**King Saud University**
**Collage of Science**
**First Semester**
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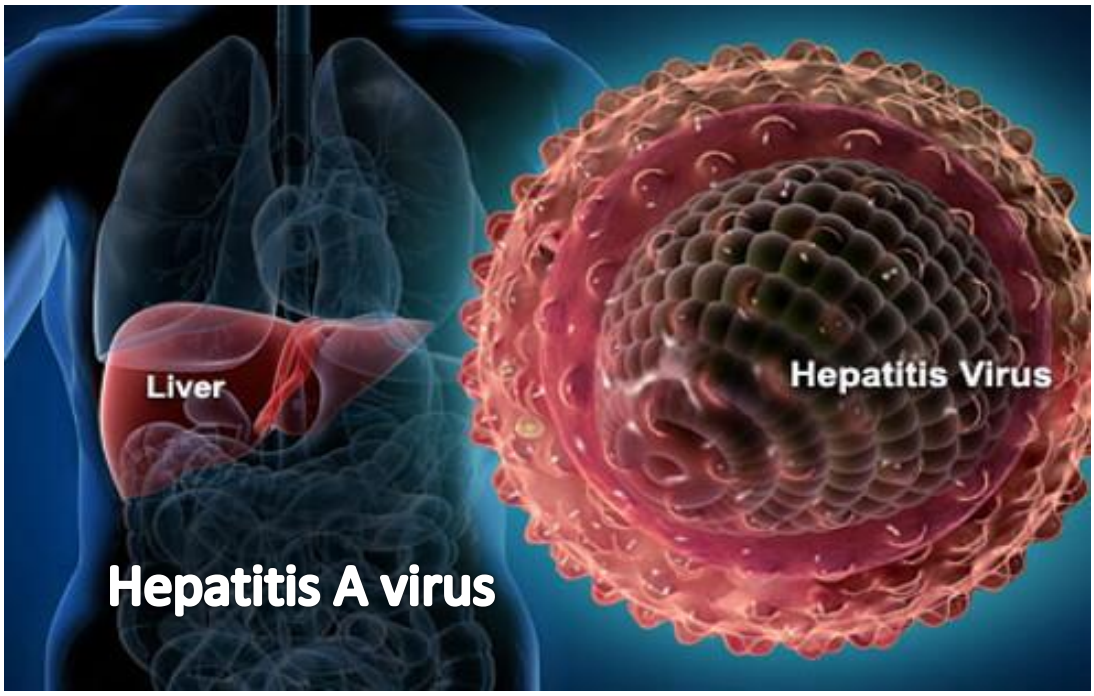
# A research of course medical virology – practical (MIC 450)



# *Topic: Hepatitis A Virus*

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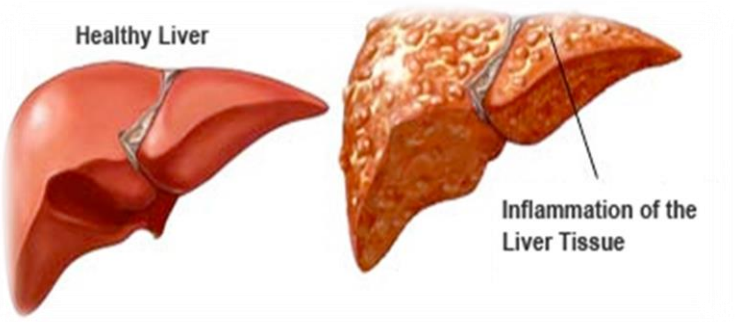
# I.INTRODUCTION:

Hepatitis is an inflammation of the liver. An inflammation is the body's response to injury or irritation. Hepatitis viruses damage a liver so badly that it is permanently scarred and can no longer repair itself. Hepatitis viruses generally do not destroy the living cells in which they replicate (Conne-Goldsmith.2011).

