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Screening of Prevalence HBV and HCV from First Six Months of 2020 in Erbil Province Kurdistan Region /Iraq

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Abstract

Nowadays, infections of the HBV and HCV are one of the major global public health problem. The objective of this study was to determine the prevalence of HBV and HCV virus from the first six months of 2020 in the of Erbil province, Kurdistan region of Iraq. Also associations between the prevalence of HBV and HCV in different age groups, has been established.

The study was conducted on (101) individuals (63 males and 38 females), of different aged population groups, in Erbil city from beginning of January 2020 to the end of June 2020. To detect the positive results of both HBV and HCV (VL), all samples subjected to detect their viral load by GeneXpert (RT PCR or RTqPCR).

Out of (101) samples, 24(%23.76) of HBV and 7(%6.93) for HVC in both sex were detect positive respectively. Prevalence of both HBV and HCV was highest in males than females. Also the highest percentage of HBV found between (19-40) years, 12(%19.04) in males and 5(%13.15) in females. In addition highest number of HCV positive was found between (41-60) years, which 2(%3.17) and 2(%5.26) for males and females respectively, while opposite results were found from other aged groups

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Introduction

Viral hepatitis B and C viruses (HBV and HCV) are responsible for the majority of chronic liver disease worldwide (Villar *et al.,* 2015). An infected individual spreads the viruses in contaminated blood and body fluids to an uninfected individual (Pardee 2019). Worldwide, there are over 350 million people with chronic HBV (Hebo *et al.,* 2019). Of the approximately 2 billion exposed, more than 600 000 deaths annually; 170 million have chronic HCV, with nearly 500 000 deaths annually (Kateera *et al.,* 2015). HBV infection prevalence varies significantly by geographic area, ranging from 0.1% to 20% in different parts of the world and from 2% to 8% in countries with intermediate endemicity for hepatitis B infection (Tozun *et al.,* 2015).

Infections of chronic hepatitis B virus (HBV) and hepatitis C virus (HCV) are typically asymptomatic for decades, and proactive screening can avoid liver disease through prompt

diagnosis and treatment. More reliable estimates of the general population and risk groups for chronic HBV and HCV infections are required (Kanda et al. , 2019). The number of deaths caused by the majority of communicable diseases, hepatitis mortality rates worldwide, rose from 0.89 million in 1990 to 1.45 million in 2013 (Moradi *et al.,* 2019). Few reproducible mortality studies have been carried out in patients with autoimmune hepatitis (AIH) and its variants (van et al. ,2019).

Recently, an epidemiological study of HBV and HCV infection has been performed among individuals between the ages of 10 and 69 living in the five geographic regions of Brazil, with average HBsAg, anti-HBc and anti-HCV seroprevalence levels of 0.37%, 7.4% and 1.38% respectively (Villar *et al.,* 2014). The plan of the global health sector at the WHO to eradicate Viral Hepatitis as a major threat to public health by 2030 needs significant efforts from governments, health authorities and communities. To help control the number of people living with Hepatitis B Virus (HBV) (WHO 2016), service provision needs to be increased. Just 5 million of the estimated 71 million people diagnosed with HCV have currently been treated (Popping et al. ,2020). Several research showed that in the first years of injection drug use, the incidence of HCV was greatest, but little is known about health seeking actions (Enkelmann et al. ,2020).

Materials and Methods

Sample Collection

A total of 101 blood samples were collected from suspected patients, who visited the private clinical sectors, from 1 January to 30 June 2020. 10 mL of blood was drawn from each suspected patients. 5 mL of the blood samples collected inside the gel tube, after clotting, centrifuged them for 15 minutes at 5000 rpm. Other remained 5 mL of blood collect inside the (EDTA) tube, centrifuged as mentioned above. A questionnaire was also prepared and used to gather personal data (name (optional), age, and gender) for each case. The samples separated regarding to their gender to (63 males and remains were females), and their ages in to four specific age groups (6-18, 19 -40, 41 -60 and 61 -86).

