

ISSN:3032-1085



Navigating the Immune Response Landscape of Hepatitis B and Hepatitis C Virus Infections

Saja Qasim Hammood

Department of biology, College of Science, Mustansiriyah University, Baghdad, Iraq 2Ministry of Education

Ellaf Abdulljabbar Jassim prf. Dr. Alia Essam Mahmood

Department of biology, College of Science, Mustansiriyah University, Baghdad, Iraq 2Ministry of Education

Ashwaq A. Kadhem

Department of biology, College of Science, Mustansiriyah University, Baghdad, Iraq 2Ministry of Education

Received: Feb 16, 2024; Accepted: Feb 28, 2024; Published: Mar 18, 2024;

Abstract: Both the hepatitis B virus (HBV) and the hepatitis C virus (HCV) are viral pathogens that primarily target the liver and can lead to chronic liver disease. The nature of the immune response that the host mounts can vary depending on the structural differences between these two viruses. HBV is a partially double-stranded DNA virus enveloped in an outer envelope with a relatively stable genome and limited antigenic variation. It consists of an inner nucleocapsid core containing viral DNA, viral polymerase, and core antigen (HBcAg). The envelope comprises surface antigens (HBsAg) important for viral entry and immune recognition. HCV is a single-stranded RNA virus with an envelope. It has a high degree of genetic diversity due to its error-prone RNA polymerase, resulting in multiple HCV genotypes and subtypes due to its high mutation rate. Both viruses activate innate immune responses, producing type I interferons (IFNs) and pro-inflammatory cytokines. HBV can actively fight these antiviral responses in several ways, such as by making viral proteins that mess up the innate immune signalling pathways. At the same time, HCV developed strategies to evade and modulate the host's innate immune system, allowing it to establish persistent infections. The adaptive immune response against HBV and HCV involves both humoral and cellular components. Antibodies against HBsAg (anti-HBs) are critical for viral clearance and protection. CD8⁺ T cells are also very important for controlling HBV infection because they find and kill infected hepatocytes. During infection, HCV-specific antibodies and CD4⁺ and CD8⁺ T cells are made. However, HCV can evade immune responses by rapidly mutating its surface proteins (e.g., E2) and modulating T-cell responses. It is important to note that the immune response to HBV and HCV is a complex and dynamic process that involves various factors beyond the structural differences described above. Host factors, viral load, viral persistence, and the interplay between innate and adaptive immune responses also significantly influence the outcome of the infection.

Keywords: Hepatitis B; Hepatitis C; Virus

Introduction

Hepatitis is an inflammation of the liver that gives rise to a broad spectrum of systemic manifestations and can cause death. It continues to be a global human health threat and economic burden. According to the World Health Organization (WHO), about 325 million people suffer from chronic hepatitis infection. There are seven strains of the

hepatitis virus [type A, B, C, D, E, F, G] (Netzler et al., 2016; Hoofnagle et al., 2012), all of which cause liver disease, but differ in some aspects, such as mode of transmission, severity of illness, geographical distribution, and prevention methods (Acorn et al., 1995; Rasche et al., 2019).

Hepatitis A virus (HAV) and hepatitis E virus (HEV) are responsible for a large proportion of cases of acute hepatitis. In contrast, hepatitis B virus (HBV) and hepatitis C virus (HCV) are the most common causes of chronic viral hepatitis. Hepatitis D virus (HDV) is a satellite virus that requires the envelope proteins of HBV for cell release and uptake (Hughes et al., 2011; Tu et al., 2017; Smith and Simmonds, 2018).

The host innate immune response is the first defense against virus invasion. During virus infection, the pathogen-associated molecular patterns (PAMPs) of the virus are recognized by host-pathogen recognition receptors (PRRs), which is the first step of antiviral innate immunity activation (Lester and Li, 2014; Streicher and Jouvenet, 2019).

Viral nucleic acid, which represents a major PAMP, can be recognized by various PRRs, including retinoic acid-inducible gene-I (RIG-I)-like receptors (RLRs), Toll-like receptors (TLRs), and nucleotide-binding oligomerization domain (NOD)-like receptors (NLRs), as well as cytosolic DNA sensors, such as cyclic GMP-AMP (cGAMP) synthase (cGAS), interferon (IFN) regulatory factors (DAIs), and interferon-gamma inducible protein 16 (IFI16). PRRs then recruit downstream adaptor proteins, which typically include mitochondrial antiviral-signaling protein (MAVS), TIR-domain-containing adaptor-inducing IFN- β (TRIF), myeloid differentiation primary response gene 88 (MyD88), and intracellular stimulator of IFN genes (STING), these adaptor proteins ultimately lead to the activation of the transcription factor nuclear factor-kappa B (NF- κ B) and interferon regulatory factor 3 (IRF3), subsequently inducing the expression of type I IFNs and pro-inflammatory cytokines, which contribute to the establishment of the “antiviral state” (Koyama et al., 2008; Wan et al., 2020; Carty et al., 2021).

The liver can mount a rapid and robust immune response under appropriate conditions as a critical frontline immune tissue. It is enriched with innate immune cells, including Kupffer cells (KCs), natural killer (NK) cells, natural killer T (NKT) cells, and hepatic dendritic cells [DCs] (Gao, 2016; Ringelhan et al., 2018).

Evidence suggests that hepatocytes are responsible for the biosynthesis of 80–90% of innate immune proteins, including complement components and secreted PRRs, and the liver can express membrane-bound PRRs, such as TLRs (Gao et al., 2008, 2011).

