception Fresh tissue biopsy Frozen tissue biopsy Buccal scrape Bronchial brushings Dried blood spots (Guthrie cards) FFPE (Blocks, scrolls and mounted slides) Cultured cells Lymphoblast cells Pleural effusion Cytology fluid, blocks and mounted slides	<u>Molecular genetic examinations for the purpose of clinical diagnosis (including predictive and prenatal) for a range of disorders</u>	In house documented methods incorporating manufacturer's instructions where relevant Automated DNA extraction using: Chemagic Star; Chemagic Star DNA 10K kit Maxwell RSC AS1550 – Maxwell® FSC DNA IQ Casework Kit DC6745 – Casework Extraction Kit (For use with AS1550) AS1400 – Maxwell® RSC Blood DNA Kit AS1720 – Maxwell® RSC FFPE Plus DNA Kit AS1440 – Maxwell® RSC RNA FFPE Kit AS1380 – Maxwell® RSC simplyRNA Blood Kit DNA Purification kit Maxwell® 16 LEV RNA FFPE kit DNA IQ ™ Casework Pro kit for Maxwell® 16 Casework Extraction kit <u>SOPs:</u> LP-GEN-QF1SmpPrp LP-MGN-Shire LP-GEN-SafetyMan LP-GEN-ExtChem LP-GEN-EZ1Extrct LP-GEN-FFPEProc LP-GEN-Maxwell16 LP-GEN-CFExt

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)	In house documented methods incorporating manufacturer's instructions where relevant
Blood components (plasma).		Cell free DNA Extraction using QIAamp Circulating Nucleic Acid kit Measurement Principle*:* Successful Fractioning/isolation of circulating cell free tumour DNA from blood plasma QC- Real-time PCR assay or "blind" SOPs LP-GEN-ExtChem LP-GEN-FFPEProc LP-GEN-Maxwell16 LP-GEN-CFExt
Peripheral Blood		RNA Extraction and cDNA preparation using: Nanodrop 8000 Superscript II reverse transcriptase system (Invitrogen) Measurement Principle: TBP end point PCR assay SOPs LP-GEN-ExtChem LP-GEN-Maxwell16 LP-GEN-CFExt

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)	In house documented methods incorporating manufacturer's instructions where relevant
DNA	Bisulphite conversion for detection of methylation in FFPE tissue: MLH1 promoter methylation	Key Equipment: QIAGEN EpiTect Bisulfite Kit Principle: Bisulphite conversion of C bases to U (Uridine) except for methylated C's that are part of CpG islands; SOPs LF-GEN-BisPyroBk FFPEProc LP-GEN-Maxwell16 LP-GEN-CFExt
DNA cDNA Bisulphite converted DNA	Gene screening services for: - TSC1/2 familial/confirmation mutation testing - ARX - DCTN1 - FOXG1 - MECP2 - TUBA1A - TUBB2B Various custom designed familial mutation analyses for genes not covered by laboratory's current screening repertoire, typically genes with variants identified in research labs and requiring confirmation/family follow-up under diagnostic conditions	Manual PCR using: Custom primers, Thermal cyclers: 2700, , 9700, PrimeG, BioRad C1000, Biorad S1000, Biorad T100 Gel electrophoresis ChemiDoc XRS+ system, 96 well Gel tanks (alpha labs), BioRad powerpac 300 Measurement Principle Targeted amplification of regions of interest. QC – gel electrophoresis for assessment of PCR product strength and conformity SOP LP-GEN-PCR LP-GEN-KnownMut

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	<u>Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)</u>	In house documented methods incorporating manufacturer's instructions where relevant
DNA		Amplification of DNA for Gene screening services using Automated PCR using: Custom primer plates SHIRE – AutoPCR module (in-house formatter and compiler) NXmaker.xls data output for Hamilton STARlets (in-house design) Thermal cyclers: 2700, 9700, PrimeG, BioRad C1000, Biorad S1000, Biorad T100 <u>SOP</u> LP-GEN-PCR LP-GEN-AutoPCR LP-GEN-Sequencing LP-GEN-Pyrosequencing
PCR products	Detection and quantification of specific variants: • LHON • m.A1555G • LRRK2 • NRAS • KRAS • BRAF • MLH1 promoter methylation •	Pyrosequencing using: PyroMark Assay Design software Qiagen Autoprep Q48 AQ pyrogram report CG pyrogram report <u>Measurement Principle</u> Target fluorescent, quantitative sequencing by synthesis <u>SOPs</u> LP-GEN-Pyrosequencing LP-GEN-Pyro checking LP-GEN-LHON LP-GEN-mA1555G LP-GEN-LRRK2 LP-GEN-NRAS LP-GEN-KRAS LP-GEN-BRAF LP-GEN-Lynch