HBV and HCV Viral Load Detection

Blood samples subjected for detection of HBV and HCV viral load by Real-Time qPCR (Real-Time Quantitative PCR) technology. Regarding to the manufacture instructions catalog number GXHBV-VL-CE-10 targets for HBV DNA, and catalog number GXHCV-VL-CE-10 targets for HCV RNA ,used for screening and diagnose the viral load of both virus in our samples. For detection of HBV (0.6-1mL plasma EDTA) for HCV (1mL of plasma of EDTA) added to their cartridges, by using 1mL micropipette with disposable pipette. Cartridges loaded inside the GeneXpert instrument, which it is full automate system and integrate specimen purification, nucleic acid amplification, and detection of the target sequence in simple or complex specimens using real-time reverse transcriptase PCR (RT-PCR) and (RT-qPCR) which uses fluorescence to detect the RNA and DNA of interest viruses. The systems require the use of single-use disposable GeneXpert cartridges that hold the RT-PCR reagents and host the RT-PCR processes.

Xpert HCV and HBV Viral Load cartridge includes reagents for the detection of HCV RNA and HBV DNA in specimens as well as two internal controls used for quantitation of HCV RNA and HBV DNA. The internal controls (High and Low) Internal Quantification Standards (IQS-H/IQS-L) monitor recovery and the presence of inhibitor(s) in the RT and PCR

reactions. To verifies that the correct volume of sample has been added in the sample chamber and ensure the sample was correctly added to the cartridge system has Sample Volume Adequacy (SVA) control. Also the Probe Check Control (PCC) verifies reagent rehydration, PCR tube filling in the cartridge, probe integrity, and dye stability. Because the cartridges are self-contained, cross contamination between samples is minimized. The results are interpreted automatically by the GeneXpert Instrument System from measured fluorescent signals. Regarding to WHO reference material for plasma and serum samples limitation by geneXpert system reported as 3.20 IU/mL, 5.99 IU/mL respectively for linear range quantification 10 to 109 IU/mL for HBV and 10 to 108 IU/mL for HCV (WHO 2016).

Statistical Analysis

The chi-square was applied to examine the relationship between the prevalence of HBV and HCV from different sexes and to determine the association the prevalence of HBV and HCV among different aged gropes. P-values < 0.05 were considered to be statistically significant.

Results

Table 1. prevalence of HBV and HCV according to gend

Viruses	Number of screened			% positive			P value
	Males	Females	Total	Males	Females	Total	
HBV	63(%62.37)	38(%37.62)	101(%100)	19(%30.1)	5(%13.15)	24(%23.76)	0.004
HCV	63(%62.37)	38(%37.62)	101(%100)	3(%4.76)	2(%5.12)	7(%6.93)	0.654

Table 2. prevalence of HBV according to four specific age groups

Gender	Different age groups %					HBV positive %					P value
	6-18	19-40	41-60	61-86	Total	6-18	19-40	41-60	61-86	Total	
Male	5 (%7.93)	27 (%42.85)	15 (%28.8)	16 (%25.39)	63 (%62.37)	1 (%1.58)	12 (%19.04)	5 (%7.93)	1 (%1.58)	19 (%30.1)	**0.0007**
Female	3 (%7.89)	18 (%47.36)	15 (%39.47)	2 (%5.26)	38 (%37.62)	0 (%0)	5 (%13.15)	0 (%0)	0 (%)	5 (%13.15)	**0.001**

