LONDON SCHOOL of HYGIENE &TROPICAL MEDICINE

UKAS MEDICAL

9148

Accessible Version
Diagnostic Laboratory Parasitology Laboratory User Handbook
November 2024

Faculty of Infectious and Tropical Diseases
London School of Hygiene & Tropical Medicine
Keppel Street
London WC1E 7HT

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Tottenham Court RD92WC

www.parasite-referencelab.co.uk

Telephone number: +44 (0) 20 7927 2427

Contents

A. Introduction

1. The Laboratory and outline of services

The Diagnostic Parasitology Laboratory is an ISO 15189 UKAS accredited medical laboratory (number 9148) based at the London School of Hygiene and Tropical Medicine, which is itself a centre of excellence for scientific research and postgraduate education. The Parasitology Laboratory provides a reference facility offering a wide range of parasitological investigations for enteric parasites, blood parasites and acanthamoebae. We also offer technical advice on methodology and laboratory procedures.

The services above are offered to all hospitals, NHS and UK Health Security Agency (UK HSA) laboratories, General Practitioners and private medical laboratories throughout the UK.

The Parasitology Laboratory also has considerable expertise in the diagnosis of parasites of non-human primates and offers this service to veterinary practitioners. Please note that this service is not in our UKAS-accredited scope.

Advice on the investigation of malaria and other parasitological diseases is always available; technical teaching sessions can also be arranged for small or large groups.

The laboratory processes over 2700 specimens annually and participates in the NEQAS Blood Parasitology, Faecal Parasitology, Canadian Blood Parasitology and SKML Acanthamoeba Molecular schemes.

Please note that all parasitology serology is performed at Clinical Parasitology, Health Services Laboratory (HSL), LLP Analytics at the Hospital for Tropical Diseases, see page 15 for contact details.

Please note that entomology investigations are conducted by the LSHTM Medical Entomologist and are not within the scope of accreditation of the Diagnostic Parasitology Laboratory.

2. Laboratory policy

Our policy is to offer a first class diagnostic and reference facility for parasitic infections. We are also strongly committed to active research into parasites and their diagnosis and to the provision of training for pathology staff, other healthcare professionals and those working in the control of infectious diseases.

3. Using this handbook

This handbook is designed to aid and advise the user on the appropriate use of the facilities to include diagnostic, teaching and advisory services. The sections are indexed to assist in understanding the structure of the laboratory, identification of staff, the diagnostic and advisory services offered and the specimens and investigations carried out by the laboratory.

Parasites and diseases are listed alphabetically and each section gives an outline of the specimens required for investigation and the tests carried out. This is not an exhaustive list and users not finding a specific requirement should contact the laboratory for help and advice. Further copies of this handbook can be downloaded from the DPL website at www.parasite-referencelab.co.uk where referral forms are available in the Resources section. Other useful information and related links are on the website.

4. Malaria Reference Laboratory requests

The UKHSA Malaria Reference Laboratory is also located in the same department and will deal with all malaria requests. A separate handbook is available to download from the MRL website at www.malaria-reference.co.uk

Specimens for malaria diagnosis should be addressed to the UKHSA Malaria Reference Laboratory where they will be dealt with appropriately.

Please note that malaria serology is performed at Clinical Parasitology (HSL), LLP Analytics at the Hospital for Tropical Diseases, see page 15 for contact details.

5. Customer Satisfaction

We encourage users of our services to make any suggestions with regard to quality improvement and customer satisfaction.
Suggestions, queries or complaints should be made to:-

Dawn Britten, Quality Manager
Email address: Dawn.Britten@lshtm.ac.uk

And or

Claire Rogers, Principal BMS.
Email address: Claire.Rogers@lshtm.ac.uk

B. Laboratory and Staff

1. Laboratory opening times

The diagnostic laboratory is open between the hours of: -

Monday to Friday 9:00 am to 5:00 pm

when staff are available for advice, information, specimen reception and processing.

Most routine specimens are sent to us by post or the DX system, any urgent specimens are usually delivered by courier and accepted during the hours stated**.**

Please telephone urgent requests prior to despatch to inform us of their impending arrival and priority status.

2. Out of hours and public holidays

We offer foremost a reference facility and therefore do not provide an on-call service. The majority of specimens we receive will have had a preliminary diagnosis made by the sender or primary laboratory and so are usually non-urgent. The laboratory is usually closed on public holidays; when there is an extended holiday period, for example Christmas and New Year, limited cover is arranged to deal with non–urgent postal specimens, and all users are informed of these arrangements prior to the holidays.
Any specimens delivered to the Diagnostic Parasitology Laboratory out of normal hours and where no prior arrangement with us has been made will be held and dealt with the next working day.