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	<u>Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)</u>	In house documented methods incorporating manufacturer's instructions where relevant
PCR products	Detection and quantification of specific variants: • Hereditary Deafness: Del *GJB6*-D13S1830 and Del *GJB6*- D13S1854 • JAK2 • F8 Intron 1	Non-fluorescent fragment sizing using: BioRad ChemiDoc XRS+ electrophoresis System <u>Measurement Principle</u> Target amplification of DNA segments containing the breakpoint junction of each deletion, as well as an internal control to check the efficiency of the PCR assay and to distinguish heterozygosity and homozygosity for the deletions <u>SOPs</u> LP-GEN-Connexin LP-GEN-F8Intron1 LP-GEN-DiagGel

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd) PCR products	<u>Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)</u> Whole or partial gene screen analysis for genetic variants causing diseases and disorders: • ARX • FOXG1 • MECP2 • TUBA1A • TUBB2B • DCTN1 • POLE • POLD1	In house documented methods incorporating manufacturer's instructions where relevant Automated Sanger Sequencing <u>Key Equipment</u> Sequence Service Database384v1.1 (Access based, in house formatter and compiler) Hamilton ML Starlet 96 with Venus operating system Applied Biosystems fluorescent capillary electrophoresis system 3730/3730XL Invitrogen E-gel system Thermal cyclers: 2700, , 9700, PrimeG, BioRad C1000, Biorad S1000, Biorad T100 <u>Measurement Principle:</u> Assessment of quality of sequencing based on read length, peak height and definition (peak morphology) <u>SOPs</u> LP-GEN-SangSeqCh LP-GEN-Sequencing - Sequencing and Genotyping LP-GEN-POLEPOLD1 – POLE/POLD1 – associated cancer susceptibility.

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd) PCR products (cont'd)	<u>Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)</u> Whole gene screen analysis for genetic variants causing diseases and disorders: (cont'd) • ARX • FOXG1 • MECP2 • TUBA1A • TUBB2B • DCTN1 • F12 • F8 • VWD	In house documented methods incorporating manufacturer's instructions where relevant Manual Sanger sequencing using: Ampure and CleanSeq Beckman Applied Biosystems fluorescent capillary electrophoresis system 3730/3730XL Thermal cyclers: 2700, 9700, PrimeG, BioRad C1000, Biorad S1000, Biorad T100 <u>Measurement Principle:</u> Assessment of quality of sequencing based on read length, peak height and definition (peak morphology) <u>SOPs</u> LP-GEN-Sequencing LP-GEN-ARX LP-GEN-FOXG1 LP-GEN-Rett LP-GEN-TUBA1A LP-GEN-TUBB2B LF-GEN-ARX2.1back LF-GEN-ARX2.2/3back LF-GEN-FOXG1_1A1/A2back LP-GEN-FI2 LP-GEN-F8 LP-GEN-CMBVWD LP-GEN-STXBP1 LP-GEN-TCF4 LP-GEN-HKPX LP-GEN-Tubulin LP-GEN-ACTB/G1 LP-GEN-CDKL5 LP-GEN-DCX LP-GEN-GPR56 LP-GEN-LIS1 LP-GEN-LynFrynsSyn LP-GEN-MECP2 LP-GEN-DCDH19 LP-GEN-SLC9A6

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd) PCR products (cont'd)	<u>Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)</u> Familial mutation analysis for clinically significant changes and variants of unknown significance in all of the following: • Cortical Brain Malformation Disorders:- *LIS1*, *DCX, ARX*, *TUBA1A*, *ACTB /ACTG1*, *GPR56*, *TUBB2B* and *CCND2* • Familial Breast Cancer: - BRCA1/BRCA2 • Autosomal Recessive polycystic kidney disease (ARPKD):- PKHD1 • Early Infantile Epilepsy Conditions: - *ARX, CDKL5, STXBP1, PCDH19 and MEF2C* • Pitt-Hopkins Syndrome (PTHS):- TCF4 • Rett Syndrome (RTT):- MECP2 • Congenital Variant of Rett:- FOXG1 • Hereditary deafness: - GJB2 • Ectodermal dysplasias:- EDA, EDAR, EDARRAD, WNT10A, GJB6, MSX1, TP63 and *IKBKG(NEMO)* • Familial Adenomatous Polyposis:- APC • *MUTYH*-associated Polyposis (MAP; MYH): - MUTYH • Familial Hypercholesterolaemia**:-** *LDLR, APOB(exon26)* and *PCSK9(*exon 7)	In house documented methods incorporating manufacturer's instructions where relevant Using Automated PCR, Automated Sanger sequencing, and (where applicable) Multiplex Ligation-dependant Probe Amplification (MLPA) <u>Key Equipment</u> Custom primer sets Commercial MLPA kits Mutation Surveyor software ENSEMBL/NCBI RefSeqGene or LRG's Hamilton ML Starlet 96 with Venus operating system <u>Measurement principle</u> Sequence output is compared with RefSeqGene or LRG sequence either in an automated manner or manually and is assessed for quality by the purity of the sequence data. Sequence changes are then interpreted against the associated polymorphism list, published data and in the context of the gene disorder and/or family. <u>SOPs</u> LP-GEN-Sequencing LF-GEN-UV LF-GEN-UVsup LP-GEN-ACTB/G1 LP-GEN-ARPKD LP-GEN-ARX LP-GEN-BRCA LP-GEN-CDKL5 LP-GEN-Connexin LP-GEN-DCX LP-GEN-ED GJB6 LP-GEN-EDA LP-GEN-EDAR LP-GEN-EDARADD