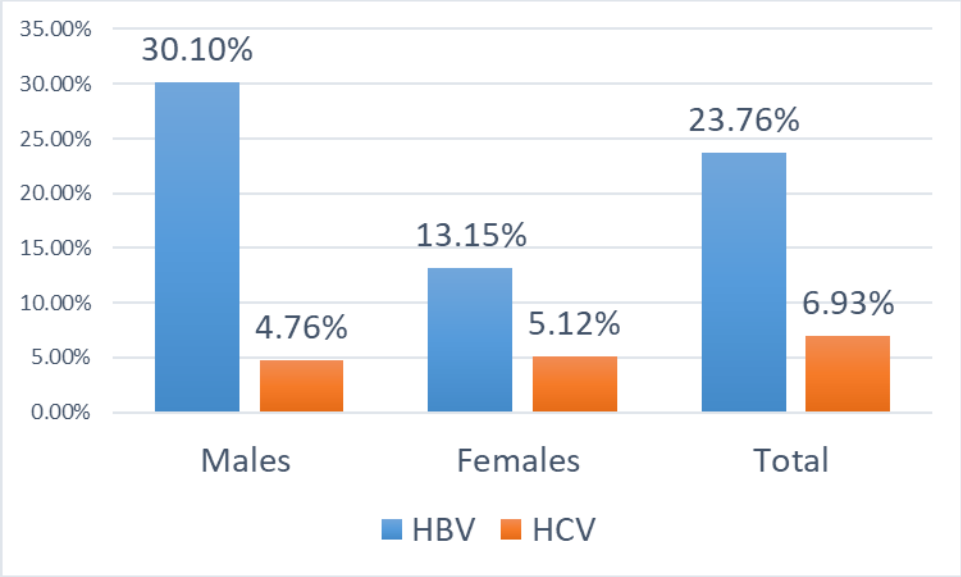

Figure 1. Prevalence of HBV and HCV according to genders

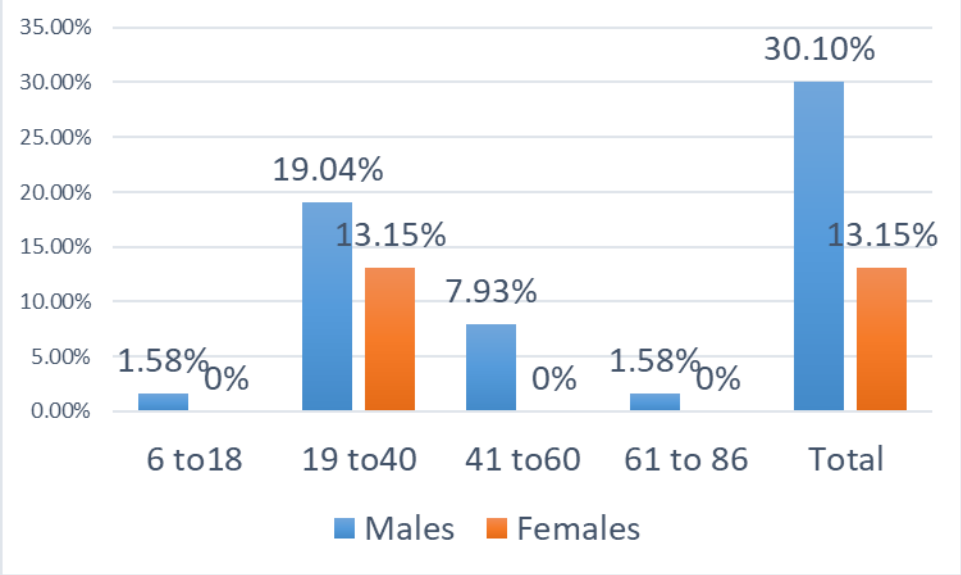

Figure 2. Prevalence of HBV according to four specific groups

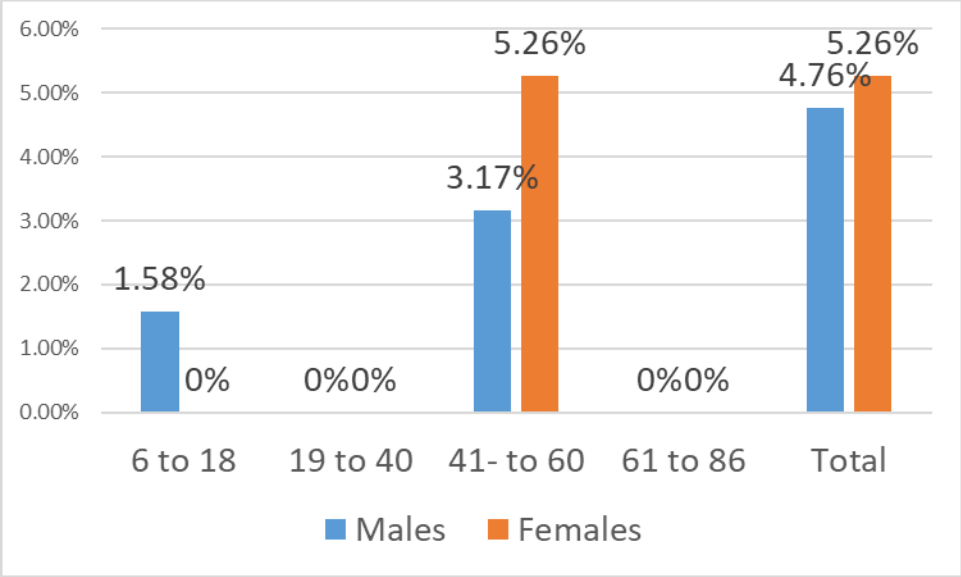

Figure 3. Prevalence of HCV according to four specific groups

Discussion

Infection with the hepatitis B virus (HBV) and hepatitis C virus (HCV) is a public health problem (Police et al. ,2020). Many men, 40-80% live with chronic hepatitis B or C, are unaware of their serostatus, and remain infectious to others (Ayele et al. ,2020). Recent data have shown that about 350 million people are chronically infected with HBV and around 200 million are infected with HCV (Hussein et al . ,2017).

In recent years, more and more data have been developed in developing countries and high/intermediate endemic areas where the most common route of infection is still vertical transmission from mother to child, and horizontal transmission between children , particularly siblings (Franco *et al.,* 2012). Previous data from 2006 to 2009 suggest that a low prevalence of hepatitis B and hepatitis C infection exists in Iraq's capital city Baghdad (Ataallah *et al.,* 2011). It also reported similar results in Basra (Al-Rubaye et al . , 2016). HBV and HVC prevalence are investigated in Erbil (Taher et al. ,2020).

The prevalence of hepatitis B surface antigen (HBsAg) and HCV antibody (anti-HCV) was identified in the Turkish population as 2.3% and 0.37% respectively (Yakaryilmaz *et al.,* 2006). At the national level, the age-standardized prevalence of hepatitis B in Iran fell from 3.02 % (95 % uncertainty interval; 2.26 to 3.96) in 2000 to 1.09 % (95 %uncertainty intervals; 0.85 to 1.37) in 2016, with a cumulative improvement of -64.84 %. In 2016 the prevalence of hepatitis B in males was more than 1.3 times greater than in females (Rezaei et al. ,2020).