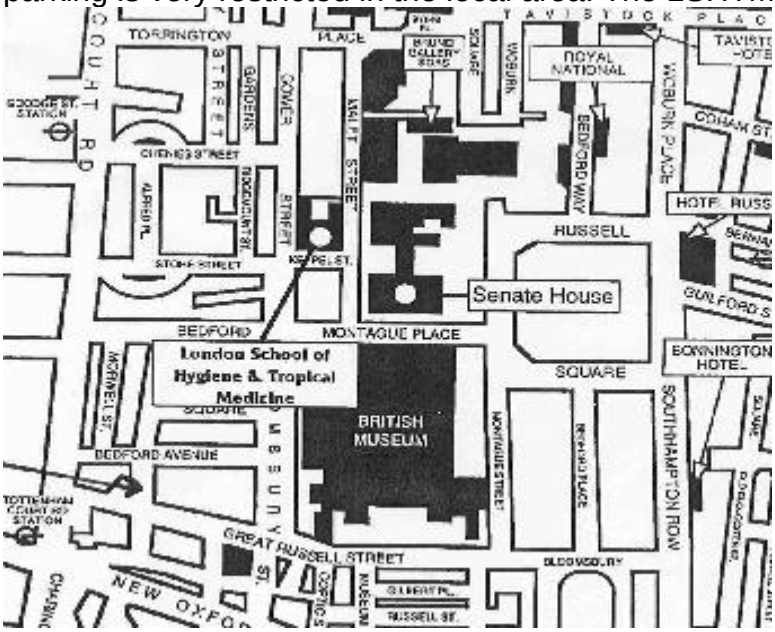
Where urgent diagnosis is required out of normal hours for malaria, Babesia or African trypanosomiasis only, specimens should be referred to: -

Clinical Parasitology at HSL, LLP Analytics at the Hospital for Tropical Diseases, telephone number: 0845 155 5000 (UCLH switchboard) and ask for the on-call Biomedical Scientist in Parasitology.

Please note that this service will incur a charge.

3. Location

The LSHTM is situated close to Tottenham Court Road, Goodge Street and Russell Square underground stations and is a 10 minute walk from Euston Main line station. Car parking is very restricted in the local area. The LSHTM has no parking facilities of its own.



4. Visitors

Please note that we are not able to see members of the general public and all patient referrals must be made by a registered health professional to the HTD, where the clinical service is delivered. There are no clinics at the LSHTM.

5. Staff and telephone numbers

Each of the following staff member has a role, position, name and telephone number described:

1. Director, Consultant Parasitologist, Professor P L Chiodini, 020 7927 2427

2. Biomedical Scientists

    a) Principal BMS, Head of Teaching & Diagnostics Unit, Claire Rogers, 020 7927 2318

    b) Lead BMS, Molecular Laboratory, Dr Debbie Nolder, 020 7927 2303

    c) Lead BMS and Quality Manager, Dawn Britten, 020 7927 2318

    d) Advanced BMS Non-Molecular Laboratory, Sarah Cheeseman, 020 7927 2427

    e) Advanced BMS Molecular Laboratory, Helen Liddy, 020 7927 2303

    f) Specialist BMS, Rita Mistry, 020 7927 2427

    g) Trainee BMS, Saba Entezam, 020 7927 2427

    h) Trainee BMS, Lindsay Stewart, 020 7927 2303

    i) Trainee BMS, Niamh Murphy, 020 7927 2427

3. Associate Practitioners

    a) AP, Helena Stone, 020 7927 2427

4. Medical Laboratory Assistants

    b) MLA, Karen Osborne, 020 7927 2427

    c) MLA, Keir Hughes, 020 7927 2427

5. Medical Entomologist, Cheryl Whitehorn, 020 7927 2344, (Member of the LSHTM Faculty of Infectious & Tropical Diseases entomology staff, who provides expertise to diagnostic laboratory)

C. Diagnostic and Advisory Services

1. Results, information and enquiries

For consultation on the investigation and diagnosis of parasitological disease, results and general information, please contact the laboratory on 020 7927 2427 from where enquiries can be answered or referred to appropriate personnel.

2. Specimens and containers

For the majority of investigations, the submitting laboratory or institution will refer specimens in an appropriate container of their choice.
For certain investigations or where special conditions are required, kits and containers are available from us upon request.

For example: -

- A collection kit containing 1 plain vial and 3 fixative vials (SAF) with collection instructions is available for investigation of both intestinal cysts and trophozoites including *Dientamoeba fragilis & Blastocystis hominis.*

See from page 16 for guidelines as to appropriate specimen type for specific investigations.

Please telephone us if in any doubt as to which container to use or to discuss specific requirements.

3. Referral forms and verbal requests

A referral form must accompany all specimens referred to the laboratory and we ask that requesting practitioners use our Diagnostic Parasitology Laboratory referral form.
Please note that investigations into Acanthamoeba keratitis and Trypanosoma cruzi should be referred using our specific Acanthamoeba and Trypanosoma cruzi referral forms. There is additionally an Animal referral form for veterinary referrals.

Hard copies of all referral forms can be supplied upon request or the forms may be downloaded electronically, as a pdf, from the laboratory website at: www.parasite-referencelab.co.uk where they can be found in the Resources section.

Verbal requests

Verbal requests for further investigations on samples already received will be recorded on the original referral form.

Verbal requests for investigations on samples not yet received must also be confirmed in writing, enclosing a referral form with the specimen.

Where samples are received without a referral form, we will attempt to telephone the referring laboratory to discuss. We will ask for a referral form to be forwarded to us via secure e-mail and give details of an appropriate e-mail address.

4. Minimum Data Set

As a guide, the following information sets our minimum data requirements to ensure patient safety. Where specimens or referral forms are received that do not meet this criteria, we shall contact the sender for more information but reserve the right to reject the sample.

