DIAGNOSIS OF INFECTIONS

Bacterial infections affect the skin; the eye; the ear; the mouth; the nose the reproductive system the digestive system the respiratory system the urinary system the nervous system the circulatory system the locomotion organs

Why diagnosis is needed?

- To administer the treatment
- For prognosis
- To initiate appropriate control measures
- To take suitable preventive steps
- To understand epidemiology
- To know the disease history
- For certification in International trade
 - To export
 - For import
- To know who is at risk



Steps in Diagnosis of Bacterial diseases

- Clinical Signs
- Laboratory examination
- 1- Microscopy
 - 2- Culture techniques
 - **3- Biochemical reactions**
 - 4- Serological identification:
 - 5- Molecular biology techniques
 - 6- Bacteriophage typing

Pre-requisite for laboratory Examination

- Suitable sample
- Proper dispatch of sample to reach the laboratory along with all the relevant history of disease (morbidity, mortality, contagiousness etc.), signs and treatment.
- In-time arrival at Laboratory
- Proper laboratory facility
- In-time processing at the Laboratory by the trained personnel

Site of sampling

Sterile sites

- Blood
- Cerebrospinal fluid (CSF)
- Body fluids (Peritoneal and pleural)

Non-sterile (normal flora)

- Respiratory tract
- Ear, eye and mouth
- Skin (wound and abscess)
- Urine (mid-stream)
- Feces

Blood/serum/plasma samp

- SST (red-gray or gold top). Contains a gel separator and clot activator.
- Sodium heparin (dark green top)
- PST (light green top). Contains lithium heparin anticoagulant and a gel separator.
- EDTA (lavender top)
- ACDA or ACDB (pale yellow top). Contains acid citrate dextrose.
- Oxalate/fluoride (light gray top)



Precautions during blood sample collection

- Patient should be allowed to sit straight with site of specimen collection directly towards you.
- The surface should be thoroughly cleaned with a ethanol swab.
- Quantity of the blood drawn should be according to requirements
- Single needle prick can be used for collection of sample for multiple voiles
- Immediately after collection sample should be properly labelled
- Sample can be stored at 4oC for maximum 3 days, however immediate processing is preferred

Urine collection



Precautions for urine sample collection

- collect your pee (urine) sample in a completely clean (sterile) container
- store it in a fridge in a sealed plastic bag if you can't hand it in straight away
- A mid-stream urine sample means you don't collect the first or last part of urine that comes out. This reduces the risk of the sample being contaminated with bacteria from:
- your hands
- the skin around the urethra, the tube that carries urine out of the body
- Do not keep it for longer than 24 hours.

Atelogical agents Uncomplicated UTI Pathogens

Escherichia coli Staphylococcus saprophyticus Klebsiella spp. Enterococcus faecalis Complicated UTI Pathogens

Escherichia coli Klebsiella spp. Enterobacter cloacae Serratia marcescens Proteus mirabilis Pseudomonas aeruginosa Enterococcus faecalis Group B streptococci

RESPIRATORY TRACT INFECTION

What are upper RI

- Common infections are localised in oropharynx, nasopharynx and nasal cavity
- Causes
 - Sore throat, nasal discharge and often fever
 - Larynx infection, otitis media, sinusitis, conjunctivitis or keratitis
 - May present with serious disease whooping cough, influenza, measles and infectious mononucleosis
- Mostly by viruses which are difficult to isolate
- Mostly caused as secondary infection my opportunistic normal flora
- Drug resistant bacilli or yeasts may dominate throat flora in patients receiving antibiotics

Aetiological agents

Antibody deficiencies

Encapsulated bacteria (Streptococcus pneumoniae, Staphylococcus aureus, Haemophilus influenzae, and Moraxella catarrhalis)

Other bacteria (Bordetella pertussis)

Opportunistic bacteria

(Pseudomonas aeruginosa)

Atypical bacteria (Mycoplasma and Chlamydophilla)

Fungi (Aspergillus and Scedosporium) → hyper-IgE syndrome

Viruses (rhinovirus, herpes simplex virus, and cytomegalovirus) T- and B-cell combined immunodeficiencies

Encapsulated bacteria (Streptococcus pneumoniae, Staphylococcus aureus, Haemophilus influenzae, and Moraxella catarrhalis)