Hepatitis A is the most common form of acute viral hepatitis worldwide. The first description of hepatitis (epidemic jaundice) is generally attributed to Hippocrates and outbreaks of hepatitis A have been recognized for centuries, affecting both military and civilian populations (Conne-Goldsmith.2011). Hepatitis A infection is caused by the hepatitis A virus (*hepatovirus*). HAV is excreted in feces, and infection mainly occurs through the fecal-oral route either by direct contact with an HAV-infected person or by ingestion of contaminated drinking water and food (Gloria-Sánchez.2013)

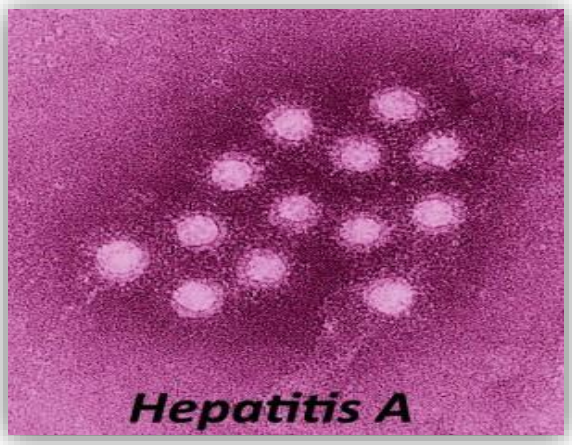
HAV replicates in *hepatocytes* and interferes with liver function, sparking an immune response that causes liver inflammation. HAV is acquired by the fecal-oral route. Person-to-person transmission is common and generally limited to close contacts (Conne-Goldsmith.2011).

Recently, 70 cases of hepatitis A in seven states have been linked to tainted frozen strawberries shipped to the United States from Egypt and served by a popular smoothie restaurant chain. As of now, 32 people have been hospitalized, but no deaths have been reported. Symptoms can take up to 50 days to appear The CD.C. Traced the infection to frozen strawberries imported from Egypt, and said it was not aware of other restaurants that may have received infected strawberries. Tropical Smoothie Cafe said it had switched to another supplier for all its restaurants nationwide, and would use only strawberries sourced from the Americas.



## II.CLASSIFICATION OF THE VIRUS:

| | |
|---|---|
| **Order** | *Picornavirales* |
| **Family** | *Picornaviridae* |
| **Genus** | *Hepatovirus* |




## III.STRUCTURE AND GENOME:

| | |
|---|---|
| **Shape** | An icosahedral symmetry is a 3-dimensional figure with 20 faces resembling a soccer ball. |
| **Size** | Small 27 to 30-nm diameter |
| **Enveloped or not** | Non-enveloped. |
| **Nucleic acid** | Positive-stranded RNA genome. |

(Zuckerman AJ. 1996), (Harvard university)

**The viral structure is further described below:**

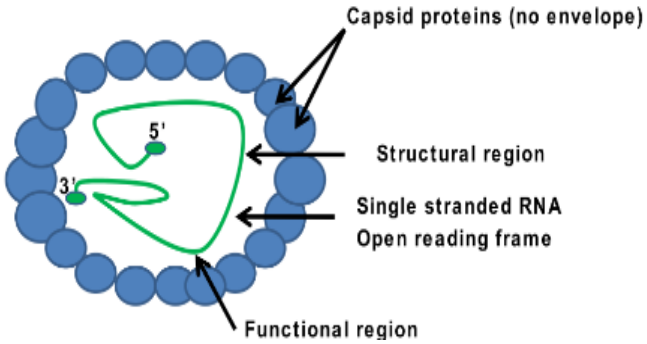



**Figure.1:** The internal structure of hepatitis A virus showing capsid proteins and envelopes, structural region, single stranded RNA (open reading frame) and functional region. (Zahid-Hussain.2001)

## ➢ Capsid

The capsid of HAV is made up of subunits called capsomeres as shown in Figure2. Each capsomere is made up of five protomers. Each protomer of HAV is made of three proteins; VP1, VP2, and VP3, which have a role in cell entry.