1. Sample:
    a. Essential:
        i. Full name OR other coded patient identifier
        ii. DOB & or Hospital number, Unit number or **NHS Number**
    b. Desirable:
        i. Sending laboratory's reference number.
        ii. Date and time specimen taken
        iii. Nature of specimen including qualifying details e.g. right or left

2. Request form:
    a. Essential:
        i. Full name OR other coded patient identifier
        ii. DOB &or Hospital number, Unit number or **NHS Number**
        iii. Investigation required
        iv. Name and address of requesting practitioner and name and address for reports if different
    b. Desirable:
        i. Sending laboratory reference number
        ii. Date and time specimen taken
        iii. Nature of specimen including qualifying details e.g. right or left
        iv. Telephone or bleep number of requesting practitioner
        v. Gender
        vi. Sending laboratory's diagnosis
        vii. Clinical information

Please note the following extract from IBMS Professional Guidance:
*"Use of the **NHS or CHI** Number on paper and electronic patient records is a mandatory requirement included within the NHS Operating Framework 2008/9. Patient data should be used to identify the sample up to the point where a NHS or CHI Number is allocated whereupon this becomes the primary identifier."*

5. Clinical Information

All relevant clinical details to include, where available

- antimicrobial therapy
- travel history
- risk status if applicable
- date of onset and duration of illness
- anatomical sites from which biopsies, foreign bodies, insects, other specimens taken
- epidemiological information, for example family or institutional outbreaks

6. Packaging

Packaging must comply fully with UK transport regulations for clinical specimens (UN 3 3 7 3 regulations).

Specimens should be in an appropriate container, securely fastened and the accompanying referral form should be placed in a separate area of the packaging so as not to be in direct contact with the specimens.

The outside of the package must be appropriately marked and clearly state:-

‘**Biological substance - category B’**

In those circumstances where specimens are unlabelled or inadequately labelled and the patient’s identification is unclear, or if they have leaked or are contaminated, they may be unsuitable for testing. In such instances the requesting laboratory or doctor will always be informed immediately by telephone to discuss the matter and to arrange for repeat specimens if necessary.

As a reference laboratory we do appreciate that some specimens cannot be repeated and every effort is made to avoid the need for repeat requests.

7. High risk specimens and safety

Specimens are regarded as HIGH RISK if taken from patients known, suspected, or at risk of having serious infectious disease.
Of note are blood-borne agents such as Hepatitis, HIV, or various viral haemorrhagic fevers or other infectious diseases such as tuberculosis or typhoid. Ova and segments of *Taenia solium* pose a parasitological high risk.

For further advice on patient management, please contact the Imported Fever Service on 0844 778 8990.

Hazard Group 3 risks:-

In addition to the standard packaging instructions given, all high risk specimens must be labelled as HIGH RISK both on the container and the request form, with a standard yellow ‘danger of infection’ sticker, and placed in a Biohazard bag.

The precise nature of the infection risk should be clearly given in the clinical details.

Hazard Group 4 risks:-

This is essentially referring to viral haemorrhagic fevers but also similar human infectious diseases of high consequence.

Specimens from a patient with a **confirmed** hazard group 4 pathogen.
Specimens containing HG4 organisms cannot be dealt with in this laboratory. The referring clinicians must not send any specimens and must discuss the case with the Imported Fever Service on 0844 778 8990.

Samples from a patient with a **possible risk** of a hazard group 4 pathogen.
The referring clinicians must first discuss the case with the Imported Fever Service on 0844 778 8990. The risk assessment and any VHF screening required as a result of this should have taken place before specimens are sent to us and we may receive specimens once the risk has been downgraded.

8. Specimen transport & reception

Most specimens are received via post, DX or courier. During normal working hours, all specimens are received at LSHTM reception from where they are forwarded to the reference laboratory. Outside normal working hours, all specimens are received at reception and will be dealt with on the next working day.

9. Urgent investigations

Urgent specimens are usually delivered by courier and accepted during normal working hours. Please telephone urgent requests before you send the samples so that we know they are coming, also ensure that you have provided full and correct contact details so that we can telephone the results back to you. Please package as described earlier and mark clearly on the outside that it is urgent.

When an urgent malaria, Babesia or African trypanosomiasis diagnosis is required outside of normal hours, specimens should be referred to Clinical Parasitology, HSL, Analytics LLP at the Hospital for Tropical Diseases (HTD). Call 0845 155 5000 (UCLH Switchboard) and ask for the on-call Biomedical Scientist in Parasitology.

Please note that this service will incur a charge.

Please note that if a specimen is enroute to LSHTM and you wish it to be re-directed to HTD, you must arrange this directly with the courier, as LSHTM staff cannot authorise a courier to re-route.

Any specimens delivered to us out of normal working hours, if special arrangements have not been made with us, will remain securely at LSHTM Reception until being transferred to the laboratory on the following working day.

D. Specimens and Investigations

1. Specimen collection

General

As a reference laboratory, many specimens received have already had preliminary investigations carried out by a primary laboratory and are sent to us in due course.

In general, specimens should be collected or transferred into an appropriate container such as a sterile universal, with an accompanying referral form giving all relevant information, including laboratory findings and sent to us with minimal delay.
Please inform us of any known infection risks.

Please provide a specimen representative of the condition under investigation and in sufficient quantity to permit a full examination – see specific guidelines on pages 17-39.

Where a specimen is submitted for a general screen with no specific parasitological investigation requested we may, as a result of our findings or as indicated to us by the specimen type or clinical details given, proceed to further investigations as deemed appropriate.