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd) PCR products (cont'd)	<u>Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)</u> Familial mutation analysis for clinically significant changes and variants of unknown significance in all of the following (cont'd): • Hyperekplexia (HKPX):- *GLRA1, GLRB* and *SLC6A5* • Lynch syndrome / Hereditary Non-polyposis Colorectal Cancer (HNPCC):- *MLH1, MSH2* and *MSH6* ○ *MLH1, MSH2* for DNA from FFPE • Neurofibromatosis Type 1 (NF1):- NF1 • Legius Syndrome (NFLS, Neurofibromatosis Type 1-like Syndrome):- SPRED1 • Optic Atrophy: - OPA1 and OPA3 • Wagner Syndrome 1 (WGN1): - VCAN (inton 7/exon 8, exon 8/intron 8 splice sites) • Parkinson's Disease (PD):- *PARK2, PINK1* and *DJ1* • Perry Syndrome:- *DCTN1* • X-linked Angelman-like Syndrome (Christianson Syndrome):- *SLC9A6* Porphyrias: - HMBS, PPOX, CPOX, FECH, *UROS* , *UROD,* • *ALAS2, XLDPP* • Nephrogenic Diabetes insipidus:- AQP2, AVPR2 • Thyroid dysfunction:– *TSHR*	In house documented methods incorporating manufacturer's instructions where relevant <u>Disorder Specific</u> LP-GEN-FAP LP-GEN-FOXG1 LP-GEN-FH LP-GEN-GIST LP-GEN-GPR56 LP-GEN-Lynch LP-GEN-Legius LP-GEN-LIS1 LP-GEN-MEF2C LP-GEN-MSX1 LP-GEN-MUTYH LP-GEN-NF1 LP-GEN-OPA1 LP-GEN-Park2 LP-GEN-PCDH19 LP-GEN-Perry LP-GEN-Rett LP-GEN-SLC9A6 LP-GEN-STXBP1 LP-GEN-TCF4 LP-GEN-TP63 LP-GEN-TUBA1A LP-GEN-TUBB2B LP-GEN-VCAN LP-GEN-WNT10A LP-GEN-HMBS LP-GEN-PPOX LP-GEN-UROS LP-GEN-UROD LP-GEN-CPOX LP-GEN-FECH LP-GEN-AVPR2 LP-GEN-AQP2 LP-GEN-TSHR

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd) DNA	<u>Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)</u> Panel for • BRCA1/BRCA2 • TSC • FAP • Lynch • MUTYH	In house documented methods incorporating manufacturer's instructions where relevant <u>Key Equipment</u> Hybex Qubit PCR MixMate MiSeq HiSeq SpeedVac Bioinformatics pipeline Next Generation Sequencing Library Preparation – Targeted/Custom and commercial NGS panels LP-GEN-ExtChem LP-GEN-Maxwell16 LP-GEN-CFExt LP-GEN-BRCA
Cell free DNA	Identification of • EFGR mutations: L858R, T790M and exon 19 deletions • Systemic Mastocytosis (CKIT) • Waldenstrom macro gloubinaemia (MYD88) • KRAS • NRAS	Droplet digital PCR (ddPCR) <u>Measurement principle</u> Define the Rain online software used to determine thresholds for each individual ddPCR assay. QuantaSoft software used to analyse data. <u>Key kits</u> KRAS multiplex kit, KRAS Q61 multiplex kit, NRAS Q61 multiplex kit <u>*SOPs*</u> LP-GEN-ddPCR LP-GEN-EGFR

