Lecture outline – 2018: Liver infections – microbiology aspects

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Infections of the Liver

Acute hepatitis is most commonly the result of virus infection: A, B, C, D and E are possible agents with A, B, C most common. Hepatitis also occurs during EBV infection, CMV infection and rarely as a result of HSV infections, and regularly in yellow fever. Infection with leptospirae and *Coxiella burnetti* are associated with abnormal liver function tests. Liver may also be involved in septicaemia due to pyogenic bacterial infections and infections caused by parasites such as *Entamoeba histolitica.*

Viral Infections

Infection by Primary hepatitis viruses

1. Hepatitis A (common)
2. Hepatitis B (common)
3. Hepatitis C
4. Hepatitis D
5. Hepatitis E

Infection by Primary hepatitis viruses

1. Cytomegalovirus disease
2. EBV infection
3. HSV infection (rarely)
4. Yellow fever

Infection by other viruses as a part of the course of illness

Bacterial infections

1. Leptospirosis
2. *Coxiella burnetti* infection (Q fever)
3. Pyogenic bacterial infections causing liver abscesses

Fungal infections

1. Invasive candidiasis in immune-compromised individuals.

Other liver Infections

1. Amoebic abscess
2. Hydatid disease

Route of spread & Risk groups of main primary hepatitis viruses

Virus	Route of spread	Risk groups
Hepatitis A	Faecal-oral	1. Those who eat or drink contaminated food e.g. shellfish and water 2. Travelers to areas of high endemicity with low hygiene standards 3. Careers or contacts of cases of acute HAV.
Hepatitis B	Vertical (i.e. Mother to baby) Sexual Contact with blood and blood products	1. Newborns of carrier mothers Members of ethnic groups with high carriage rates 2. Sexually promiscuous, both heterosexual and male homosexuals 3. Intravenous drug abusers who9 share needles 4. Patients receiving or exposed to blood or blood products e.g, Haemophiliacs 5. Healthcare workers exposed to blood or blood products
Hepatitis C	Contact with blood and blood products Natural route of transmission unknown Sexual transmission (inefficient compared to HBV)	1. Intravenous drug abusers who9 share needles 2. Patients and Healthcare workers receiving or exposed to blood or blood products as per HBV
Hepatitis D	Can only infect simultaneously with HBV or Patients who are already HBV infected Therefore routes as for HBV	As per HBV
Hepatitis E	Faecal-oral	Travellers to HEV endemic areas

Microbiological diagnosis of acute infective hepatitis

Specimen collection and transport:

Potential risk in handling specimens to lab staff specimens should be marked as **'Risk of infection'** to ensure safe transport.

1. Blood for Standard Investigations:

- Full blood count
- Liver function tests

2. Blood for microbiology (HAV, HBV & HCV serology) to exclude HAV and HBV infection

Diagnosis of HAV and HBV relies on detection of viruses or of antibodies to viruses using serological techniques.

Both viruses cannot be grown in routine tissue culture

Not useful to send a faecal sample to the laboratory for HAV diagnosis.

Interpretation of lab reports:

Hepatitis A

Presence virus specific IgM antibodies to HAV in a serum sample is a definitive indication recent HAV infection (acute HAV infection). IgM only present for a short time following infection

Presence of virus specific IgG antibodies only indicates patient has been infected with HAV some unspecific time during his/her life

Hepatitis B

Diagnostic assays available for HBV infection based on detection of virus antigens or virus specific antibodies (hepatitis B markers) in the laboratory.

Antigen	**Antibody**
Hepatitis B core antigen (HBcAg)	Anti-HBc
Hepatitis B e antigen (HBeAg)	Anti-HBe
Hepatitis B surface antigen (HBsAg)	Anti-Hbs

'e' is for extractable. eAg is a breakdown product of cAg.

HBV replication in hepatocytes release excess surface protein spills over into blood stream.

Therefore, primary screening assay for HBV infection is detection of **HBsAg in blood**.

Presence of HBsAg in blood indicate patient is infected with HBV.

Laboratory confirmation recent HBV infection:

Best serological marker of recent infection is the presence of IgM class antibodies to the virus

For HBV, standard diagnostic assay to look for the presence of **IgM against HBV core antigen**

Significance and usefulness of other HBV markers:

Purpose of testing HBeAg and anti-HBe:

eAg is a breakdown product of core antigen.

It is a surrogate marker of the extent of HBV replication occurring in the liver

The 'e' status of all carriers should be determined.