Faeces

Unless otherwise requested, faecal specimens are given a general parasitological screen to include:-

- examination for adult worms and segments
- formol-ether concentration and microscopy for ova, cysts and larvae
- amoebic culture

If the presence of trophozoites is to be investigated, faeces must either be fresh or collected into SAF preservative (available upon request). Ideally when investigating trophozoites, the sample should be transported by courier at 37 degrees centigrade or ambient temperature, but certainly not refrigerated.
See also guidelines for intestinal parasites and specific parasitic diseases.

Blood

See specific parasitic diseases for guidelines on volume, anticoagulants, blood smears etc.

Serology

We do not perform serological tests, all parasitology serology is performed by Clinical Parasitology, HSL Analytics LLP at the Hospital for Tropical Diseases (HTD) and those received by us will be forwarded to the HTD. Referring laboratories should send specimens directly to the HTD:

Address:
Clinical Parasitology, HSL Analytics LLP
Hospital for Tropical Diseases (HTD)
Mortimer Market

Capper Street
London
WC1E 6JB

Telephone 020 7307 9400 and ask for Parasitology Serology

Tissues and biopsies

Under aseptic conditions, transfer material to a sterile universal container. If the sample is very small, add 0.5ml of sterile saline to prevent drying. Please refer to specific investigations for more detail.

Worms and worm segments

Adult worms and tapeworm segments should be sent without preservative in a suitable container. If there is likely to be a delay of more than 24 hours, then 70% alcohol should be added to the specimen.

Ectoparasites and Entomology

Soft-bodied specimens (such as mites, maggots, other larvae) should be sent preserved in 70% alcohol in a suitable container; if left unpreserved they may degrade during the transportation period. Hard bodied specimens (beetles, flies, anything with a rigid exoskeleton) should be sent dry, as certain morphological features are lost in flies preserved in alcohol (this is particularly true of mosquitoes). If possible, please add silica gel to dry samples to minimise risk of fungal growth during transportation.
Where extracted from a body site and there is a risk of infection, specimen may be fixed in 10% formol water, rinsed in water and transferred to 70% alcohol. Do not leave in formol water as this hardens some specimens. Please allow specimen to remain intact if possible, giving full clinical details including travel history and site of extraction if relevant. If in any doubt please call to discuss.
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2. Specimen retention

Specimens are retained for varying periods of time according to type. Should additional tests be requested after the initial referral, please observe the following guidelines:-

Faeces, urine, blood, body fluids, CSF:-
A minimum of 2 weeks at 2-8 degrees centigrade after receipt

Worms, segments, insects:-
A minimum of 2 weeks after receipt

Tissues, biopsies:-
4 weeks after final report, at 2-8 degrees centigrade

Blood sent for T cruzi PCR :–
After DNA extraction, 1.0 millilitre aliquots of blood are held at -20 degrees centigrade indefinitely (where sample volume permits).

3. Parasitic diseases and their laboratory investigation

The above guidelines and the following specific parasitic diseases are not intended as an exhaustive list of parasitological investigations available; please contact the laboratory to discuss individual cases, specific requirements or investigations not listed.

Please refer to the following pages where all are listed in alphabetical order.

All parasitology serology is now performed at Clinical Parasitology at HSL, LLP Analytics at the Hospital for Tropical Diseases.

Amoebiasis:
Intestinal amoebiasis, amoebic dysentery and invasive amoebiasis

Causative organism: *Entamoeba histolytica* (pathogenic) *or Entamoeba dispar* (non-pathogenic)

Diagnosis: formol-ether concentration and microscopical examination of faeces for cysts

direct microscopy of fresh faeces or pus for trophozoites

amoebic culture from faeces or pus

microscopy of Iron-Haematoxylin stained faecal preparations

specific antigen ELISA for the detection of *E. histolytica* adhesin in faeces and culture

Specimens: faeces in plain container for concentration and culture

for OCP screen, minimum 1 gram faeces or 2 millilitre if sample is liquid
for ELISA, minimum 0.2 gram faeces or 400 microlitre if liquid
if TechLab ELISA has been requested, ideally the faeces should be no more than 48 hours old for optimal test sensitivity

SAF-fixed faeces for Iron-Haematoxylin staining of cysts and trophozoites

fresh faeces or rectal scrapings for trophozoite investigation to be carried out within 1 hour of sample being taken – please inform laboratory in advance
ideally when investigating trophozoites, the sample should be transported by courier at 37 degrees centigrade or ambient temperature, but certainly not refrigerated

*pus from liver or lung abscess – please inform laboratory in advance

* Advice on collection of these samples is available from the Doctor on Duty in Infectious & Tropical Diseases at the Hospital for Tropical Diseases – telephone 0845 155 5000 (UCLH switchboard)

N.B. Due to the intermittent nature of passage of trophozoites and cysts in faeces, it may be necessary to examine 3 or more samples, collected on different days as this increases sensitivity of detection.

Free living amoebae:

Amoebic keratitis

Causative organism: *Acanthamoeba species.*

Diagnosis: microscopy and culture
PCR

The greatest diagnostic sensitivity for Acanthamoeba is achieved through use of both *in vitro* culture, microscopy and a probe-based qPCR.

Specimens: see detail below for suitable sample types and sample preparation

Please ensure all containers are tightly screwed and use Parafilm (NOT Sellotape) to prevent leakage during transit.