However, viruses have also evolved strategies to evade the innate immune response to optimize their replication capacity. Research regarding hepatitis viruses, in particular HBV and HCV, has identified several molecular mechanisms hepatitis viruses employ to evade the host innate immune system (Hong et al., 2015; Liu et al., 2015; Yi et al., 2015; Feng and Lemon, 2019; Wang et al., 2019).

Viral Hepatitis

Viral hepatitis is a systemic ailment causing liver inflammation and damage, often asymptomatic in the initial weeks. Acute hepatitis cases in both children and adults may self-resolve and are attributed to various agents, including hepatitis A, B, C, D, E, G, and F viruses (Jawetz et al., 2013; Saha et al., 2016). Notably, chronic forms of hepatitis B (HBV) and hepatitis C (HCV) frequently progress to cirrhosis, posing a risk of liver failure or cancer and contributing to significant morbidity and mortality. Among viral hepatitis types, HAV, HBV, and HCV are ubiquitous globally (WHO, 2019). Transmission occurs through diverse routes, including sexual, parenteral, and blood transfusion. Hepatitis manifests with fatigue, flu-like symptoms, dark urine, pale stool, abdominal pain, and loss of appetite (Saha et al., 2016; WHO, 2019).

Reports indicate that over 2.3 billion individuals worldwide are infected with hepatitis viruses, with an annual toll of 1.4 million deaths, particularly in cases progressing to cirrhosis and liver cancer (Jefferies et al., 2018). Chronic HBV infection affects over 350 million people globally, resulting in nearly 1 million deaths annually. Additionally, approximately 150-200 million individuals live with chronic HCV infections, contributing to around 350,000 deaths each year (Mohammed et al., 2019). Chronic HBV infection is recognized as a significant public health challenge, akin to HIV, tuberculosis, and malaria (Revill et al., 2019).

Hepatitis Viruses Classification

Hepatitis viruses, classified as A, B, C, D, E, F, and G, exhibit variations in their properties and transmission modes, as seen in Table 1 (Robotis and Boleti, 2008). HBV, HCV, and HGV are enveloped, while HEV and HAV are non-enveloped (Yates et al., 2011; Ranjbar et al., 2013; Netzler et al., 2016).

Their genetic constitution differs, with HBV having a partially double-stranded DNA genome and HAV, HCV, HEV, and HGV having positive single-stranded RNA genomes; HDV, in contrast, features a negative-sense single-stranded spherical RNA genome (Netzler et al., 2016; Murray et al., 2020; Netter et al., 2021).

Transmission routes differ, with HAV and HEV transmitted fecal-orally, while HBV, HCV, HDV, and HGV are

transmitted parenterally through blood contact and sexual intercourse (Hoofnagle et al., 2012; Carroll et al., 2015; Meo et al., 2010). HDV is a satellite virus found in the presence of HBV, either through co-infection or superimposed on chronic hepatitis B (Urban et al., 2021).

Pathogenicity varies, with HAV and HEV considered less severe, HDV-HBV co-infection as the most severe form of chronic viral hepatitis, followed by HCV and then HBV, HGV, and HFV are rare types of viral hepatitis (Rizzetto and Alavian, 2013; Moosavy et al., 2017).

Table 1: classification of Hepatitis Viruses

Virus	Envelope	Genome	Transmission	Pathogenicity	References
HAV	No	+ssRNA	Fecal-oral	Less Severe	(Netzler et al., 2016; Hoofnagle et al., 2012)
HBV	Yes	Partially dsDNA	Parenteral, Sexual	Severe	(Yates et al., 2011; Carroll et al., 2015)
HCV	Yes	+ssRNA	Parenteral, Sexual	Moderate	(Ranjbar et al., 2013)
HDV	No	-ssRNA	Parenteral, Body Fluids	Most Severe	(Netter et al., 2021; Meo et al., 2010)
HEV	No	+ssRNA	Fecal-oral	Less Severe	(Netzler et al., 2016; Hoofnagle et al., 2012)
HGV	Yes	+ssRNA	Parenteral, Sexual	Rare	(Yost et al., 2018)
HFV	-	-	-	Rare	(Not specified)

Hepatitis Viruses Structure

1. Hepatitis B Virus Structure

The hepatitis B virus is characterized by pleomorphic forms (three forms). The first form is filamentous (with 17–22 nm in diameter and length 50–230 nm), and the second is a spherical body with 17–22 nm in diameter composed of the lipid and protein that forms the surface antigen (HBsAg) and is created in excess through the life cycle of the virus, and both forms lacking a core (not infectious) (Cao et al., 2019) the third form is spherical with a double-shelled sphere with a diameter of 42–47 nanometers which is known as an infectious virion (Dane particle), it is characterized by being the largest form that contains a DNA molecule, in addition to many antigenic components, including hepatitis B surface antigen (HBsAg) which are heterogeneous antigens made up of proteins of varying size, the small one S-HBsAg has only the S domain, and the middle M-HBsAg contains the PreS2 and S domains, while the large L-HBsAg has the PreS1, PreS2 and S domains (Tan and Ho, 2014).