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	<u>Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)</u>	In house documented methods incorporating manufacturer's instructions where relevant
DNA	Detection of whole or partial gene deletions and duplications and probe specific mutations of the following genes/disease areas: • DMD/BMD • SMA • CMT/HNPP • Lubs X-linked Mental Retardation Syndrome (MRXSL)	Multiplex Ligation-dependant Probe Amplification (MLPA) method using : <u>Key Equipment</u> Genotyping xls output (in house design) Applied Biosystems fluorescent capillary electrophoresis system 3730/3730XL GeneMapper software GeneMarker software Thermal cyclers: 2700, , 9700, PrimeG, BioRad C1000, Biorad S1000, Biorad T100 *Commercial kits:* MRC-Holland <u>Measurement Principle</u> Targeted specific probe ligation followed by amplification of ligated products and Dosage Quotient (DQ) analysis for the determination of larger exon/multi-exon deletions and duplications <u>SOPs</u> LP-GEN-Sequencing LP-GEN-MLPA LP-MGN-DMDBMD, LP-GEN-SMA LP-GEN-CMT1A LP-GEN-Lynch

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd) Extracted DNA from: Peripheral blood, Products of Conception	<u>Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)</u> Detection of chromosome abnormalities associated with: Postnatal disorders including Abnormal Adult (Behavioural) Abnormal Adult (Learning disability) Abnormal Adult (Short stature) Abnormal Child (Dysmorphism) Abnormal Child (Developmental Delay) Abnormal Child (Congenital Abnormality) Abnormal Child (intellectual disability) Reproductive disorders including Foetal Wastage (Recurrent Miscarriage) Foetal Wastage (Neonatal death) Foetal Wastage (Abnormal Foetus) Follow up testing of prenatal karyotypic abnormalities	In house documented methods incorporating manufacturer's instructions where relevant Array Comparative Genome Hybridisation analysis (aCGH) using: <u>Key Equipment</u> Thermal cyclers: 2700, 9700, PrimeG, BioRad C1000, Biorad S1000, Biorad T100 Agilent scanner Nanodrop/Qubit Speed/Vac Concentrator Hybridisation oven <u>Measurement Principle:</u> Microarray whole genome profile using Oxford Gene Technology platform technology. Analysis and reporting of copy number imbalances against a reference, in line with ACC Constitutional Array CGH Best Practice Guidelines (2011) v2.00 and RCOG prenatal guidelines 2011 <u>SOPs:</u> LP-GEN-AC1SetUp LP-GEN-AC2ScnDataH LP-GEN-AC3AnInt

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)	In house documented methods incorporating manufacturer's instructions where relevant
PCR products	Molecular Rapid Aneuploidy testing	Fragment sizing: Fluorescent QF-PCR Key Equipment Genotypingservice.xls output (in house design) Applied Biosystems fluorescent capillary electrophoresis system 3730/3730XL GeneMapper software GeneMarker software Peak Scanner software Thermal cyclers: 2700, , 9700, PrimeG, BioRad C1000, Biorad S1000, Biorad T100 Measurement Principle Capillary electrophoresis of QF-PCR derived DNA fragments and Semi-automated analysis using ABI Genemapper software to facilitate the calculation of dosage quotients using Microsoft Excel macro driven work sheets, in accordance with ACGS Best Practice Guidelines for QF-PCR for the diagnosis of aneuploidy (2018) V4 SOPs: QF-PCR Samples Manual Sections 1 to 8: [LP-GEN-QF1SmpPrp], [LP-GEN-QF2SetUp], [LP-GEN-QF3AnIntn], [LP-GEN-QF4ChkRep], [LP-GEN-QF5PCRkit], [LP-GEN-QF6Qualit], [LP-GEN-QFTrbShoot]

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	<u>Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)</u>	In house documented methods incorporating manufacturer's instructions where relevant
PCR products (cont'd)	Disorder/test provision • Exclusion of Maternal Cell contamination • Foetal sexing • Molecular tissue/tumour identity services	Measurement Principle: Capillary electrophoresis of QF-PCR derived DNA fragments followed by comparative analysis to DNA from either familial (maternal), standard male/female or normal/reference tissue sample respectively SOPs LP-MGN-MCC QF-PCR Samples Manual Sections 1 to 7: [LP-GEN-QF1SmpPrp], [LP-GEN-QF2SetUp], [LP-GEN-QF3AnIntn], [LP-GEN-QF4ChkRep], [LP-GEN-QF5PCRkit], [LP-GEN-QF6Qualit], [LP-GEN-QFTrbShoot]