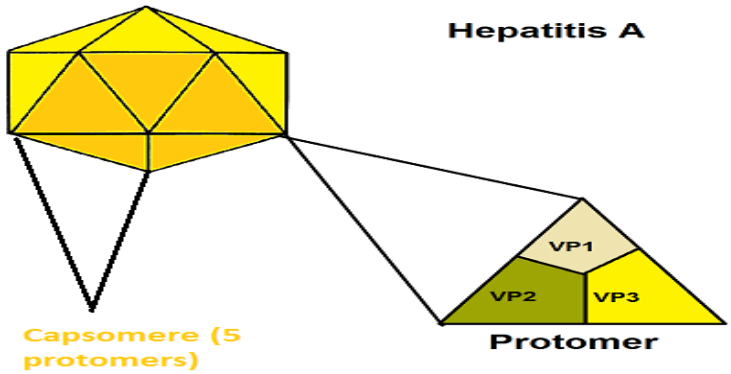



**Figure.2:** capsid of the virus is made of subunits called protomers. Each protomer is made of three proteins.

## ➢ Genome:

The genome has an attached protein VPg that acts as a primer for copying the genome (replication). As shown in Figure3, the genome is divided into three segments: P1, P2, and P3. The first segment encodes genes to make capsid proteins. The other two segments make other genes needed for replication (Jiang P, Liu Y, Ma H-C, Paul AV, Wimmer E.2014).


**Figure.3:** The genome of HAV is a single strand of RNA that is divided into three sections.

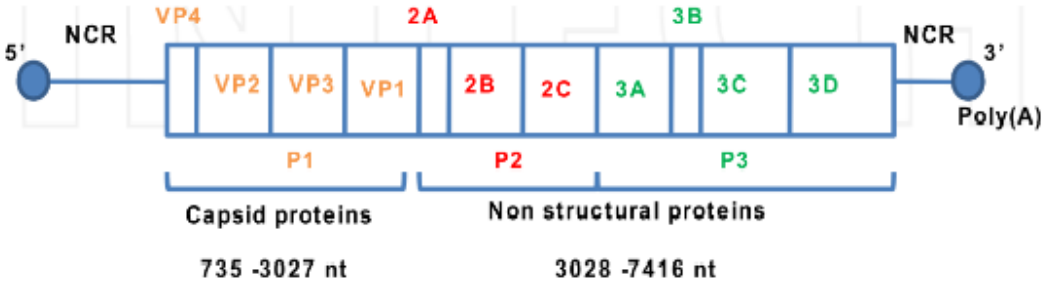



**Figure.4:** Genomic structure of hepatitis A virus. HAV genome is divided into a 5' non-coding region (5' NCR), a giant open reading frame, and a non-coding region (3' NCR). The coding region is subdivided into regions P1, P2 and P3. (Zahid-Hussain.2011)

## IV.PROTEINS (VIRULENCE FACTORES):

- **4 structural proteins (VP1, 2,3,4)**
  - Capsid Polypeptides
  - Anti-HAV binds VP1 and/or VP3
- **7 non-structural proteins (2A,B,C; 3A, B, C, D)**
  - 3C (protease)
  - 3D (RNA Polymerase)

(Harvard University)

The HAV proteins is organized into three regions, named P1, P2, and P3. The viral capsid protein (P1) is further divided into VP4, VP2, VP3 and VP1 regions. while the non-structural protein P2 and P3 polyprotein regions are divided into 2A, 2B, 2C and 3A, 3B, 3C, 3D respectively (Figure 4). HAV non-structural polyprotein contain the enzymes and accessory proteins essential for viral replication. It is processed into precursors intermediates, and mature proteins by the proteolytic activities of encoded viral proteins.HAV 2A, 2B, 2C protein encodes 45, 251 and 335 amino acids respectively. The 2A and 3C are identified as processing enzyme in hepatitis A virus. The translated 2A regions function as intermediary, partially located on the surface (VP1) and some are assembled inside the virion. Both 2B and 2C proteins play an important role in the replication of the viral RNA. P3 polyproteins encodes 3A, 3B, 3C and 3D proteins with 74, 23, 219 and 489 amino acids respectively, protein 3A have sequences that integrate into the host membrane bilayer and appear to be involved in both the rearrangement of the target membranes during infection and the tethering of the RNA replication complex to these membranes. The 3C protein is the sole processing enzyme, and the primary cleavage takes place at the 2A/2B site. While 3D is the RNA dependent RNA polymerase. Some mutations in 2B, 2C and 3A proteins are identified to enhance viral replication or to induce cytopathogenic effects in the viruses adapted to cell cultures. The VP4 is very small and not detected in the mature virion. (Vives-Adrián L, Garriga D, Buxaderas M, Fraga J, Pereira PJB, Macedo-Ribeiro S.2015, Totsuka A, Moritsugu Y.1999, Zahid-Hussain.2011).