- **Clinical samples** (corneal scrapes, biopsies, fluids, swabs etc.) should be sent in a small volume (1 – 2 millilitre ideal) of sterile saline or sterile distilled water in a small (less than 5 millilitre) sterile vial or tube.

  Material from a **needle or blade scrape** should be rinsed into the saline or water. Please remove blades or needles after rinsing. Do NOT leave the blade in the tube as it rusts: this inhibits our PCR and may have a detrimental effect on culture isolation.

  Swabs or washings appear to be less efficient in detecting the organism. If swabs must be sent then please add a small volume of sterile saline or sterile distilled water to the swab to prevent drying. Please do not send dry swabs.

  Punch **biopsies** or portions of excised cornea may also be submitted: put sample into a small volume of sterile saline or distilled water in a small sterile vial.

- **Non-clinical samples -** contact lenses: should be sent in their lens cases (ie in used contact lens fluid.) Culture is performed on lenses and fluids; PCR is performed on fluids only. N.B. Isolation of *Acanthamoeba* from contact lens-related specimens, whilst suggestive, does not necessarily implicate the amoeba as causing the patient's symptoms. Amoebic genera (other than *Acanthamoeba*), flagellates, ciliates and other organisms may be found in contaminated washing fluids and on lenses, particularly with poor lens hygiene.

- **Culture-positive samples**: please send original culture plate if possible, or blocks of agar from the plate in a sterile vial.

- **Commercial contact lens solutions** we do NOT test commercial contact lens solutions (other than that already in patients' contact lens cases).

All specimens should be submitted for testing together with a completed *Acanthamoeba* referral form

Free living amoebae:

Granulomatous Amoebic Encephalitis (GAE)

Primary Amoebic Meningoencephalitis (PAM)

Causative organisms; *Acanthamoeba species*., *Balamuthia species*. (GAE), *Naegleria fowleri* (PAM)

Diagnosis: microscopy, culture, PCR

Specimens: CSF and biopsy material

N.B. These infections require urgent diagnosis; telephone advice should be obtained as soon as infection is suspected.

Investigations for GAE and PAM are currently performed at:

Clinical Parasitology, HSL Analytics LLP
Hospital for Tropical Diseases (HTD)
Mortimer Market
Capper Street
London
WC1E 6JB

Telephone 020 7307 9400 and ask for Parasitology Microscopy

Babesiosis

Causative organism: *Babesia species.*

Diagnosis: microscopical examination of thin and thick blood films for parasites stained with Giemsa and Field's

Specimens: 2 thin (methanol-fixed) and 2 thick (unfixed) blood films sent in a slide container.
Blood taken into anticoagulant (EDTA should be used) and films made with a minimum of delay and preferably within 2 hours of taking the blood.

Blood should be taken at peak of parasite density as indicated by fever; however parasites may be found in the absence of fever and the examination of blood films should NOT be delayed. Repeat blood films may be necessary to demonstrate infection.

N.B. Travel history and splenic status is important in the diagnosis of this infection.

Cryptosporidiosis

Causative organism: *Cryptosporidium species.*

Diagnosis: microscopy after acid fast staining of faecal smears using a modified Ziehl-Neelsen stain or fluorescent microscopy after phenol auramine staining for detection of oocysts.

immunochromatographic rapid antigen test

Specimen: faeces in a plain container or fixed in SAF
Minimum 1 millilitre for antigen test

N.B. Due to the intermittent nature of passage of oocysts in faeces, it may be necessary to examine 3 or more samples, collected on different days as this increases sensitivity of detection.

Cyclosporiasis

Causative organism: *Cyclospora spp.*

Diagnosis: microscopical examination of faecal smears stained by modified Ziehl-Neelsen for oocysts and direct microscopy following formol-ether concentration

Specimens: faeces in plain container

N.B. Due to the intermittent nature of passage of oocysts in faeces, it may be necessary to examine 3 or more samples, collected on different days as this increases sensitivity of detection.

Cystoisosporiasis (Isosporiasis)

Causative organism: *Cystoisospora belli (Isospora belli)*

Diagnosis: microscopical examination of faecal smears stained by modified Ziehl-Neelsen for oocysts and direct microscopy following formol-ether concentration

Specimens: faeces in plain container

N.B. Due to the intermittent nature of passage of oocysts in faeces, it may be necessary to examine 3 or more samples, collected on different days as this increases sensitivity of detection.

Enterobiasis

Threadworm or Pinworm

Causative organism: *Enterobius vermicularis*

Diagnosis: microscopical examination for ova

Specimens: adhesive tape smears of perianal skin

perianal swab

faeces in clean container (low sensitivity)

Adhesive tape or swab preferred.

Cut a 10cm strip of Sellotape, or similar, and press middle 3 to 5cm firmly against the right and left perianal folds, sticky side down. Stick tape onto a microscope slide and place in a slide box.

or

Moisten a swab in sterile saline and repeatedly roll over the whole of the perianal area; break off into a small volume of saline in a sterile universal.

Carry out either procedure first thing in the morning before bathing or defaecation. Repeated samples over 4 to 6 consecutive days may be necessary as this increases sensitivity of detection.

BEWARE eggs are highly infectious and resistant to drying!