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	<u>Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)</u>	In house documented methods incorporating manufacturer's instructions where relevant
PCR products (cont'd)	Disorder/Test provision (cont'd) • CALR • Nucleotide repeat disorders Fragile X (FMR1&FMR2) SCA1,2,3,6,7&17 FRDA DM1&DM2 C9ORF72 ARX HD α and β globin gap PCRs	Measurement Principle: Targeted fragment size analysis; separation by capillary electrophoresis and translation to diagnostic reference ranges versus known controls and in accordance with current disorder specific best practice guidelines (where available). SOPs LP-GEN-JAK2CALR LP-GEN-SCAs LP-GEN-FRDA LP-GEN-DM1 LP-GEN-DM2 LP-GEN-C9ORF72 LP-GEN-ARX LP-GEN-HD LP-GEN-FragileX LP-GEN-Amplidex
PCR products	Linkage analyses: ARPKD (PKHD1)	Measurement Principle Fluorescent targeted fragment size analysis within a family group (minimum affected, Father, Mother), separation by capillary electrophoresis and manual translation to familial haplotype(s). SOPs LP-GEN-ARPKD

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)	In house documented methods incorporating manufacturer's instructions where relevant
PCR products (cont'd)	Microsatellite instability analysis for: Lynch Syndrome Somatic colorectal tumours	Commercial kit: Promega MSI analysis System; Measurement Principle Measure of instability of 5 quasi-monomorphic mononucleotide repeat markers Gene Marker Software SOP LP-GEN-Lynch LF-GEN-MSIwksbk LP-GEN-MicroInst
DNA	Real-time allelic discrimination of • APOE • CF • MYH • HFE • FVL • MTHFR	Key Equipment Applied Biosystems 7500 Fast Real Time PCR System StepOne Plus Instrument and analysis software Measurement Principle; TaqMan PCR; allelic discrimination (Mutant: Normal primer specific ratio) SOPs LP-GEN-PCR LP-GEN-APOE LP-GEN-CF LP-GEN-MUTYH LP-GEN-RealTPCR

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd) DNA (cont'd)	<u>Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)</u> Quantitative aCGH follow up: Confirmation of dosage imbalance.	In house documented methods incorporating manufacturer's instructions where relevant Key Equipment StepOne Plus Instrument and analysis software Measurement Principle: Quantitative detection of nucleic acid sequences using the comparative C_T method and analysis using the StepOne Plus Instrument and analysis software to facilitate the calculation of dosage quotients which are used to determine copy number of the targets being tested. Key SOPs: LP-GEN-qPCRSetUp LP-GEN-qPCRAnalysis LP-GEN-qPCRChk&Auth LP-GEN-qPCRWS LP-GEN-qPCRRprtg

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)	In house documented methods incorporating manufacturer's instructions where relevant
DNA	Diagnosis of Fragile X	Key equipment Applied Biosystems fluorescent capillary electrophoresis system 3730/3730XL GeneMapper software GeneMarker software Thermal cyclers: 2700, , 9700, PrimeG, BioRad C1000, Biorad S1000, Biorad T100 Commercial kit: AmplideX FMR1 PCR Kit from Asuragen; Measurement Principle: amplification and detection of the CGG repeat region in the 5' untranslated region of the FMR1 gene SOP LP-GEN-AmplideX
Cell free DNA from maternal circulation, extracted in-house	Trisomies 13, 18 and 21, and sex chromosome aneuploidy in single pregnancies	Key equipment: Illumina TruSeq Nano DNA Sample prep Kits Illumina cfDNA assay on the Hiseq. VeriSeq NIPT analysis software system using NIPT-16 assay. Measurement Principle: Testing of foetal blood from maternal circulation for the common chromosmal trisomies using next generation sequencing SOP LP-GEN-NIPTAnalysis LP-GEN-NIPTLib LP-GEN-NIPTcfExt

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	<u>Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)</u>	In house documented methods incorporating manufacturer's instructions where relevant
DNA	Gene variants associated with Ectodermal Dysplasia Ehlers Danlos Aortopathy & Marfans Optic Atrophy Primary immunodeficiency	Next generation sequencing: Trusight One NGS Panel test Automated set-up using Hamilton ML Starlet 96 with Venus operating system Key equipment HiSeq Germlineenrichment software Thermal cycler Plate shaker Hybex Qubit Tapestation 4200 Hot block Commercial kit: Trusight Rapid Capture SOP LP-GEN-TruSightONE LP-GEN-TSOViPaAnl LP-GEN-VarInvestigation LP-GEN-NGSAutoTru, LP-GEN-NGSAuto; validation doc MP-GEN-ValdVerifNGSSTAR