## V.TRANSMMISSION:

Hepatitis A is primarily transmitted via the oral-fecal route, which means that something we eat or drink has been contaminated by feces infected with the virus. Once ingested (taken in via food or drink), the hepatitis A virus passes through the stomach and into the intestines. From there, the virus enters the blood. The portal vein carries the infected blood from the intestines to the liver. Once in the liver, hepatitis A viruses invade the hepatocytes. The viruses start to churn out new viruses. Unlike most viruses, hepatitis A does not burst from the cell and destroy it. Instead, the hepatocytes expel the new "daughter viruses" into the bile. The bile is released into the stool, which then carries huge quantities of the virus out of the body. Those viruses can then infect the next unsuspecting person. The hepatitis A virus is very hardy. It can live in water as hot as 140°F (60°C). Can also survive in temperatures below freezing (32°F, or 0°C). The hepatitis A virus can survive on surfaces such as bathroom and kitchen counters for more than a month at normal room temperatures. It can live in both freshwater and salt water for many months.

**People become infected with hepatitis A through:**

- Close contact with family members or while traveling to areas with poor sanitation. People can get hepatitis A when infected people do not thoroughly wash their hands after going to the bathroom.
- Child care workers change diapers, they may pass the virus from one baby to another.
- Toddlers who do not wash their hands properly can easily pass hepatitis A to playmates and caregivers.
- Contamination can occur during processing and distribution of food or during preparation at home or in restaurants (eat or drink food or water that has been contaminated by stools (feces) containing the hepatitis A virus)
- Fruits, vegetables, shellfish, ice, and water are common sources of the disease.
- Raw or incompletely cooked shellfish such as oysters, clams and mussels have a particularly high incidence of transmitting the Hepatitis A virus because they live in bodies of water that may be polluted with HAV.

**The following groups are considered to be at risk of getting HAV:**

- Men who have sex with men.
- People who share needles or syringes when they inject drugs.
- People with certain blood-clotting problems.
- People with job-related risks, such as doctors, nurses, paramedics, scientists who work with primates, and laboratory workers who handle infected blood or stools.

**The virus is more easily spread under poor sanitary conditions, and when good personal hygiene is not observed (**Conne-Goldsmith.2011)

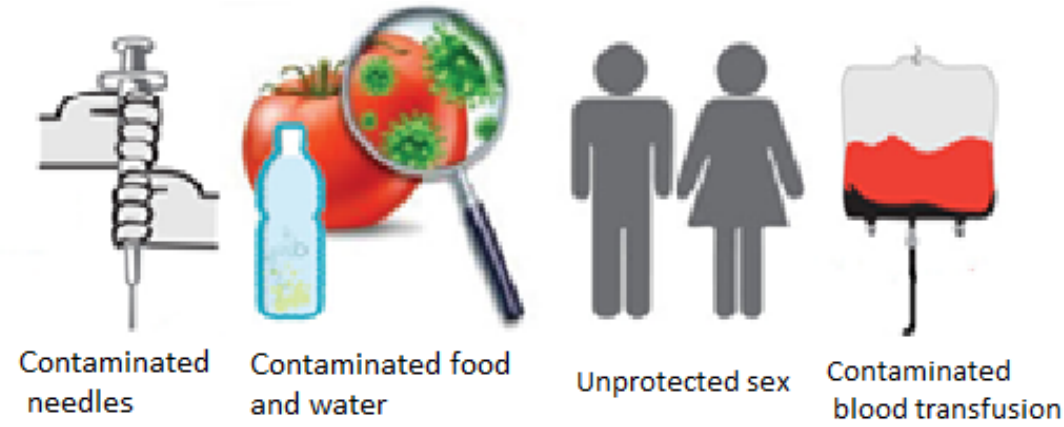



**Figure.5:** Transmission of HAV

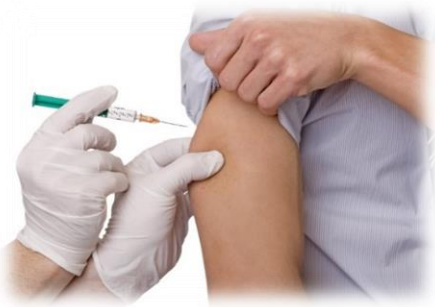
# VI.PREVENTION:

Improved sanitation, food safety and immunization are the most effective ways to combat HAV. Basic good hygiene can go a long way toward preventing infection with hepatitis A (Conne-Goldsmith.2011).