Interpretation of HBeAg and Anti-HBe results:

(i). HBVeAg positive: anti-HBe negative:

Extensive HBV replication occurring in the liver with spill over into serum.

Serum extremely infectious, e. g. via needle-stick injury

Increased risk of long term liver damage

(ii). HBe Ag negative, Anti-HBe positive

Low level replication in liver

Serum very much less infectious, e.g. transfusion of unit of blood needed to transmit infection

Reduced risk of long term liver damage

(iii). HBe Ag negative, Anti-HBe negative

In process of seroconverting to eAg (i.e. will become Anti-HBe positive)

Regard as HBeAg positive individuals until Anti-HBe is detectable

Refer: Serology results obtained from various categories of patients infected with HBV

Note:

HBeAg and anti-HBe are only surrogate markers of what is going on in the liver of HBsAg carriers. More exact measure is the presence and quantity of HBV DNA in serum. This assay not routinely available.

Hepatitis C

Detection of antibodies to HCV indicate infection

Detection HCV RNA in serum by PCR indicate chronic carriage

Hepatitis D

Detection of delta antigen, anti-delta antibodies

Hepatitis E

HEV serology

Assay not widely available.

Diagnosis by exclusion

Cytomegalovirus (CMV):

Sample of blood → rapid diagnostic techniques

Presence of replicating CMV in peripheral blood correlates much better with CMV induced disease.

1. Detection of early antigen fluorescent foci (the DEAFF test). CMV infected may remain morphologically normal for many days but express viral antigen on cell surface. Inoculate patient's material to cell cultures. Presence of virus 24-48 hours later by staining cells with FITC labeled Mabs specific for early antigens (express early in the CMV replication cycle)of CMV.
2. Direct CMV Ag detection. Peripheral blood cells taken directly from the patient are stained with FITC labeled Mabs to detect CMV Ag expression occurring *in vivo*.
3. CMV genome detection. Presence of the CMV genome in peripheral blood by PCR method.

4. Routine tissue culture which is the standard assy for demonstration of CMV inappropriate as it take up to 4wks for the virus to produce CPE.
5. A rise in antibody titre to CMV in paired samples of sera.

Epstein-Barr virus infection (EBV) → infectious mononucleosis

Specimens:

(A). Blood sample for (i). full blood count including

differential (lymphocytosis, i.e. 53% and

film (atypical lymphocytosis)

(ii). Liver function tests, i.e. ALT raised i.e. 86U/L (normal range = 5-50)

indicate hepatic involvement

(iii). 'monospot ' or Paul Bunnel test (detect presence of heterophile antibodies)

Acute EBV positive for Monspot/Paul Bunnel test but 10% individual negative. Also CMV, Toxoplasma, HIV induced IM are negative.

(iv). Viral serology

Demonstration of EBV specific IgM in acute serum sample

Demonstration of EBV antigens EBNA, LMP.

(B). Detection of EBV genome by *in situ* hybridization

Leptospirosis:

Spirochaetes, *Leptospira interrogans***.**

Contact with rodent urine, livestock, contaminated water (farming, water sports)

Investigation: Sample of blood for serology.

(i). Rise in antibodies in paired serum samples

(ii). IgM antibodies to the organism

Liver Abscesses:

What microbes cause liver abscesses?

Most are pyogenic and caused by bacteria.

1. *Streptococcus milleri*
2. *Staphylococcus aureus*
3. *Bacteroides* species
4. Coliforms such as *Escherichia coli* originating from the large bowel
5. Fungal infections of the liver usually found only in severely immunocompromised such as bone marrow transplant patients. i.e. invasive candidiasis
6. Entamoeba histolitica→ liver involvement following amoebic colitis may occur in patients or travelers from endemic areas.

Microbiological Investigations:

Pyogenic abscesses:

Specimens: purulent fluid, if systemic involvement blood for culture, swabs of the pus. Actual pus or tissue is better than swab when isolating anaerobes. Pus may be collected and forwarded to the lab in a sterile container or a syringe sealed with rubber plug.

Gram staining: gives an early idea of likely pathogen

gram positive cocci in chains→ Streptococci (likely pathogen *Streptocci milleri*)

Amoebic liver abscesses:

Eosinophilia in 80%.

Amoeba may be seen in biopsy of edge of abscess. Rarely seen in aspirated fluid.

Rise in antibodies.

Hydatid disease:

abdominal mass/liver abscess (occupation may be relevant)

Ultra sound but not aspiration

serology