The total number of screened for HBV and HBC were 101 blood samples, 63(%62.37) of them were males and 38(%37.62) were females. The present study indicated that %23.76 in males and %13.15 in females of the samples were positive for HBV, while the number of HCV positive dramatically decreased to %4.76 in males and %5.12 in females. For both viruses statistically showed significant for HBV the p-value was 0.004, whereas for HCV was non-significant and the p-value 0.654. (table 3.1 and figure 3.1). Although the prevalence of HBV and HCV among diabetic patients is higher, it has a low prevalence compared with other patients studied (Merza et al. ,2020). At 1992 epidemiology estimated that the prevalence of HBsAg in the general Chinese population was 75 % (Said et al . , 2019). Chronic hepatitis B patients reported slightly lower health-related life scores in various areas relative to stable control subjects in the province of AL-Najaf / Iraq (Kadhim et

al. ,2020).

There are 3 prequalified rapid diagnostic assays for hepatitis B surface antigen (HBsAg) by WHO that is responsive, accurate, and effective in HBV screening: SD Bioline, Determines 2, and VIKIA. POCTs are less accurate and sensitive to other biomarkers but have some applicability. The GeneXpert HBV viral load assay is sensitive and specific with a wide range of dynamics (Jackson et al. ,2020). The HBV DNA quantification is used to initiate and monitor antiviral treatment (Abravanel et al. ,2020). The GeneXpert HBV-VL test, which has the potential for near-point molecular treatment testing, showed excellent performance and proved to be a reliable tool for quantification of HBV DNA (Khodare et al. ,2020). The research found a seroprevalence rate of 13.3 % of HBeAg among prospective asymptomatic blood donors (Rufai *et al.,* 2014).