**To Avoid Transmission of HAV:**

- People should wash their hands for at least thirty seconds with plenty of soap and warm water, after going to the bathroom, changing diapers, playing with young children, and before preparing food.
- People who prepare food for others in restaurants should wear gloves.
- Vigorously washing raw produce in tap water greatly reduces the risk of contracting hepatitis- A.
- Vigorously washing raw produce in tap water greatly reduces the risk of contracting hepatitis- A.
- Vaccination

**The best way to prevent Hepatitis A is by getting vaccinated**



# VII.REPLICATION CYCLE:

HAV enters the cell through endocytosis, replication, translation and assembly occur in cytoplasm of infected cells (shown in figure 4). (Zahid-Hussain.2011)

**The steps of Replications cycle for HAV include:**

- **Attachment:** Binding of the virus to a cell surface receptor. This interaction leads to virus internalization and destabilization of the capsid, which allows the release of the genome from the endosome into the cytoplasm (Bergelson JM. 2010, Levy HC, Bostina M, Filman DJ, Hogle JM. 2010).

- **Penetration:** the process of entering the virus to a host cell (Zahid-Hussain.2011).

- **Uncoating:** process whereupon the viral capsid is removed in order to release the viral nucleic acid into the host (Zahid-Hussain.2011).

- **Replication:** process where by a virus uses its host to copy its genome, generate and assemble a protein capsid, and thereby reproduces itself (Inside the cell the virus uncoats, releases viral RNA and begins transcription). The newly made viral RNA can be used as either mRNA for translation, a template for replication, or a substrate of encapsidation into a protective coat as soon as enough genomes and capsid proteins have been synthesized (Zahid-Hussain.2011).

- **Assembly:** The capsid is assembled, and the genome is packaged (Zahid-Hussain.2011).

- **Release:** The virus is released through cell lysis, which disrupts the host cell's membrane (Zahid-Hussain.2011).