Filariasis

Causative organisms: of particular importance are *Loa loa, Wuchereria bancrofti. Brugia malayi, Onchocerca volvulus*

Diagnosis: membrane filtration and microscopical examination of peripheral blood for microfilaria (except *O. volvulus*)

examination of skin snips for microfilariae of *O*. *volvulus*

examination of histological material for adults

Specimens: 10 to 20 millilitre of citrated (preferred anticoagulant) blood (observe periodicity* if known, if not take at any time).

Additionally 4 unfixed thick films to be made from the blood with a minimum of delay.

Skin snips for *O. volvulus* placed into physiological saline; advice should be sought before taking skin snips.

*Depending on clinical and travel history, possible periodicity should be observed when taking blood.

The following filarial worms have a species name, period of activity in the body, geographical location and collection time described:

1. Wuchereria bancrofti, Periodic, nocturnal, Asia, Africa, Caribbean, South America, West Pacific, 10pm to 4am, peak midnight
2. Wuchereria bancrofti, Subperiodic, nocturnal, Thailand, Vietnam, 8 to 10pm , peak 9pm
3. Wuchereria bancrofti, Subperiodic, diurnal, South East Pacific, 2 to 6pm, peak 4pm
4. Brugia malayi, Periodic, nocturnal, South & East Asia, 10pm to 4am, peak midnight
5. Brugia malayi, Subperiodic, nocturnal, South East Asia, 8 to 10pm, peak 9pm
6. Brugia timori, Nocturnal, Lesser Sunda islands of Indonesia, including Timor, 10pm to 4am, peak midnight
7. Loa loa, Diurnal, West & Central Africa equatorial rainforests, 10 am to 3pm, Peak 1pm
8. Mansonella ozzardi, Central and South America, Caribbean, Non-periodic
9. Mansonella perstans, Tropical Africa, South America, Non-periodic

Giardiasis

Causative organism: *Giardia intestinalis* (synonym, *G. lamblia, G. duodenalis*)

Diagnosis: microscopy of fresh faeces for trophozoites and cysts

formol-ether concentration and microscopical examination for cysts

immunochromatographic rapid antigen test

Specimens: faeces in plain container (minimum 1 millilitre for antigen test)

if fresh and for trophozoites, examination should be carried out within 4 hours of specimen being produced.
ideally when investigating trophozoites, the sample should be transported by courier at 37 degrees centigrade or ambient temperature, but certainly not refrigerated

N.B. Due to the intermittent nature of passage of trophozoites and cysts in faeces, it may be necessary to examine 3 or more samples, collected on different days as this increases sensitivity of detection.

Hydatid infection

Causative organisms: *Echinococcus granulosus*, *E. vogeli* and *E.oligarthrus* for cystic hydatid and *E. multilocularis* for alveolar hydatid.

Diagnosis: microscopical examination for hooks and protoscoleces in hydatid sand

Specimens: fluid or contents of cysts (fixed in formalin)

N.B. Clinical advice on the management of hydatid disease should be sought before considering aspiration of a cyst as leakage of fluid may cause further dissemination or an anaphylactic reaction. Advice is available from the Doctor on Duty in Infectious & Tropical Diseases at the Hospital for Tropical Diseases– telephone 0845 155 5000 (UCLH switchboard)

Insects and other arthropods

Soft-bodied specimens (such as mites, maggots, other larvae) should be sent preserved in 70% alcohol in a suitable container; if left unpreserved they may degrade during the transportation period. Hard bodied specimens (beetles, flies, anything with a rigid exoskeleton) should be sent dry, as certain morphological features are lost in flies preserved in alcohol (this is particularly true of mosquitoes). If possible, please add silica gel to dry samples to minimise risk of fungal growth during transportation.

Where specimen has been excised from the patient and there is a risk of infection, the specimen should be fixed in 10% buffered formalin (as used for histology) or 10% formol water/saline, then rinsed in distilled water and transferred to 70% ethanol.

Allow specimen to remain intact if possible, giving full clinical details including travel history and site of extraction if relevant.

Please note that entomology investigations are conducted by the LSHTM Medical Entomologist and are not within the scope of accreditation of the Diagnostic Parasitology Laboratory

Intestinal parasitic infections with helminths and protozoa (general)

A wide range of nematodes, cestodes, trematodes and protozoa are dealt with in this laboratory; some are listed individually e.g. amoebiasis, cryptosporidiosis, cyclosporiasis, giardiasis, microsporidiosis, schistosomiasis, strongyloides.

See also pages for individual parasites

Diagnosis: macroscopical examination of faeces for adult worms and segments

direct microscopy of fresh faeces for trophozoites

formol-ether concentration of faeces and microscopy for ova, cysts and larvae

iron-haematoxylin staining and microscopy of SAF-fixed faeces for protozoal trophozoites

Specimens: faeces in plain container for adult worms, segments and concentration- for OCP screen, minimum 1 gram faeces or 2 millilitre if the sample is liquid. **N.B.** do not refrigerate if Strongyloides culture required.

SAF fixed faeces for trophozoites (especially suitable when a fresh sample is not practical and for fragile organisms e.g. *Dientamoeba fragilis & Blastocystis hominis*) sample volume as indicated on collection pot

fresh faeces for trophozoites
ideally when investigating trophozoites, the sample should be transported by courier at 37 degrees centigrade or ambient temperature, but certainly not refrigerated.

N.B.