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)	In house documented methods incorporating manufacturer's instructions where relevant
DNA	Gene variants indicating a pre-disposition to breast, colorectal and rare cancers	Next generation sequencing using TruSight Cancer Panel (validated for >1700 exons 94 genes). Automated set-up using Hamilton ML Starlet 96 with Venus operating system Key equipment NextSeq In-house developed software for data analysis Thermal cycler Plate shaker Hybex Qubit Tapestation 4200 Hot block Commercial kit: Trusight Rapid Capture SOP LP-GEN-TruSightCan
	Myeloid malignancies Chronic lymphocytic leukaemia Myeloproliferative disorders	Next Generation Sequencing using Trusight Myeloid Panel Key equipment MiSeq, Thermal cycler, Plate shaker, Hybex, Qubit, Hot block, Tapestation 4200 SOP LP-GEN-MyeloidNGS - Myeloid TruSight Panel LP-GEN-HaemNGS - Haematological NGS panel (Workflow and Analysis)

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)	In house documented methods incorporating manufacturer's instructions where relevant
	Familial Hypercholesterolaemia	SureSeq NGS Custom FH Panel - Illumina Key Equipment as above with Covaris ME220 SOP LP-GEN-FHNGSlibPrep - SureSeq NGS Library Preparation For Familial Hypercholesterolaemia (FH) panel LP-GEN-FH - Familial Hypercholesterolaemia.
	Pan-Cancer	Roche Pan Cancer Panel – Key equipment – NextSeq -Illumina SOP Custom desined Pan-Cancer Panel - Roche NextSeq – Illumina LP-GEN-PanCanLibPrp - Pan-Cancer NGS Library Preparation LP-GEN-PanCanAnlyss - Solid Tumour NGS Panel Pan-Cancer Panel)Workflow and Analysis)

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)	In house documented methods incorporating manufacturer's instructions where relevant
DNA	Glioma service: Pyrosequencing of mutation hotspots in IDH1, IDH2, or BRAF, MGMT methylation pyrosequencing and/or 1p/19q FISH	Pyrosequencing using: PyroMark Assay Design software Qiagen's Autoprep Q48 instrument AQ pyrogram report CG pyrogram report Fluorescence in situ hybridisation (FISH) using: Standard light microscopes from Zeiss Fluorescence microscopes from Zeiss Slide denaturation system x 2: Abbott Statspin Thermobrite, MRC Scientific Denaturation and Hybridisation Key equipment: Thermal cyclers: 2700, , 9700, PrimeG, BioRad C1000, Biorad S1000, Biorad T100 Mini plate shaker SOP LP-GEN-Glioma LP-GEN-Pyro checking LP-GEN-Pyrosequencing

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	<u>Molecular genetic examinations for the purpose of clinical diagnosis (cont'd)</u>	In house documented methods incorporating manufacturer's instructions where relevant
DNA from whole blood	Whole genome sequencing for analysis of single nucleotide variants including indels	Equipment: Maxwell: Chemagic NanoDrop Qubit: - Tapestation 4200 Bioanalyser NovaSeq 6000 Dragen: Documentation: LP-GEN-SampSort LP-GEN-Maxwell16 LP-GEN-ExtChem LP-GEN-SamplePreprocess LP-GEN-FlexWGS LP-GEN-QubitFlur LP-GEN-TapeStation LP-GEN-NovaSeq LP-GEN-HPCBioinf LP-GEN-AutoQC-SOP LP-GEN-NGSQC LP-GEN-AutoQCSOP LP-GEN-SOPDRAGEN LP-GEN-RWGSAnalysis LP-GEN-VarInvestigation

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd) Amniotic fluid, Chorionic villus, Pleural effusion or other prenatal fluids, Peripheral blood, Bone marrow, Products of conception Skin/tissue biopsy Lymph node biopsy, Tumour material,	Cytogenetic examinations for the purpose of clinical diagnosis (including predictive and prenatal) for a range of disorders	In house documented methods incorporating manufacturer's instructions where relevant Cell culture using: Key Equipment 2x double Napco 6000 (incubators) (nos. 1-4) 1x Thermo Forma Solis 2 (incubator) (no. 8) Inverted microscopes : Olympus CK2 Microscopes from Zeiss Measurement Principle: Initiation of short and long term cell culture and harvesting/fixation to produce interphase or metaphase nuclei for analysis by cytogenetic or molecular cytogenetic techniques, in line with ACGS and ACC Best Practice Guidelines. SOPs:- LP-GEN-BndgStng LP-GEN-SlideMaking LP-GEN-Harvestng LP-GEN-ShTmSetUp LP-GEN-LnTmSetUp