In addition to HBcAg, the icosahedral nucleocapsid (which consists of 180 capsomers HBcAg with a diameter of 30–35 nm) has a function in the replication of the virus and is considered the main factor for infection of the cell that surrounds the viral DNA genome and their polymerase (Coppola et al., 2015), while hepatitis B e antigen (HBeAg) is secreted in the blood and the presence of both these proteins acts as markers of viral replication however the antibodies to these antigens are markers of declining replication. The genome of HBV is a relaxed circular double-stranded DNA (rcDNA); the full length of the genome is 3020–3320 nucleotides long (the 5' end of the full-length strand is linked covalently to the viral DNA polymerase), and the short length strand is 1700–2800 nucleotides long (Ali et al., 2009), the viral genome encodes 4 overlapping open reading frames (ORFs: S, C, P, and X) the S ORF encodes to the HBsAg (LHBs, MHBs, SHBs envelope proteins), the C ORF (core or C gene has the pre-core and core regions) encodes for the HBcAg, HBeAg respectively, while P ORF encodes to the polymerase (reverse transcriptase), and X ORF encodes to the HBxAg (16.5-KD protein) which has multiple functions, including signal transduction, DNA repair, transcriptional activation, and inhibition of protein degradation (Kosinska et al., 2017).

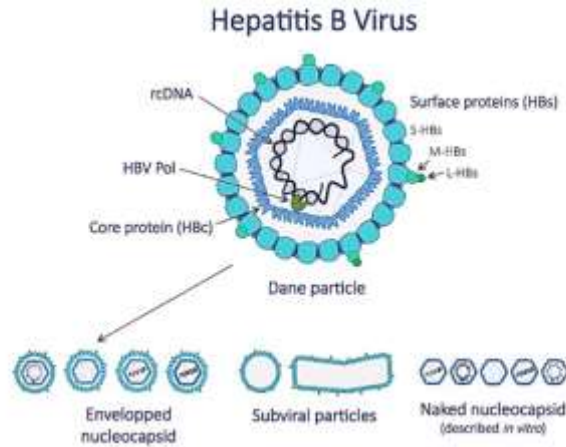


Figure 1-1: Hepatitis B virus (HBV) structure (Hu et al., 2017)

2. Hepatitis C virus structure

The hepatitis C virus is an envelope spherical virus containing positive sense single strand RNA surrounded by capsid form complex protein called nucleocapsid all of the components covered by envelop (Gastaminza et al., 2010), it's a glycoprotein which plays a major role in the process of entering the virus into the cell through its association with cell receptors such as heparan sulfate proteoglycan syndecan-1 or syndecan-4, CD81, scavenger receptor type B class 1 protein (SRB-1) and high-density lipoprotein (HDL) binding molecule very low-density lipoproteins (VLDL), and apolipoproteins (Apo) A1, B, C, and E (Vercauteren et al., 2014; Morozov and Lagaye, 2018) finally, the lipid membrane is derived from host lipid membrane component and is composed primarily of cholesterol, cholesteryl esters, phosphatidylcholine, and sphingomyelin, and the last two play a role in the entry of HCV into host cells (Blaising and Pêcheur, 2013).

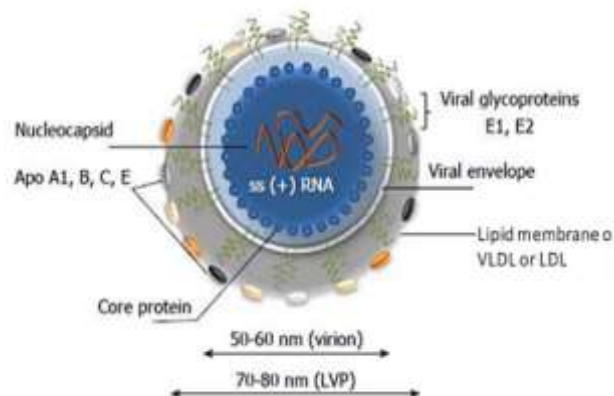


Figure 1-2: Hepatitis C virus (HCV) structure (Grassi et al., 2016)

Hepatitis viruses immune responses

1. Hepatitis B immune response

In general, the immune response is responsible for pathogens clearance and the appearance of signs of inflammation. Usually, hepatitis B virus can invade liver cells without appearance of any damage to the cells. Therefore, the immune response is important in controlling the spread of the virus and is also responsible for the inflammatory events that cause liver pathogenesis (Chisari et al., 2010).

The activation of the immune response is initiated through the cellular receptors represented by the TLRs, which leads to a series of events, including the production of antiviral cytokines such as interferon (IFN)- α , and the stimulation of innate immune cells such as natural killer (NK) cells and NKT cells which finally lead to the stimulation of adaptive immunity by activation and proliferation of T and B cells and generating the memory cells (Ma et al., 2018).

As for the hepatitis B virus, it attachment and enter through the hepatocyte receptors

represented by transporting polypeptide (NTCP) receptor, the viral nucleic acid is released and incorporated into the cell nucleic acid, this virus has developed distinct strategies to escape from the host's immunity (Herrscher et al., 2020), and employ hepatocytes for its reproduction and then will be recognized by Kupffer cells which lead to the production of much pro-inflammatory proteins (interleukin-1 beta (IL-1 β), interleukin-6 (IL6), tumor necrosis factor-alpha (TNF α), and interleukin-8 (IL-8)) (Hösel et al., 2009), then generated a strong response of CD4⁺ T cells and CD8⁺ T cells to control and eliminate HBV (Thomson and Knolle., 2010) , in addition to the role of B cells that are stimulated by T cells to produce anti-HBs, anti-HBe and anti-HBc. (Gerlich et al., 2013) Through the above mechanisms, it is possible to control the acute infection, but sometimes it is possible for the infection to develop and become chronic (Simmons et al., 2013) as a result of several factors, including those related to the virus (long incubation phase, an absence of danger signals, viral evasion etc.) and others related to the immune response of the host (genetic and environmental factors etc.) (Kgatle and Setshedi., 2016).

