HISTOPATHOLOGY USER GUIDE

Contents

Overview

Histopathology services are provided by the Christie Pathology Partnership.

The information in this user guide relates to Histopathology Services.

Expert clinical and scientific advice is available on the investigation of patient pathological specimens, the diagnostic report and on any further investigations which may be required.

Authorised results will be available to view on CWP. All request forms should be filled in correctly with all relevant clinical details.

Contact details shown below (during normal working hours):

Contact details of key members of staff

The contact number for the histopathology department is 0161 446 3290.
The full contact details of all laboratory personnel can be found on the staff directory.

NB all telephone numbers should be prefixed with 0161 when calling from outside Manchester

Medical Staff	Name	Telephone	E-mail
Consultant Histopathologist	Dr Sudha Desai	446 3571	sudha.desai1@nhs.net
Consultant Histopathologist	Dr Bipasha Chakrabarty	446 3907	bipasha.chakrabarty@nhs.net
Consultant Histopathologist	Dr Lia Menasce	446 3273	lia.menasce@nhs.net
Consultant Histopathologist	Dr Patrick Shenjere	446 3274	p.shenjere@nhs.net
Consultant Histopathologist	Dr Paula Hyder	446 3292	paula.hyder@nhs.net
Consultant Histopathologist	Dr Pedro Oliviera	956 1161	pedro.oliviera1@nhs.net
Consultant Histopathologist	Dr Anshuman Chaturvedi	446 7071	a.chaturvedi@nhs.net
Consultant Histopathologist	Dr Angelia Ong	446 8533	angelia.ong@nhs.net
Consultant Histopathologist	Dr Sangeeta Verma	446 8025	sangeeta.verma@nhs.net
Consultant Histopathologist	Dr Rola Salama	Ext 1687	rola.salama@nhs.net
Scientific staff			
Histology Service Manager	Ms Catherine Billington	446 3289	catherine.billington3@nhs.net
BTR Clinical Scientist	Mrs Angela Cramer	446 3211	angela.cramer@nhs.net
Histology Office	Emma Herring	446 8540	the-christie.pathology.office@nhs.net

The location of the laboratory

The Histopathology laboratory is situated within the Pathology Department (Department 45) located at the Wilmslow Road end of the 1st floor corridor, above the Wilmslow Road entrance. Follow the silver signage for Pathology. Alternatively, you could download the Christie app for real time navigation on your phone. Search for 'The Christie' on the App Store or Google Play.

Opening Times of the Histopathology laboratory

Please note: There is **NO** out of hour's service for Histopathology. Specimens arriving on Saturday, Sunday or bank holidays will not be processed until the next normal working day

Monday to Friday 07:00 – 17:00	Routine service - Please ensure that samples for routine requests are sent to the laboratory within these hours

Services offered by the Histopathology Laboratory

- Routine Histopathology
- Special Stain Histopathology
- Immuncytochemistry
- Breast and Gastric Receptor staining and analysis

Histopathology offers a diagnostic service for oncology surgical specimens taken in Theatre at The Christie with a frozen section service provided during routine hours. The department also processes peritoneal washings for Gynae specimens, so that they can be reported in conjunction with the surgical specimen. Histopathology also provides a review and opinion service for surgical samples taken at external hospitals.

MDT review of Christie patients could cause the pathologist to return to any previous histology cases and request extra tests to be carried out, which may not have been indicated either by the clinical status of the patient or that the test may not have been available at that time. As Histology blocks are stable in the longer term, this allows further testing to be carried out.

The laboratory carries out H&E staining, a variety of special stains, a limited number of in-situ hybridisation probe staining and a large repertoire of immunocytochemistry staining of the prepared surgical specimens. The current test repertoire of the Histopathology Department can be found in the Schedule of Accreditation issued by the United Kingdom Accreditation Service (UKAS) and can be accessed via the following link: 8697 Medical Single (ukas.com).

Due to the complex nature of the surgical samples and a move towards personalised medicine, the department may need to send material away for further molecular or genetic testing.