1. Due to the intermittent nature of passage of parasites in faeces, it may be necessary to examine 3 or more samples, collected on different days as this increases sensitivity of detection.
2. We can supply kits for the collection of faeces in SAF – please contact us if required

Leishmaniasis

Visceral, Cutaneous & Mucosal leishmaniasis

Causative organism: Leishmania species.

Diagnosis: microscopy of stained aspirates and smears.

histological examination of ready-stained tissue sections for presence of parasites

Specimens: This laboratory only performs microscopy of impression smears or tissue sections from e.g. bone marrow, lymph, spleen, liver, tissue. Ideally the sending laboratory should send at least two smears, fixed and unstained, although we can also examine ready-stained smears.

For a full investigation of cutaneous or visceral leishmaniasis please contact Clinical Parasitology, HSL Analytics LLP, telephone 020 7307 9400 and ask for Parasitology which offers a range of investigations including culture, serology (for visceral Leishmaniasis) and PCR.

Malaria

Please refer to the separate UKHSA Malaria Reference Laboratory handbook for full details of this service, handbook and referral forms available at

www.malaria-reference.co.uk

N.B. Serology is occasionally useful in detecting evidence of past infection, but its main indication in the UK is for blood donor screening. **Serology has no place in the diagnosis of acute malaria, for which blood films are mandatory.**

Serology is no longer performed by this laboratory, but may be obtained from the Clinical Parasitology, HSL Analytics LLP at the Hospital for Tropical Diseases (HTD) see page 14-15

Microsporidiosis

Causative organisms: including *Enterocytozoon bieneusi*, *Encephalitozoon intestinalis*

Diagnosis: microscopical examination of strong trichrome stained faecal or urine smears for microsporidial spores

Specimens: faeces

urine

N.B. Due to the intermittent nature of passage of spores, it may be necessary to examine 3 or more samples, collected on different days as this increases sensitivity of detection.

Schistosomiasis

(Bilharzia)

Causative organism: *Schistosoma mansoni, S. haematobium, S. japonicum, S. intercalatum and S. mekongi*

Diagnosis: formol-ether concentration and microscopy of faeces for ova

microscopical examination of urine after concentration or filtration

microscopical examination of squash preparations of tissue for ova

examination of stained histology sections

Specimens: faeces in plain container, minimum 1 gram faeces or 2 millilitre if sample is liquid

urine in a plain, sterile container - either a midday urine specimen (between noon and 3 pm) or a 24-hour collection of terminal urine - if a single urine specimen is to be submitted there should ideally be a minimum volume of 10 millilitre
1 millilitre of undiluted formaldehyde should be added to urine to preserve any eggs that may be present.
N.B. eggs may be found trapped in the blood and mucus in the terminal portion of the urine specimen.

tissue, unfixed biopsy material (rectal, sigmoid, bladder) material for squashes

ready-stained sections for histological examination.

N.B. when screening after return from an endemic area, it is advisable to examine both urine and faeces.

Due to the intermittent nature of passage of parasites, it may be necessary to examine 3 or more samples, collected on different days as this increases sensitivity of detection.

Strongyloidiasis

Causative organism: *Strongyloides stercoralis*

Diagnosis: microscopical examination for larvae and adults
isolation of larvae

Specimens: faeces, for OCP screen, minimum 1 gram faeces or 2 millilitre if sample is liquid
Duodenal or jejunal aspirate

For isolation of strongyloides, 8-10 gram faeces and as much as possible for aspirates

N.B. Please do not refrigerate specimen if isolation is required

Due to the intermittent nature of passage of parasites in faeces, it may be necessary to examine 3 or more samples, collected on different days as this increases sensitivity of detection.

Trichinosis

Causative organism: *Trichinella spiralis*

Diagnosis: microscopical examination for larvae by tissue squash

Specimens: *unfixed or fixed muscle biopsy

N.B. it is widely considered unnecessary to perform biopsy for the diagnosis of this parasite, the alternative being serology (performed at Clinical Parasitology, HSL, HTD, see page 15)

Advice on collection of samples is available from the Doctor on Duty in Infectious & Tropical Diseases at the Hospital for Tropical Diseases – telephone 0845 155 5000 (UCLH switchboard)

Trypanosomiasis – African
Sleeping sickness

Causative organism: *Trypanosoma brucei rhodesiense*, *T.b. gambiense*

Diagnosis: microscopical examination of blood films for trypomastigotes

microscopical examination of cerebrospinal fluid where neurological involvement *****

Specimens: 2 methanol-fixed thin and 2 unfixed thick blood films for microscopy; blood taken into anticoagulant (preferably heparin) should be used and films made with a minimum of delay and preferably within 2 hours of taking the blood.

*CSF

*Advice should be sought before attempting to take CSF sample for diagnosis, due to risk of introducing trypanosomes into the CNS from the blood, please contact the Doctor on Duty in Infectious & Tropical Diseases at the Hospital for Tropical Diseases - telephone 0845 155 5000 (UCLH switchboard)

Trypanosomiasis – South American
Chagas disease

Causative organism: *Trypanosoma cruzi*

Diagnosis: microscopy of thick and thin blood films to identify trypanosomes for investigation of suspected acute stage Chagas disease (CD) or reactivated cases only: acute CD is extremely rarely seen in the UK.

blood films for microscopical examination should be submitted, in parallel with blood for qPCR, for investigation of acute CD in the following patient groups: neonates (congenital), transplant-recipients, needle-stick injuries and travellers. Also for cases of suspected reactivation of chronic infection.