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd) Bone marrow Blood Formalin fixed paraffin embedded tissue lymph node biopsy	Cytogenetic examinations for the purpose of clinical diagnosis (cont'd)	In house documented methods incorporating manufacturer's instructions where relevant Haematological culture and processing by: Cell culture Plasma cell separation Cell counting RNA isolation Key Equipment Thermo Forma Solis 2 incubator no. 8 Inverted microscopes Microscopes (Main Lab) Horiba Medical ABX MicrosES60 cell counter RoboSep cell isolation platform Principle Magnetic cell separation, initiation of direct, short and long term cell culture and harvesting/fixation to produce interphase or metaphase nuclei for analysis by cytogenetic or molecular cytogenetic techniques, in line with ACGS Best Practice Guidelines. SOPs:- LP-GEN-CYTOSCC LI-GEN-LNChkSht LP-GEN-RCLysis LP-GEN-Maxwell16 LP-GEN-Reporting LP-GEN-AnlysChck LP-GEN-BndgStng LP-GEN-SlideMaking LP-GEN-Harvestng LP-GEN-ShTmSetUp

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	Cytogenetic examinations for the purpose of clinical diagnosis (cont'd)	In house documented methods incorporating manufacturer's instructions where relevant
Cultured cells or fixed tissue	Cytogenetic chromosome analysis	Key Equipment Microscopes MetaSystems image analysis system. Measurement Principle Microscopic analysis of metaphase chromosome preparations with reference to standard ISCN 2013. and in accordance with ACGS Best Practice Guidelines (2016) SOPs LP-GEN-Reporting LP-GEN-AnlysChck
Cell suspensions. Tissue sections (FFPE) Blood smears Bone marrow smears Cytospin slides Tumour touch preparations	For diagnosis of the following disorders: Acute Myeloid Leukaemia Acute Lymphoblastic Leukaemia Chronic Myeloid Leukaemia Chronic Lymphocytic Leukaemia Myelodysplastic syndrome Myeloproliferative neoplasms Myeloma Lymphoma	Fluorescence in situ hybridisation (FISH) using Key Equipment Standard light microscopes from Zeiss Fluorescence microscopes from Zeiss Slide denaturation system x 2: Abbott Statspin Thermobrite, MRC Scientific Denaturation and Hybridisation Measurement Principle: Microscopic analysis of fluorescently labelled probes in accordance with ACGS/ACC Best Practice Guidelines and manufacturers guidelines. SOPs LP-GEN-FISHStUpPrc LP-GEN-FISHAnlChk
	Her 2 ROS1	LP-GEN-HER2 - Human Epidermal Growth Factor 2 (HER2). LP-GEN-ROS1FISH – ROS1 Proto-Oncogene receptor tyrosine kinase

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	<u>Cytogenetic examinations for the purpose of clinical diagnosis (cont'd)</u>	In house documented methods incorporating manufacturer's instructions where relevant
DNA	Amplification Refractory Mutation system for diagnosis of Cystic Fibrosis	Key Equipment Applied Biosystems fluorescent capillary electrophoresis system 3730/3730XL GeneMapper software GeneMarker software Thermal cyclers: 2700, 9700, PrimeG, BioRad C1000, Biorad S1000, Biorad T100 *Commercial kit:* Yourgene Health. - Elucigene CF-EU2v1 50 test kit; Measurement Principle Allele specific amplification of 50 common European CFTR mutations SOPs LP-GEN-Elucigene LP-GEN-Sequencing LP-GEN-CF
	Amplification Refractory Mutation system for testing for DYPD	*Commercial kit:* Elucigene ONDYB1DPYD ARMS PDR Kit
Fixed, fresh and frozen tissues; excisional and incisional biopsies, surgical resection and post mortem specimens, body fluids containing cellular material Tissue sections (FFPE)	Fluorescent in-situ hybridisation Sarcoma (EWSR1,SYT(SS18),DDIT3, FOX01, COL1A1-PDGFB, FUS, MDM2, ALK, MYC, NR4A3, PAX-3FOXO1, ETV6, TFE3, CIC). Lung cancer (ALK)	Equipment Dakocytomation hybridizer Gant Boekel Slide warmer Stuart Scientific SI 60 incubator Principle Microscopic analysis of fluorescently labelled probes in accordance with ACGS/ACC Best Practice Guidelines and manufacturers guidelines Key SOPs LP-GEN-FISHAnlChk LP-GEN-FISHStUpPrc