The department also provides a Breast and Gastric receptor analysis service for external laboratories, which refer their work to the department. The Breast Tumour Receptor service performs immunohistochemistry (IHC) and analysis on paraffin embedded tissues for breast and gastric receptors, including HER2, Oestrogen Receptors (ER), Progesterone Receptors (PR), Ki67, PD-L1 22C3 and Androgen Receptor (AR). The Breast Tumour Receptor Service also offers non-routine testing by special request for trials and other research

HER2

- HER2 IHC cases showing >10% strong, complete membrane staining of invasive cancer cells are regarded as HER2 positive.
- Less than 10% of cells with strong complete membrane staining or greater than 10% of invasive cells are with moderate membrane staining are forwarded for assessment by FISH.
- Current HER2 testing guidelines are followed which recommend a two-tier service using IHC to detect HER2 protein expression with analysis of equivocal HER2 (2+) cases by FISH to detect gene amplification.
- HER2 FISH, using an approved dual colour assay, to determine HER2 status for the use of adjuvant trastuzumab (Herceptin) therapy, in support of NICE guidance in early breast cancer (see Cytogenetics User Guide).

Gastric HER2

- If HER2 testing in advanced gastric or gastroesophageal cancer by IHC and FISH is required. **Please refer all cases of gastric tumours directly from the pathologist to avoid delay in testing.**
- Further information on arrangements for HER2 testing can be obtained from Dr Mansoor (was.mansoor@nhs.net), Medical Oncologist at The Christie

PD-L1 22C3 to identify patients for treatment with pembrolizumab
(UKAS accreditation approved: 8697 Medical Single (ukas.com))

- PD-L1 22C3 is a Companion Diagnostic for Gastric or GOJ Adenocarcinoma, where a CPS ≥ 10 identifies gastric or GOJ adenocarcinoma patients for treatment with KEYTRUDA® (pembrolizumab).

Referrals for FISH only are welcome

Enquiries regarding testing for all cell markers by IHC or FISH are welcome.

BTR Test	Target Reporting Time (Calendar days)
Immunohistochemistry	7 days
FISH	14 days

- Turnaround times are assessed as time of receipt in the laboratory to release of report for HER2 IHC & HER2 FISH.
- Turnaround times are dependent on the time taken for the sample to reach the laboratory from the requestor

Technical background to Breast and Gastric Analysis

HER2 signals are counted in 20 to 60 non-overlapping invasive cancer cell nuclei by at least two analysts and over three or more distinct tumour fields. Additional cells are scored in borderline cases.

- A ratio of 1.80-1.99 or a ratio <2.0 and average HER2 gene copy number between 4.0 and 6.0 will be reported as 'Borderline but Not Amplified' and include a clear statement that the tumour is HER2 Negative.
- A ratio of >2.0 and/or mean HER2 copy number >6.0 will be reported as Positive.
- Monosomy or partial monosomy of chromosome 17 leading to a dual probe ratio >2.0 will be reported as 'Positive with Monosomy

References:

(Rakha EA et al. J Clin Pathol 2014;0:1-7). Aneusomy i.e. deletions / gains (polysomy) are common in breast cancer and measurement of chromosome number is critically important (ref Walker et al, J Clin Pathol 2008)

IVD/CE marked. PD-L1 IHC 22C3 pharmDx can be used for qualitative immunohistochemical assessment using monoclonal mouse antibody. It is used in the detection of PD-L1 protein in formalin-fixed, paraffin-embedded (FFPE) gastric or gastroesophageal junction (GOJ) adenocarcinoma. PD-L1 protein expression in gastric or GOJ adenocarcinoma is determined by the Combined Positive Score (CPS), which is the number of PD-L1 staining cells (tumour cells, lymphocytes, macrophages) divided by the total number of viable tumour cells, multiplied by 100

The Histopathology department works closely with the MCRC Biobank and samples fresh surgical tissue for inclusion in consented trials and research projects. We also provide a service to process material taken for trials directly into paraffin wax blocks. Archived paraffin wax blocks can also be retrieved for use in Trials.

Disposal of unwanted Formalin

Out of date or unused formalin (from trials etc.) can be brought to the Histology lab for disposal. The formalin should be decanted into a 5l plastic container and labelled as waste Formalin. (an empty container and waste label can be acquired from Histology if necessary, and formalin can be decanted in Histology under fume extraction, NB we do not have the staff to carry out this task for other departments)

Consent

It is the responsibility of the requesting clinician to obtain consent from the patient for the collection, processing and retention of specimens.