PCR for investigation of suspected acute CD (essential) and for screening / monitoring of chronic CD.

N.B. Serology is the standard method for diagnosis of CD in the chronic stage and this should be performed before qPCR is considered. *T. cruzi* serology is performed at Clinical Parasitology, HSL at the Hospital for Tropical Diseases, see p14-15 for full details.

Specimens: thick and thin blood films
thin films should be methanol-fixed; thick films should be unfixed

blood collected into EDTA

N.B. for optimal sensitivity of qPCR, please send:
adults: at least 10 millilitre EDTA blood
neonates / babies: 1-2 millilitre EDTA blood
older children: 1-2 millilitre EDTA blood if using paediatric bottles or 10 millilitre EDTA blood if using adult bottles (preferred)

All samples should be accompanied by a completed *T. cruzi* qPCR form

Worms - general

Diagnosis: macroscopical examination of adult worms and segments
For example *Ascaris lumbricoides, Enterobius vermicularis,Taenia spp., Diphyllobothrium latum*

formol-ether concentration of faeces and microscopy for ova and larvae

Specimens: faeces in plain container for adult worms, segments and concentration, minimum 1 gram faeces or 2 millilitre if sample is liquid

Whole worms and segments

If there will be a delay in sending, please put specimen in 70% alcohol in suitable container

4. Laboratory Schedule & Turnaround Times

Generally, specimen processing is begun on the day of receipt. Specimens that require microscopy only may have results available that same day whereas investigations processed in batches, those requiring culture or worm identification will take variable periods of time. As a reference laboratory some investigations are highly unusual and so target turnaround times **serve as a guide only**. When complete, final reports are produced and posted the same or next working day – interim or final telephone reports are always available upon request.

Turnaround time guideline:-
from receipt of specimen to release of report (telephoned or posted letter), in working days).

Intestinal Parasitology:-
Specimen processing takes between 1 and 7 working days, depending upon the range of investigations required for each specimen (for example, concentration, specific staining, microscopy, Techlab ELISA, amoebic culture, Stronglyloides isolation.

Acanthamoeba culture:-
Culture usually takes up to 7 calendar days; microscopy results, where applicable, available in the interim. All positive results telephoned in advance of final report.

Acanthamoeba PCR:-
Up to 10 working days. All positive results telephoned in advance of final report.

T. cruzi PCR
Suspected acute (neonate, travel, transplant-associated, needle-stick injuries) are treated as urgent and results are usually available within 1-2 working days. Please contact us to discuss the case before the sample is sent.
Chronic cases: assay run 1-2 times monthly, batched – please contact us if regarded as more urgent e.g. possible reactivation

Worm Identification
1-7 working days

Microscopy of stained/unstained specimens (other than faeces)
e.g. hydatid, blood parasites, pus: 1-3 working days depending on urgency

Entomology-
This is variable depending upon specimen. If your request is urgent, we advise that you contact us before sending the sample to discuss.

5. Charges

For current scale of charges please contact the laboratory for information.

Should there be any change to this, all laboratories will be given, wherever possible, a minimum of 2 months advance notice.

E. Results and Reports

1. Written reports

Reports are printed and dispatched by 1st Class Royal Mail each working day.

In most cases it can be assumed that the written report is final, however if further results are to follow, or if a repeat specimen is required, this will be clearly stated. Interim reports, where necessary, will normally be given by telephone and confirmed in a full and final written report.
Interpretation of results and comments on individual cases will be given where required.

2. Telephone reports

Results of urgent investigations, those which may aid immediate patient management, or any results specifically requested by the sending laboratory will be telephoned as soon as they become available.
The name and status of the person to whom results are given will be required for our records and we will ask for the results we have given to be read back to us in order to confirm correct communication. The Laboratory Director will telephone to discuss results where clinical interpretation or advice is required.

The results of some investigations may be rapidly available and to aid the management of certain infections will be telephoned immediately. Examples are:

- Primary diagnosis of malaria by microscopy and or molecular and immunochromatographic techniques
- *P. falciparum* or *P. knowlesi* where undiagnosed by the requesting laboratory
- *T. cruzi* PCR positive
- Trypanosomes in blood films
- Diagnosis of *E. histolytica* by microscopy and antigen–specific ELISA
- *Giardia intestinalis*
- Hydatid
- Babesia
- *T. solium*
- Any other pathogen where prompt initiation of treatment is considered necessary.

All telephoned reports, whether initiated by the reference laboratory or the requesting laboratory, will be confirmed with a written report.

3. Archiving of reports & security of information

All reference laboratory copies of written reports (to which the original request form is attached) are held in secure, locked storage for a minimum period of 5 years.

All staff have a duty of patient confidentiality and as part of the induction process are aware that all patient-related information is confidential and all data is held in accordance with the Data Protection Act 2018 and GDPR. Staff should adhere to the School's Data Protection policy and related guidance. It is a mandatory requirement that all staff undertake the LSHTMs on-line training and guidance in security of information and GDPR.

4. Obtaining information and results

Staff are always available during laboratory opening hours to discuss results and to give advice and information.

Please contact the laboratory on 020 7927 2427 from where queries can be answered or referred to appropriate personnel.

Updates to the Handbook

Please note that this guidance is valid on the day that you print or download it. Always check for the latest version at www.parasite-referencelab.co.uk