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	Molecular Diagnostics: Leukaemia and Lymphoma examinations for the purpose of clinical diagnosis	In house documented methods incorporating manufacturer's instructions where relevant
Blood/Bone marrow	Molecular screens for the following translocations (provides diagnostic, prognostic and minimal residual disease (MRD) information)	ABI 9700 PCR machine PCR amplicon analysis
	RUNX1-RUNX1T1 CBFB-MYH11 PML-RARA BCR-ABL1 (p190) BCR-ABL1 (p210)	LP-HAE-MLt(8;21) LP-HAE-MLInv16 LP-HAE-ML15;17 LP-HAE-MLp190 LP-HAE-MLp210Diag
Blood/Bone marrow	RQPCR BCR-ABL1 (p210 quantitative)	Applied Biosystems Taqman 7500 Fluorescent PCR amplicon analysis against standard curve Absolute copy number determined via manual procedure SOP LP-HAE-MLp210QPractical
Blood/Bone marrow/Blood and Bone marrow cell fractions	Molecular screens for chimerism following transplant using microsatellite analysis.	ABI 9700 PCR machine, fluorescently labelled PCR amplicon analysis using Applied Biosystems Ampfister Identifier Kit and Genescan 3100, Applied Biosystems fluorescent capillary electrophoresis system 3730/3730XL
Blood/Bone marrow/Fixed Paraffin sections/Fresh tissue	Molecular screens for the detection of suspected lymphoproliferative disorders to confirm clonality	ABI 9700 PCR machine Fluorescently labelled PCR amplicon analysis using Invivoscribe Biomed 2 primer sets and auto-clonality setup (using Hamilton STARlet instruments) and Applied Biosystems fluorescent capillary electrophoresis system 3730/3730XL

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	<u>Molecular Haematology: Red Cells & Iron examinations for the purpose of clinical diagnosis</u>	In house documented methods incorporating manufacturer's instructions where relevant
Whole blood, bone marrow or buccal smear	HFE genotyping for the variants commonly associated with HFE-related hereditary haemochromatosis in people of North European descent HFE:p.Cys282Tyr HFE:p.His63Asp (NM_000410.3(HFE):c.845G>A & HFE:c.187C>G)	StepOne Plus Instrument and allelic discrimination StepOne analysis using TaqMan probes and fluorescence detection SOP LP-GEN-HFE LP-GEN-REALTPCR
Whole blood, bone marrow or buccal smear		Chemagic STAR semi-automated method using specific binding of DNA from lysed cells by M-PVA magnetic beads
DNA extracted from whole blood	Identification of common single and double alpha globin gene deletions and triplication for the following disorders: Alpha plus thalassaemia of types ($-\alpha^{3.7}$,$-\alpha^{4.2}$) Alpha zero thalassaemia of types (--SEA,--FIL,--THAI,--MEDI,-(α)20.5,--BRIT,--SA), and anti3.7ααα.	Manual GAP-PCR PCR set-up and gel electrophoresis, using Eppendorf Mastercycler and two ABI Veriti Thermocyclers for gene amplification. BioRad and GRI electrophoresis tanks. ChemiDoc XRS+ system, digital camera and thermal print gel documentation. Key SOPs LP-GEN-ExtChem LP-GEN-Maxwell16 LP-HAE-RCUVIDOC LP-GEN-RCHMDL10 LP-GEN-RCHMLL11

Materials/Products tested	Type of test/Properties measured/Range of measurement	Standard specifications/ Equipment/Techniques used
HUMAN TISSUE AND FLUIDS (cont'd)	<u>Molecular Haematology: Red Cells & Iron examinations (cont'd)</u>	In house documented methods incorporating manufacturer's instructions where relevant
DNA extracted from whole blood, bone marrow or buccal smear	Diagnosis and prognosis of the following disorders of red cell/iron metabolism: Dinucleotide (TA) repeat polymorphism in the UGT1-A1 gene promoter G-gamma globin gene promoter region C>T XmnI site polymorphism alpha plus thalassaemia type HBA2:c.95+2_95+6delTGAGG HbD-Punjab (HBB:c.364G>C) and HbO-Arab (HBB:c.364G>A) HbS (HBB:c.20A>T) HFE:c.845G>A & HFE:c.187C>G.	Manual RFLP-PCR PCR set-up and gel electrophoresis, using Eppendorf Mastercycler and two ABI Veriti Thermocyclers for gene amplification. BioRad and GRI electrophoresis tanks. UviDoc UV illuminator, digital camera and thermal print gel documentation. PCR LP-HAE-RCHMDL7 LP-HAE-RCHMDL8,9, 22 LP-GEN-ExtChem LP-GEN-Maxwell16
END		