Instructions for requesting tests

The language and criteria here need to match Pathgen Acceptance and Histology Acceptance. (decide and copy and paste into all 3 docs)

All specimens must be fully labelled and accompanied by a completed Histology or Breast Receptor request form. Inadequately or incorrectly labelled samples will not be processed. The Breast Receptor form must be accompanied by an authorised Histology Report.

THE REQUEST FORM:

Required:

- Forms should be labelled with an addressograph label showing patients full name, hospital number, date of birth and NHS Number
- All relevant clinical information **MUST** be included
- High risk status MUST be indicated where appropriate on the form.
- Legible signature of specimen taker to enable contact from the laboratory in case of queries
- Requestor's contact number.
- Details of previous treatment required to inform subsequent Histology report

Desirable:

- Consultant, location and date
- The time the specimen was taken

THE SPECIMEN:

Specimens should be placed in appropriately sized specimen containers filled with 10% Buffered Formalin. Bone Marrow Trephines should be placed into a 60ml pot containing AZF. Specimens for frozen sections need to be placed into a sterile 60ml sized pot without fixative and taken immediately to the Histopathology laboratory for receipt and processing. Fluid for peritoneal washings should also be brought to Pathology as soon as possible and refrigerated if not dealt with straight away.

Required:

- Addressograph labels should be attached to specimen containers.
- Specimen details should be written on the label of the pot, and should

exactly match details on the Histology card.
- Legible signature of specimen taker.

Desirable:

- Date and time specimens were taken
- Location of patient

URGENT SAMPLES

- Requests to process samples urgently should be clearly written on the Histology form.
- 31/62 target stickers should be attached to the form if appropriate.

Non-compliance:

1. Any unlabelled specimens will be returned to Surgical Theatre, or the staff member taking the specimen will be contacted to come and identify the specimen.
2. The specimen **will not** be processed until it has been labelled. A note will be made on the final report that the specimen was received unlabelled and that the laboratory cannot take responsibility for actions taken as a consequence of the report issued.
3. Incorrectly / inadequately labelled specimens - an attempt will be made to contact the requestor to allow correction / completion of the labelling. Where there is conflicting information the person taking the specimen will be asked to re-label it. If the name is correct but other details are incorrect the specimen can be sent back for correction.

Instruction for transportation of samples

Histopathology samples should NEVER be sent via the pneumatic tube system, but should be delivered to Pathology Reception by hand.

Various personnel within the Trust will be involved in transport of specimens to and from the laboratory. In order to protect their own and others safety the following guidelines should be followed.

- Cover any cuts and grazes with a waterproof dressing.
- Touch specimen containers as little as possible, washing hands as soon as practicable afterwards.
- Diagnostic samples must be sealed in the plastic bag and request form put into the open pocket of the bag. This is particularly important for fresh specimens, and peritoneal fluids, which need to reach the laboratory as soon as possible.
- Carry all specimens in the trays or boxes, where provided, never in pockets.
- If a specimen leaks into a tray or box, tell the laboratory reception staff and they will refer the specimen to a member of Histology staff.

- If a specimen is dropped or broken, do not touch it or try to clear up the mess. Stay with the specimen to prevent other people touching it and send someone to the laboratory for assistance. If you spill the fixative onto your clothing, you must remove it at once and then wash your hands and put on clean clothing.
- Report the accident to a supervisor as soon as possible.
- Keep specimen containers upright at all times.

Histology specimens should never be transported in the POD system, as they are irreplaceable.

Reporting of Results

Histology specimens must be fixed and processed before the resulting slides can be reported by the Consultant Pathologist. This process can take several days, the expected TRT for Histology results would ideally be 10 working days or less. However, due to the complexity of the patient samples received here at the Christie, this may take longer as a result of sending samples for complex molecular testing in referral laboratories.

Breast/Gastric Receptors are reported within 7 days for IHC and 14 for FISH. (For further info on FISH see Cytogenetics user guide).

Please note that the primary method for transmission of reports is to the Clinical Web Portal (CWP).