Chen and Tian 2019 explained that viral hepatitis B inflammation is within five stages, the first stage is immune tolerance, during which the virus has the ability to high replication and has weak inflammation, followed by the stage of active immunity with a cellular response, which includes CD8⁺ T cell and the production of antibodies, which are the result of infection and liver injury, the third stage represents by inactive immune response stage with a weak replication of the virus which leads to limited inflammation, the fourth stage, the immune response is reactivated, which coincides with the development of chronic hepatitis, the occurrence of fibrosis, cirrhosis and hepatocellular carcinoma. The final stage of the disease is the stage of immune exhaustion.

- **Immune escape mechanisms of the HBV include:**

It has been shown by many studies that HBV-specific CD8⁺ T cells in both the peripheral blood and liver microenvironment of patients with CHB always exhibit an exhausted phenotype (Bengsch et al., 2014).

Study by Fu and his colleagues 2020 has shown that patients with CHB possess suppressive mechanisms such as regulatory T cells and increased expression of co-inhibitory receptors such as cytotoxic T lymphocyte antigen 4 (CTLA-4), T-cell immunoglobulin and myosin domain 3 (Tim-3), inhibits the antiviral response, and it has been shown that hepatocytes of patients with CHB did not express certain co-stimulatory molecules (for example, CD80 and CD86), which means that affected hepatocytes may not be able to transduce the second signal required for CD8⁺ T cell activation (Fisicaro et al., 2020).

Tregs and macrophages may contribute to the suppression of immunity in patients with CHB through the production of immunomodulatory cytokines such as interleukin (IL-10) and tumor growth factor (TGF-1) (Wang et al ., 2022) .Furthermore, it has been shown through studies that myeloid-derived suppressor cells (MDSC) in the liver microenvironment have the ability to suppress T-cell signalling in part through hydrolyze arginine and significantly inhibit the T-cell effect or function (Dai et al., 2022).

One of the reasons for the progression of viral hepatitis to the stage of CHB is continuous treatment with IFN- α which is able to induce large numbers of CD24⁺ CD38^{hi} regulatory B cells (Bregs) and enhance the immunosuppressive response, resulting in the downregulation of CD8⁺ T cells and effector functions of killer cells (Fu et al ., 2020), also other types of cells in the liver play a role during chronic hepatitis B virus infection

such as the Kupffer cells that secrete immunomodulatory cytokines (Tacke ., 2017), for example, TGF- β 1, IL-10 (De Simone et al ., 2021), and strongly express PD-L1 or PD-L2 during CHB infection, thus suppressing antiviral immune responses and leading to immune tolerance (Tacke ., 2017).

2. Hepatitis C immune response

The hepatitis C virus is the main cause of chronic hepatitis and hepatocellular carcinomas, thus this virus will be exposed to innate and acquired immune mechanisms after entering it through its receptors. (Li et al., 2013)

Hepatitis C virus is an RNA virus that enters hepatocytes through its own receptors, such as heparan sulfate proteoglycan syndecan-1 or syndecan-4, CD81, scavenger receptor type B class 1 protein (SRB-1) and high-density lipoprotein (HDL) binding molecule very low- density lipoproteins (VLDL), and apolipoproteins (Apo) A1, B, C, and E (Vercauteren et al., 2014; Morozov and Lagaye 2018) and then uses the cellular machinery to rapidly proliferate, leaving infected cells to infect new cells, by this way HCV avoid an immune response (Dustin et al., 2016).

After the binding of the PAMPs of the virus to its receptors on the cells that include it and that will activate the innate response that recruits immune cells such as antigenic presenting cell and lymphocytes that will produce signaling molecules (cytokine such as interferon) that have an anti-viral role and inhibit its replication (Streicher and Jouvenet., 2019).

These interferon's have a role in activating innate and adaptive immune cells, where antibodies against this virus are formed within 7 to 8 weeks, and CD4⁺,CD8⁺T cells that can distinguish virus antigens presented through APCs with MCH II and MHC I molecules, respectively, are on their cell surface, and several studies found that there is a close correlation between an increase in the lymphocyte response (CD4⁺T and CD8⁺T) (Ashfaq et al., 2011; Abdelwahab., 2016) with the appearance of clinical symptoms and an increase in liver enzymes, over time (Shin et al., 2011), the infection turns into a chronic one as a result of the virus's continuous replication, by this way the virus will protect itself, as it cannot insert its genetic material within the cellular genetic material (Zeisel et al., 2013) , so as in the case of infection with HBV, over time, it leads to cirrhosis of the liver and the occurrence of hepatocellular carcinoma(HCC) (He et al., 2016).

The virus has developed strategies to evade these immune responses through, for example, its susceptibility to mutations as a result of the lack of RNA-dependent RNA polymerase to proofread, which helps it to escape immunity either through the formation of quasispecies due to change of the virus epitopes that deplete of the T cells. In addition, there is mutations may reduce the ability of the virus to multiply, which leads to a decrease in its presentation by antigenic presenting cells, which reduces the immune response of CD8⁺ T-cell spatially in chronic infection (Lucas et al., 2018)

The virus has the ability to encode several proteins that inhibit the immune response, such as Core, NS3/4A, NS4B, and NS5A(Ding et al., 2013), in addition to reduce the genes that stimulate interferon (ISGs) expression which these products have ability to degrading viral RNA or blocking translation of viral mRNA(Gokhale et al., 2014).