Viruses (respiratory syncytial virus, adenovirus, parainfluenza 3, paramyxovirus, and cytomegalovirus)

Opportunistic bacteria (Pseudomonas aeruginosa and Pneumocystis jiroveci)

Atypical mycobacteria (including BCG vaccine strain)

Fungi (Aspergillus, Scedosporium, Candida, Histoplasma, and Cryptococcus) Phagocytic immunodeficiencies

Catalase-positive microorganisms (Burkholderia, Pseudomonas, Serratia, and Staphylococcus aureus)

Fungi (Aspergillus, Nocardia, and Candida)

Atypical mycobacteria (including BCG vaccine strain) Complement immunodeficiencies

Encapsulated bacteria (Neisseria sp. and

(Neisseria sp. and Streptococcus pneumoniae)

Etiological agents of respiratory infections according to the PIDs category.

Specimen collection

- A plain cotton wool swab used
- Sampling from tonsils, posterior pharyngeal wall or inflamed areas
- Swab should b rubbed with rotation over the area
- Adequate view of throat shod be ensured
- Swab should be immediately replaced in its tube and transported (or stored at 4oC)



shutterstock.com • 1116273128



- Deep nasal samples collected with thin special swabs
- For sinusitis pus/ nasal aspirate or saline wash should b examined
- Swab can be used to collect discharge from external meatus of patient's ear infected with chronic otitis





Specimen for lower RI

- Sputum most common which is mixture of bronchial secretions and inflammatory exudates coughed up into mouth and expectorated
- Busy and uninstructed staff may send collection of saliva
- Repeat samples may b required occasionally
- Collection in 50-100ml caped plastic container
- Collection after the patient wakes up is preferable
- Transportation must be in sealed bags



WOUND SPECIMEN

Pus sample collection

- Wound infections may be caused by one to many organisms depending on the site of the infection.
 - dermatophytes infections in the keratinized layer;
 - superficial skin infection by aerobes
 - only anaerobes are commonly isolated from abscesses of the perennial, inguinal, and buttock area,
 - whereas non-perennial infections are caused by mixed facultative aerobic organisms.
 - postoperative wound are often infected with a mixture of aerobes and anaerobes

- the ideal specimen is an aspirate from a previously undrained abscess,
- or a tissue biopsy.
- a minimum volume of 1mL (up to 5 mL) of pus should be collected. Large volumes of purulent material maintain the viability of anaerobes for longer.
- should be collected in a sterile syringe – any air bubbles should be expelled.
- Needle safely and tightly capped





Type of microorganisms	Na	%
Coagulase-negative Staphylococcus	62	43.4
Staphylococcus aureus (MSRA= nº17-11.9%)	41	28.7
Klebsiella pneumoniae	18	12.6
Enterococcus faecalis	17	11.9
Enterobacter aerogenes	12	7
Enterococcus ssp	7	4.9
Pseudomonas aeruginosa	5	3.5
Acinetobacter ssp	5	3.5
Morganella morganii	3	2.1



CEREBROSPINAL FLUID

CSF collection

- Lumbar puncture done to collect sample by traned physicians
- 3-5ml collected
- Best site is interspace between 3 and 4 lumbar vertebrae



© The Reyal Children's Hospital, Melbourne, Australia 2012 Kids Health Info www.rch.org.au Illustration: RCH Educational Resource Centre

Etiological agents

- 1. Herpes simplex virus (HSV) types 1 and 2,
- 2. Mumps virus,
- 3. Enterovirus,
- 4. Varicella zoster virus (VZV),
- 5. Streptococcus pneumoniae,
- 6. **HiB**
- 7. Neisseria meningitidis.

Important steps in specimen

- particularly checks to ensure that the correct specimen has been sent
- the name on the specimen is the same as that on the request form.
- how to handle and store specimens that require immediate attention, e.g. c.s.f., blood cultures, unpreserved urine, swabs not in transport media, faecal specimens containing blood and mucus, and wet slide preparations.
- Examples of specimens which should not be accepted for microbiological investigations include:
- dry faecal swabs,
- saliva instead of sputum,
- eye swabs that have not been freshly collected,
- any specimen not collected into a correct container,
- a leaking specimen (sample may be contaminated).