Uncertainty of Measurement

All test results are subject to a degree of uncertainty of measurement. This may be due to a range of factors, including:

- Biological variation within individuals
- Analytical measurement imprecision
- Pre-analytical factors

If you require more information regarding the effects of these factors on the outcome of an individual test result, please contact the lab on 0161 446 3290.

Clinical advice and interpretation

Clinical advice on examinations and interpretation of results is available by contacting a Consultant Histopathologist or Specialist Registrar via telephone.

For BTR interpretation queries please contact Angela Cramer or a member of the BTR team on 0161 446 3211

Opinion Service

The Histology Dept. also provides an opinion service, which can be accessed by sending relevant blocks/slides with a covering letter providing all relevant clinical information to the Pathology Office. Blocks and slides should be packaged appropriately according to current postal regulations, and will be returned as soon as possible after reporting.

Comments/Complaints Procedure

Any complaints or concerns about any aspect of the service should be raised initially with the Histology Service manager, Ms Catherine Billington, tel: 0161 446 3289

We are keen to know about any problems arising from the laboratory service. Feedback from our users will help in our constant efforts to improve our service; a user satisfaction survey is available.

Data Protection

The Christie Pathology Partnership (CPP) is committed to deliver a first class confidential service ensuring that all patient information is processed fairly, lawfully and transparently. Confidential information about patients can only be used for healthcare and relevant business purposes. All staff are required to comply fully with The Trust Governance Operating Framework for handling of patient confidential information.

In addition to this all HCPC registered staff follow the HCPC confidentiality guidance for registrants

The CPP also follows the Synlab group Privacy Policy

Quality assurance and Accreditation

Quality Statement:

The laboratory examinations, procedures and reports of test results are compliant with the requirements for quality and competence in medical laboratories according to United Kingdom Accreditation Service against the International Standard ISO15189:2012. UKAS **Registration Number: 8697.** A list of accredited tests can be accessed on the UKAS website. The department participates in all appropriate National External Quality Assurance Schemes (NEQAS) where available. Documentation relating to Internal Quality Control and performance in NEQAS are

available for scrutiny by users of the service. Performance is monitored and subject to rigorous control, to ensure that analyses are accurate, precise and results are comparable with other laboratories. A list of accredited tests can be accessed on the UKAS website.

Certain IHC tests are not accredited by UKAS – see list below:

ADD EBER, CK19 These are out of UKAS scope
Barbara to do gap analysis and provide update

2SC	CD22	Lysozyme	RB358
AFP	CD35	Mast Cell Tryptase	ROS-1
ALK-ETOP (D5F3)	CD38	MIC-2	SATB2
Annexin	CD42b	MUC-4	SDHB
AR	CD44	Myo D1	SF-1
BAP-1	Claudin	Napsin A	SOX11
BOB-1	c-Myc	NK1C3 (CD63)	SOX2
BRAF	Collagen IV	NKX3.1	STAT6
CAIX	CXCL13	Oct 2	T cell Receptor βF1
Caldesmon	FH	Oct 4	TIA-1
Calponin	Glycophorin	Pax 8	Tyrosinase
Cath K	Hepar-1	PD-1	Uroplakin II
CD11a	HNF1B	Perforin	Uroplakin III
CD11c	ICOS	Podoplanin	
CD138	LEF1	PTEN	

The department is approved by the Institute of Biomedical Sciences (IBMS) as a **Training Laboratory** and all our qualified scientists are registered with the Health & Care Professions Council (HCPC).

The laboratory also regularly monitors the UKAS accreditation status of the referral laboratories used for specialist testing.

Names and addresses of referral laboratories

Referral laboratory	UKAS reg no.	Accreditation status

Genomics Diagnostics Lab St Mary's Oxford Road Manchester M13 9WH Tel: 0161 276	**UKAS 9865**	**Accredited**
Histopathology Dept The Royal Marsden Fulham Road London SW3 6JJ	**UKAS 9929**	**Accredited**
Histopathology Dept Manchester University NHS FT Wythenshawe Hospital Southmoor Road Wythenshawe M23 9LT	**UKAS 9083**	**Accredited**

Useful Links

Lab Tests Online: Lab Tests Online.org.uk

11. Document Locations

Electronic Version	1. Histopathology Intranet Site/Christie Website
Any other printed copies of this document are unauthorised.	

12. Procedure Amendments

This replaces all previous versions of the document