T cells are depleted and exhausted by continuous antigenic stimulation during infection (Kim et al., 2014) however, when it becomes chronic, T cells of both types,

CD4⁺ and CD8⁺, are depleted as a result of overexpression of inhibitory receptors (iRs) such as T-cell immunoglobulin, cytotoxic T- lymphocyte antigen 4 (CTLA-4), CD160, mucin domain-containing protein 3 (Tim-3), elevated anti-inflammatory interleukin 10 (IL-10) and programmed cell death-1 (PD-1) plasma levels. (Kared et al., 2013).

- Immune escape mechanisms of the HCV include:

One of the hallmarks of HCV infection is its ability to evade the host immune system through various mechanisms. The virus evades the immune response through the genetic features that characterize it, as it shows remarkable genetic diversity due to its high replication rate and error-prone RNA-dependent RNA polymerase. (Dustin et al., 2017)

This diversity allows the virus to generate viral variants, thereby evading host immune responses continuously.

Several HCV genotypes and subtypes have been identified, each with distinct immune evasion capabilities. (Martinez et al., 2020). This virus has multiple strategies to counteract the host's innate immune defenses. It inhibits the production of interferons (IFNs) and interferon-stimulated genes (ISGs), crucial for antiviral defense. HCV also targets pattern recognition receptors (PRRs) involved in viral detection, such as Toll-like receptors (TLRs) and retinoic acid-inducible gene I (RIG-I)-like receptors (RLRs), to dampen the host's immune response. (Iwasaki, 2012)

Additionally, HCV encodes several proteins, such as core, E1, E2, and NS3-NS5B, which play critical roles in evading host immune responses. These proteins interfere with various innate and adaptive immune system components, including interferon signaling pathways, antigen presentation, and T-cell responses (Chigbu et al., 2019) HCV alters the adaptive immune response by modulating antigen presentation and impairing T-cell function. It downregulates major histocompatibility complex (MHC) class I molecules, limiting the presentation of viral antigens to cytotoxic T lymphocytes (CTLs). HCV also induces the expansion of regulatory T cells (Tregs) and stimulates the production of immunoregulatory cytokines, suppressing effector T cell response (Larrubia et al., 2014).

Conclusion

In conclusion, it is important to note that the immune response to HBV and HCV is a complex and dynamic process that involves various factors beyond the structural differences described above. Host factors, viral load, viral persistence, and the interplay between innate and adaptive immune responses also significantly influence the outcome of the infection.

References

1. White, P.A., Netzer, N.E. and Hansman, G.S., 2016. Foodborne viral pathogens. CRC Press.
2. Hoofnagle, J.H., Nelson, K.E. and Purcell, R.H., 2012. Hepatitis e. New England Journal of Medicine, 367(13), pp.1237-1244.
3. Acorn, C. J., Millar, C. C., Hoffman, S. K., and Lyle, W. M. (1995). Viral hepatitis: a review. Optom. Vis. Sci. 72, 763–769. doi: 10.1097/00006324-199510000-00010
4. Rasche, A., Sander, A.-L., Corman, V. M., and Drexler, J. F. (2019). Evolutionary biology of human hepatitis viruses. J. Hepatol. 70, 501–520. doi: 10.1016/j.jhep.2018.11.010
5. Hughes, S. A., Wedemeyer, H., and Harrison, P. M. (2011). Hepatitis delta virus. Lancet 378, 73–85. doi: 10.1016/S0140-6736(10)61931-9
6. Tu, H., Lai, X., Li, J., Huang, L., Liu, Y. and Cao, J., 2019. Interleukin-26 is overexpressed in human sepsis and contributes to inflammation, organ injury, and mortality in murine sepsis.