In our results showed that there are association among the different aged group that infected by HBV in male, the statistical of p-value was 0.0007, the highest number was 19-40 aged groups 12(%19.04) while in 41-60 ages were 5(%7.93) .On the other hands for two other remains groups have the lowest number of positive HBV 1(%1.58) respectively .In female the prevalence of HBV positive has the largest number was 5(%13.15) in aged groups 19-40 years and three other groups there were no any infected HBV positive and statistically significant (table 3.2 and figure 3.2). HBV prevalence at 2.2% and carrier rate at 0.3% in men and 0.1% in women (Goudeau *et al.,* 1995). At first sex, the number checked and those positive in the age group is as follows: < 10 years (7/25), 11-14 years (24/143), 15-18 years (76/433), 19-22 years (14/103), > 23 years (2/16)(Forbi *et al.,* 2008). The prevalence of hepatitis B surface antigen (HBsAg) and total anti-HBc (core) was 6.8% and 35.6% in Turkish ,Kurds, and 2.2% and 12.7% in Iraqis respectively (Chironna *et al.,* 2003). Based on the prevalence in the general population of carriers of hepatitis B surface antigen, countries are classified as high (> 8%), intermediate (2-7%) or low (< 2%) endemic. Iraq is among the middle prevalence countries (Hussein *et al.,* 2015). Also, approximately 5% of the general population in western Iran has previous HBV exposure and less than 1% is HBsAg carrier (Alavian *et al.,* 2012). The prevalence of hepatitis B surface antigen (HBsAg) varies according to different reports, but in Iranian provinces, a prevalence of 1.2%-5% was registered (Ziaee et al . , 2016).

At the point of care, molecular testing can turn out to be a game-changer for HCV diagnosis and treatment monitoring through increased sensitivity, reduced turnaround time, and performance ease. One such assay has recently been released on GeneXpert (Gupta *et al.,* 2017). Although the RNA point-of-care technology for novel hepatitis C virus (HCV) has the potential to improve diagnosis in resource-limited settings (Mohamed et al. ,2020). Viral load GeneXpert HCV (within 105 minutes). (Latham *et al.,* 1998). For GeneXpert HCV Viral Load and venipuncture blood samples, finger-stick whole-blood samples were used (Bajis et al . , 2019; Calvaruso *et al.,* 2019; Saludes *et al.,* 2020). Regarding HCV infection 3(%4.76) for different aged groups mostly between 41-60 years, their prevalence was 2(%3.17) and it is higher than from other groups and statistically significant in males the p-value was 0.0008. However for female groups all positive HCV in 41-60 aged groups (2(%5.26) and cannot find any positive HCV from reaming groups, therefor statistically no significant and its p-value was 0.11 (table 3.3 and figure 3.3). In Iraq HCV prevalence rises dramatically with maternal age, women around 30 years of age are considered to be a risk group for HCV infection (Al-Kubaisy 2012).evelopment are people who tend to be lazy and do not care about their environment.

Conclusions

We have shown that nearly a quarter of the blood sample positive for HBV is also positive for HBV, a marker of active viral replication and transmission, which can be taken as an indicator of active HBV in Erbil individuals. In addition, the prevalence of HCV in these samples of Erbil was considered low, which could lead to the prevention and health care of these populations.

References

Abravanel, F., Lhomme, S., Trémeaux, P., Migueres, M., Harter, A., Haslé, C., Bruel, P., Alric, L., Métivier, S., Raymond, S. and Izopet, J., 2020. Performance of the Xpert HBV Viral Load assay versus the Aptima Quant assay for quantifying hepatitis B virus DNA. *Diagnostic Microbiology and Infectious Disease*, *96*(2), p.114946.

Alavian, S.M., Tabatabaei, S.V., Ghadimi, T., Beedrapour, F., Kafi-abad, S.A., Gharehbaghian, A. and Abolghasemi, H., 2012. Seroprevalence of hepatitis B virus infection and its risk factors in the west of Iran: a population-based study. *International journal of preventive medicine*, *3*(11), p.770.

Al-Kubaisy, W., 2012. Specific HCV antibodies, RNA, and genotypes detection correlated to the age of pregnant women in Iraq. *International Journal of Infectious Diseases*, *16*, p.e67.

Al-Rubaye, A., Tariq, Z. and Alrubaiy, L., 2016. Prevalence of hepatitis B seromarkers and hepatitis C antibodies in blood donors in Basra, Iraq. *BMJ open gastroenterology*, *3*(1).

Ataallah, T.M., Hanan, K.A., Maysoun, K.S. and Sadoon, A.A., 2011. Prevalence of hepatitis B and C among blood donors attending the National Blood Transfusion Center in Baghdad, Iraq from 2006-2009. *Saudi Med J*, *32*(10), pp.1046-50.