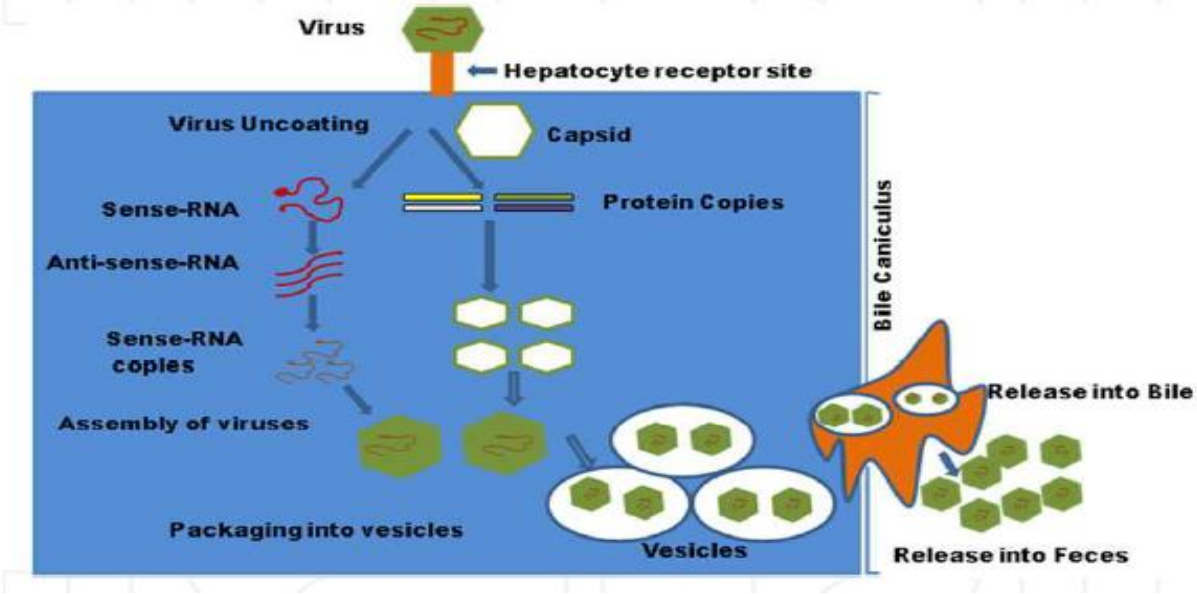



**Figure.6:** Diagrammatic representation of life cycle and replicative phase of hepatitis A virus.

## VIII.SYMPTOMS:

Hepatitis A virus; now preventable by vaccine. Most people recover and the infection do not become chronic. Most people do not have any symptoms of hepatitis A.

**If symptoms of hepatitis A occur, they include:**

- feeling tired
- muscle soreness
- upset stomach
- fever
- loss of appetite
- stomach pain
- diarrhea
- dark-yellow urine
- light-colored stools
- yellowish eyes and skin, called jaundice

Symptoms of hepatitis A can occur 2 to 7 weeks after coming into contact with the virus. Children younger than age 6 may have no symptoms. Older children and adults often get mild, flulike symptoms. The patient should see a doctor right away if he or a child has symptoms of hepatitis A.

# IX.DIAGNOSIS:

Virological diagnosis of HAV infection could be made by the detection of HAV RNA (in serum, stool or liver) or HAV antigen (in stool) during the late incubation period or early symptomatic phase of hepatitis A, serological diagnosis by measurement of specific circulating antibody is more practical, less cumbersome and widely available.

The diagnosis of hepatitis A requires the detection of immunoglobulin M antibody to HAV (lgM anti-HAV) in a patient who presents with, or has recently had, clinical features of hepatitis (icteric or anicteric disease) or in an individual with inapparent, asymptomatic infection m whom serum aminotransferase elevations may be detected. In occasional individuals monitored because of exposure to the disease, serological confirmation of acute HAV infection may occur in the absence of detectable arninotransferase elevations. Sensitive and specific radioimmunoassays and enzyme-linked immtinoassays are available for the detection of total anti-HAV and the lgM component. Automated microparticle enzyme immunoassays which can detect both lgM anti-HAV and IgG anti-HAV appear to be more sensitive and equal in specificity to the current commercially available assays% Peak IgM anti-HAV levels are reached during the acute or early convalescent phases and decline thereafter, often disappearing by 3 to 4 months after the onset of illness.

However, IgM anti-HAV may persist for more than six months in as many as 25% of patients ~4. Persistence longer than 12 months is distinctly unusual: fiflsepositive reactions may be responsible for some instances. In contrast. IgG anti-HAV appears early but reaches peak levels during the convalescent phase and usually remains detectable, in slowly declining titres, for decades (Raymond S. Koff. 1992).

# X.TREATMENT:

Hepatitis A goes away on its own in most cases. Most people get well within a few months.

**Hepatitis A management:**

- Slow down. Cut back on daily activities until all of energy returns. Asp patient start to feel better take time in getting back to the regular routine.
- Drinking plenty of water to avoid dehydration. Fruit juices and broth are other good choices.
- Eating a healthy mix of foods, it is important for patient to get good nutrition.
- Make sure that doctor knows all the medicines taking by patient, including herbal products.

If hepatitis A causes more serious illness, patient may need to stay in the hospital to prevent problems while his/her liver heals.

**Vaccination:**

The HAV vaccine is the most important preventive strategy against HAV (Mena,Garcia- Basteiro et al. 2015)

**There are currently 2 types of HAV vaccines:**

- Liver attenuated.
- Formaldehyde inactivated vaccines (most widely used worldwide and are the only ones recommended for HIV-infected subjects).

There are several HAV vaccines currently available. Most are adjuvant with **aluminum hydroxide**:

- Havrix 1440
- VAQTA 50

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