- Critical Care, 23(1), pp.1-12.
7. Smith, D. B., and Simmonds, P. (2018). Classification and genomic diversity of enterically transmitted hepatitis viruses. *Cold Spring Harb. Perspect. Med.* 8:a031880. doi: 10.1101/cshperspect.a031880
 8. Lester, S. N., and Li, K. (2014). Toll-like receptors in antiviral innate immunity. *J. Mol. Biol.* 426, 1246–1264. doi: 10.1016/j.jmb.2013.11.024
 9. Streicher, F., and Jouvenet, N. (2019). Stimulation of innate immunity by host and viral RNAs. *Trends Immunol.* 40, 1134–1148. doi: 10.1016/j.it.2019.10.009
 10. Koyama, S., Ishii, K. J., Coban, C., and Akira, S. (2008). Innate immune response to viral infection. *Cytokine* 43, 336–341. doi: 10.1016/j.cyto.2008.07.009
 11. Wan, D., Jiang, W., and Hao, J. (2020). Research advances in how the cGAS-STING pathway controls the cellular inflammatory response. *Front. Immunol.* 11:615. doi: 10.3389/fimmu.2020.00615
 12. Carty M, Guy C, Bowie AG. ,2021. Detection of viral infections by innate immunity. *Biochemical pharmacology*,1(183):pp114316.
 13. Wang, Y., Cui, L., Yang, G., Zhan, J., Guo, L., Chen, Y., et al. (2019). Hepatitis B e antigen inhibits NF- κ B activity by interrupting K63-linked ubiquitination of NEMO. *J. Virol.* 93:e0667-18. doi: 10.1128/JVI.00667-18
 14. Gao, B. (2016). Basic liver immunology. *Cell Mol. Immunol.* 13, 265–266. doi: 10.1038/cmi.2016.09
 15. Ringelhan, M., Pfister, D., O'Connor, T., Pikarsky, E., and Heikenwalder, M. (2018). The immunology of hepatocellular carcinoma. *Nat. Immunol.* 19, 222–232. doi: 10.1038/s41590-018-0044-z
 16. Gao, B., Jeong, W.-I., and Tian, Z. (2008). Liver: an organ with predominant innate immunity. *Hepatology* 47, 729–736. doi: 10.1002/hep.22034
 17. Gao, B., Seki, E., Brenner, D. A., Friedman, S., Cohen, J. I., Nagy, L., et al. (2011). Innate immunity in alcoholic liver disease. *Am. J. Physiol. Gastrointest. Liver Physiol.* 300, G516–G525. doi: 10.1152/ajpgi.00537.2010
 18. Liu, Y., Li, J., Chen, J., Li, Y., Wang, W., Du, X., et al. (2015). Hepatitis B virus polymerase disrupts K63-linked ubiquitination of STING to block innate cytosolic DNA-sensing pathways. *J. Virol.* 89, 2287–2300. doi: 10.1128/JVI.02760-14
 19. Yi, Z., Chen, J., Kozlowski, M., and Yuan, Z. (2015). Innate detection of hepatitis B and C virus and viral inhibition of the response. *Cell Microbiol.* 17, 1295–1303. doi: 10.1111/cmi.12489
 20. Feng, Z., and Lemon, S. M. (2019). Innate immunity to enteric hepatitis viruses. *Cold Spring Harb. Perspect. Med.* 9:a033464. doi: 10.1101/cshperspect.a033464
 21. Hong, Y., Zhou, L., Xie, H., and Zheng, S. (2015). Innate immune evasion by hepatitis B virus-mediated downregulation of TRIF. *Biochem. Biophys. Res. Commun.* 463, 719–725. doi: 10.1016/j.bbrc.2015.05.130
 22. Jawetz, Melnick, & Adelberg.,2013. hepatitis viruses.Medical Microbiology. Brooks G F, Butel J S & Morse S A. (eds.) 26th (eds.). Appleton and Lange. Lurman. Eine Icterus epidemie. Berlin Klin Wschr, 1885, pp.20-22.
 23. Saha, S., Sikdar, P., Lavanya, D. and Babu, M.N., 2016. An Overview on Hepatitis and its Prevention. *International Journal of Pharma And Chemical Research*, 2(4), pp.210-219.
 24. Jefferies, M., Rauff, B., Rashid, H., Lam, T. and Rafiq, S., 2018. Update on global epidemiology of viral hepatitis and preventive strategies. *World journal of clinical cases*, 6(13), p.589.

25. Mohammed, M.O., Hasan, A.B. and Maarouf, F.B., 2019. Epidemiological Study of Hepatitis B, C and HIV Cases among the Foreigners Visiting Sulaimani City from 2013 Through 2016. *Kurdistan Journal of Applied Research*, pp.211-223.
26. Revill, P.A., Chisari, F.V., Block, J.M., Dandri, M., Gehring, A.J., Guo, H., Hu, J., Kramvis, A., Lampertico, P., Janssen, H.L. and Levrero, M., 2019. A global scientific strategy to cure hepatitis B. *The Lancet Gastroenterology & Hepatology*, 4(7), pp.545-558.
27. Robotis JF, Boleti H., 2007. Viral Hepatitis. *xPharm. The Comprehensive Pharmacology Reference*, 1–10. doi: 10.1016/B978-008055232-3.60928-6.
28. Yates, S., Penning, M., Goudsmit, J., Frantzen, I., van de Weijer, B., van Strijp, D. and van Gemen, B., 2001. Quantitative detection of hepatitis B virus DNA by real-time nucleic acid sequence-based amplification with molecular beacon detection. *Journal of clinical microbiology*, 39(10), pp.3656-3665.
29. Yost, S.A., Wang, Y. and Marcotrigiano, J., 2018. Hepatitis C virus envelope glycoproteins: A balancing act of order and disorder. *Frontiers in immunology*, 9, p.1917.
30. . Ranjbar, M.M., Ghorban, K., Alavian, S.M., Keyvani, H., Dadmanesh, M., Ardakany, A.R., et al., 2013. GB virus C/hepatitis G virus envelope glycoprotein E2: computational molecular features and immunoinformatics study. *Hepatitis monthly*, 13(12).
31. White, P.A., Netzer, N.E. and Hansman, G.S., 2016. *Foodborne viral pathogens*. CRC Press.
32. Murray, P.R., Rosenthal, K.S. and Pfaller, M.A., 2020. *Medical microbiology E-book*. Elsevier Health Sciences. p:809(550-555).
33. Netter, H.J., Barrios, M.H., Littlejohn, M. and Yuen, L.K., 2021. Hepatitis Delta Virus (HDV) and delta-like agents: Insights into their origin. *Frontiers in Microbiology*, 12, p.1448.
34. Hoofnagle, J.H., Nelson, K.E. and Purcell, R.H., 2012. Hepatitis e. *New England Journal of Medicine*, 367(13), pp.1237-1244.
35. Carroll, K.C., Butel, J. and Morse, S., 2015. *Jawetz melnick and adelbergs medical microbiology 27 E*. McGraw-Hill Education. p:502-508
36. Meo, S.A., Assad, A.A., Sanie, F.M., Baksh, N.D., Al-Qahtani, A. and Shaikh, Z.A., 2010. Transmission of hepatitis-B virus through salivary blood group antigens in saliva. *J Coll Physicians Surg Pak*, 20(7), pp.444-448.
37. Urban, S., Neumann-Haefelin, C. and Lampertico, P., 2021. Hepatitis D virus in 2021: virology, immunology and new treatment approaches for a difficult-to- treat disease. *Gut*, 70(9), pp.1782-1794. Rizzetto M, Alavian SM. 2013. Hepatitis delta: The rediscovery. *Clin Liver Dis*, 17: 475–487.
38. Rizzetto M, Alavian SM. 2013. Hepatitis delta: The rediscovery. *Clin Liver Dis* 17: 475–487
39. Moosavy, S.H., Davoodian, P., Nazarnezhad, M.A., Nejatizadeh, A., Eftekhari, E. and Mahboobi, H., 2017. Epidemiology, transmission, diagnosis, and outcome of Hepatitis C virus infection. *Electronic physician*, 9(10), p.5646
40. Cao, J., Zhang, J., Lu, Y., Luo, S., Zhang, J. and Zhu, P., 2019. Cryo-EM structure of native spherical subviral particles isolated from HBV carriers. *Virus research*, 259, pp.90-96.
41. Tan, W.S. and Ho, K.L., 2014. Phage display creates innovative applications to combat hepatitis B virus. *World journal of gastroenterology: WJG*, 20(33), p.11650.
42. Coppola, N., De Pascalis, S., Onorato, L., Calò, F., Sagnelli, C. and Sagnelli, E., 2016. Hepatitis B virus and hepatitis C virus infection in healthcare workers. *World journal of hepatology*, 8(5), p.273.
43. Ali, B.A., Salem, H.H., Wang, X.M., Huang, T.H., Xie, Q.D. and Zhang, X.Y., 2009. Detection of hepatitis B polymerase gene in early embryonic cells from golden hamster oocyte and human spermatozoa carrying HBV DNA. *Inter J Virol*, 5(4), pp.164-169.

44. Kosinska, A.D., Bauer, T. and Protzer, U., 2017. Therapeutic vaccination for chronic hepatitis B. *Current opinion in virology*, 23, pp.75-81.
45. Hu, J. and Liu, K., 2017. Complete and incomplete hepatitis B virus particles: formation, function, and application. *Viruses*, 9(3), p.56.
46. Gastaminza, P., Dryden, K.A., Boyd, B., Wood, M.R., Law, M., Yeager, et al., 2010. Ultrastructural and biophysical characterization of hepatitis C virus particles produced in cell culture. *Journal of virology*, 84(21), pp.10999-11009.
47. Vercauteren, K., Mesalam, A.A., Leroux-Roels, G. and Meuleman, P., 2014. Impact of lipids and lipoproteins on hepatitis C virus infection and virus neutralization. *World Journal of Gastroenterology: WJG*, 20(43), p.15975.
48. Morozov, V.A. and Lagaye, S., 2018. Hepatitis C virus: Morphogenesis, infection and therapy. *World journal of hepatology*, 10(2), p.186.
49. Blaising, J. and Pécheur, E.I., 2013. Lipids—A key for hepatitis C virus entry and a potential target for antiviral strategies. *Biochimie*, 95(1), pp.96-102.
50. Grassi, G., Di Caprio, G., Fimia, G.M., Ippolito, G., Tripodi, M. and Alonzi, T., 2016. Hepatitis C virus relies on lipoproteins for its life cycle. *World journal of gastroenterology*, 22(6), p.1953.
51. Chisari, F.V., Isogawa, M. and Wieland, S.F., 2010. Pathogenesis of hepatitis B virus infection. *Pathologie Biologie*, 58(4), pp.258-266.
52. Ma, Z., Cao, Q., Xiong, Y., Zhang, E. and Lu, M., 2018. Interaction between hepatitis B virus and toll-like receptors: current status and potential therapeutic use for chronic hepatitis B. *Vaccines*, 6(1), p.6.
53. Herrscher, C., Roingeard, P. and Blanchard, E., 2020. Hepatitis B virus entry into cells. *Cells*, 9(6), p.1486.
54. Hösel, M., Quasdorff, M., Wiegmann, K., Webb, D., Zedler, U., Broxtermann, M., et al., 2009. Not interferon, but interleukin-6 controls early gene expression in hepatitis B virus infection. *Hepatology*, 50(6), pp.1773-1782.
55. Thomson, A.W. and Knolle, P.A., 2010. Antigen-presenting cell function in the tolerogenic liver environment. *Nature Reviews Immunology*, 10(11), pp.753- 766.
56. Gerlich, W.H., 2013. Medical virology of hepatitis B: how it began and where we are now. *Virology journal*, 10(1), pp.1-25.
57. Simmons, R.A., Willberg, C.B. and Paul, K., 2013. Immune evasion by viruses. *eLS*.
58. Kgtale, M.M. and Setshedi, M., 2016. Immunopathogenesis of hepatitis B virus infection and related complications. *EMJ Hepatol*, 4, pp.84-92.
59. Bengsch, B., Martin, B. and Thimme, R., 2014. Restoration of HBV-specific CD8⁺ T cell function by PD-1 blockade in inactive carrier patients is linked to T cell differentiation. *Journal of hepatology*, 61(6), pp.1212-1219.
60. Fisicaro, P., Barili, V., Rossi, M., Montali, I., Vecchi, A., Acerbi, G., et al., 2020. Pathogenetic mechanisms of T cell dysfunction in chronic HBV infection and related therapeutic approaches. *Frontiers in Immunology*, 11, p.849.
61. Wang, D., Sun, Z., Zhu, X., Zheng, X., Zhou, Y., Lu, Y., Yan, P., Wang, H., Liu, H., Jin, J. and Zhu, H., 2022. GARP-mediated active TGF- β 1 induces bone marrow NK cell dysfunction in AML patients with early relapse post-allo- HSCT. *Blood, The Journal of the American Society of Hematology*, 140(26), pp.2788-2804.
62. Dai, X.K., Ding, Z.X., Tan, Y.Y., Bao, H.R., Wang, D.Y. and Zhang, H., 2022. Neutrophils inhibit CD8⁺ T cells immune response by arginase-1 signaling in patients with sepsis. *World Journal of Emergency Medicine*, p.0.

63. Tacke, F., 2017. Targeting hepatic macrophages to treat liver diseases. *Journal of hepatology*, 66(6), pp.1300-1312
64. De Simone, G., Andreatta, F., Bleriot, C., Fumagalli, V., Laura, C., Garcia- Manteiga, et al., 2021. Identification of a Kupffer cell subset capable of reverting the T cell dysfunction induced by hepatocellular priming. *Immunity*, 54(9), pp.2089-2100.
65. Li, C., Ji, H., Cai, Y., Ayana, D.A., Lv, P., Liu, M. and Jiang, Y., 2013. Serum interleukin-37 concentrations and HBeAg seroconversion in chronic HBV patients during telbivudine treatment. *Journal of Interferon & Cytokine Research*, 33(10), pp.612-618.
66. Vercauteren, K., Mesalam, A.A., Leroux-Roels, G. and Meuleman, P., 2014. Impact of lipids and lipoproteins on hepatitis C virus infection and virus neutralization. *World Journal of Gastroenterology: WJG*, 20(43), p.15975.
67. Morozov, V.A. and Lagaye, S., 2018. Hepatitis C virus: Morphogenesis, infection and therapy. *World journal of hepatology*, 10(2), p.186.
68. Dustin, L.B., Bartolini, B., Capobianchi, M.R. and Pistello, M., 2016. Hepatitis C virus: life cycle in cells, infection and host response, and analysis of molecular markers influencing the outcome of infection and response to therapy. *Clinical microbiology and infection*, 22(10), pp.826-832.
69. Streicher, F. and Jouvenet, N., 2019. Stimulation of innate immunity by host and viral RNAs. *Trends in Immunology*, 40(12), pp.1134-1148.
70. Ashfaq, U.A., Javed, T., Rehman, S., Nawaz, Z. and Riazuddin, S., 2011. An overview of HCV molecular biology, replication and immune responses. *Virology journal*, 8(1), pp.1-10.
71. Abdelwahab, S.F., 2016. Cellular immune response to hepatitis-C-virus in subjects without viremia or seroconversion: is it important?. *Infectious agents and cancer*, 11(1), pp.1-12.
72. Shin, E. C., Sung, P. S., & Park, S. H., 2016. Immune responses and immunopathology in acute and chronic viral hepatitis. *Nature Reviews Immunology*, 16(8), 509.
73. Zeisel, M.B., Lupberger, J., Fofana, I. and Baumert, T.F., 2013. Host- targeting agents for prevention and treatment of chronic hepatitis C—perspectives and challenges. *Journal of hepatology*, 58(2), pp.375-384.
74. He, Q., He, Q., Qin, X., Li, S., Li, T., Xie, L., et al., 2016. The relationship between inflammatory marker levels and hepatitis C virus severity. *Gastroenterology Research and Practice*, 2016.
75. Lucas, M., Deshpande, P., James, I., Rauch, A., Pfafferott, K., Gaylard, E., et al., 2018. Evidence of CD4⁺ T cell-mediated immune pressure on the Hepatitis C virus genome. *Scientific reports*, 8(1), pp.1-10.
76. Ding, Q., Cao, X., Lu, J., Huang, B., Liu, Y.J., Kato, N., et al., 2013. Hepatitis C virus NS4B blocks the interaction of STING and TBK1 to evade host innate immunity. *Journal of hepatology*, 59(1), pp.52-58.
77. Gokhale NS, Vazquez C, Horner SM., 2014. Hepatitis C Virus. Strategies to Evade Antiviral Responses. *Future Virol.*;9(12)
78. Kim, S.J., Syed, G.H., Khan, M., Chiu, W.W., Sohail, M.A., Gish, R.G., et al., 2014. Hepatitis C virus triggers mitochondrial fission and attenuates apoptosis to promote viral persistence. *Proceedings of the National Academy of Sciences*, 111(17), pp.6413-6418.
79. Kared, H., Fabre, T., Bedard, N., Bruneau, J. and Shoukry, N.H., 2013. Galectin-9 and IL-21 mediate cross-regulation between Th17 and Treg cells during acute hepatitis C. *PLoS pathogens*, 9(6), p.e1003422.
80. Dustin LB. Innate and Adaptive Immune Responses in Chronic HCV Infection. *Curr Drug Targets*. 2017;18(7):826-843. doi: 10.2174/1389450116666150825110532

81. Martinez MA, Franco S. Therapy Implications of Hepatitis C Virus Genetic Diversity. *Viruses*. 2020 Dec 29;13(1):41. doi: 10.3390/v13010041
82. Iwasaki A. A virological view of innate immune recognition. *Annu Rev Microbiol*. 2012;66:177–196
83. Chigbu DI, Loonawat R, Sehgal M, Patel D, Jain P. Hepatitis C Virus Infection: Host-Virus Interaction and Mechanisms of Viral Persistence. *Cells*. 2019 Apr 25;8(4):376. doi: 10.3390/cells8040376
84. Larrubia JR, Moreno-Cubero E, Lokhande MU, García-Garzón S, Lázaro A, Miquel J, Perna C, Sanz-de-Villalobos E. Adaptive immune response during hepatitis C virus infection. *World J Gastroenterol*. 2014 Apr 7;20(13):3418-30. doi: 10.3748/wjg.v20.i13.3418.