Ayele, A., Abera, D., Hailu, M., Birhanu, M. and Desta, K., 2020. Prevalence and associated risk factors for Hepatitis B and C viruses among refugees in Gambella, Ethiopia. *BMC Public Health*, *20*, pp.1-10.

Bajis, S., Grebely, J., Cooper, L., Smith, J., Owen, G., Chudleigh, A., Hajarizadeh, B., Martinello, M., Adey, S., Read, P. and Gilliver, R., 2019. Hepatitis C virus testing, liver disease assessment and direct-acting antiviral treatment uptake and outcomes in a service for people who are homeless in Sydney, Australia: The LiveRLife homelessness study. *Journal of viral hepatitis*, *26*(8), pp.969-979.

Calvaruso, V., Bronte, F., Ferraro, D., Reina, G., Conte, E., Rini, F., Magro, B., Petta, S., Di Marco, V. and Craxì, A., 2019. Point-of-care HCV RNA testing in the setting of DAA therapy: HCV-FiS (HEpatitis C Virus Fingerstick Study). *Liver International*, *39*(12), pp.2240-2243.

Chironna, M., Germinario, C., Lopalco, P.L., Carrozzini, F., Barbuti, S. and Quarto, M., 2003. Prevalence rates of viral hepatitis infections in refugee Kurds from Iraq and Turkey. *Infection*, *31*(2), pp.70-74.

Enkelmann, J., Gassowski, M., Nielsen, S., Wenz, B., Roß, S., Marcus, U., Bremer, V. and Zimmermann, R., 2020. High prevalence of hepatitis C virus infection and low level of awareness among people who recently started injecting drugs in a cross-sectional study in Germany, 2011–2014: missed opportunities for hepatitis C testing. *Harm Reduction Journal*, *17*(1), pp.1-10.

Forbi, J.C., Onyemauwa, N., Gyar, S.D., Oyeleye, A.O., Entonu, P. and Agwale, S.M., 2008. High prevalence of hepatitis B virus among female sex workers in Nigeria. *Revista do Instituto de Medicina Tropical de São Paulo*, *50*(4), pp.219-221.

Franco, E., Bagnato, B., Marino, M.G., Meleleo, C., Serino, L. and Zaratti, L., 2012. Hepatitis B: Epidemiology and prevention in developing countries. *World journal of hepatology*, *4*(3), p.74.

Goudeau, A. and Dubois, F., 1995. Incidence and prevalence of hepatitis B in France. *Vaccine*, *13*, pp.S22-S25.

Gupta, E., Agarwala, P., Kumar, G., Maiwall, R. and Sarin, S.K., 2017. Point-of-care testing (POCT) in molecular diagnostics: Performance evaluation of GeneXpert HCV RNA test in diagnosing and monitoring of HCV infection. *Journal of Clinical Virology*, *88*, pp.46-51.

Hebo, H.J., Gemeda, D.H. and Abdusemed, K.A., 2019. Hepatitis B and C viral infection: prevalence, knowledge, attitude, practice, and occupational exposure among healthcare workers of Jimma University Medical Center, Southwest Ethiopia. *The Scientific World Journal*, *2019*.

Hussein, N.R. and Taher, A.M., 2015. The seroprevalence of HBV and HCV Infection in newly recruited police in Duhok city, Kurdistan Region, Iraq. *Int J Sci Basic Appl Res*, *24*(1), pp.112-6.

Hussein, N.R., Haj, S.M., Almizori, L.A. and Taha, A.A., 2017. The prevalence of hepatitis B and C viruses among blood donors attending blood bank in Duhok, Kurdistan region, Iraq. *Int J Infect*, *4*(1), p.e39008.

Jackson, K. and Gish, R.G., 2020. Point of Care Diagnostic Testing for Hepatitis B Virus. *Current Hepatology Reports*, pp.1-9.

Kadhim, D.J., 2020. Health-Related Quality of Life among a Sample of Chronic Hepatitis B Patients in AL-Najaf Province/Iraq. *Iraqi Journal of Pharmaceutical Sciences (P-ISSN: 1683-3597, E-ISSN: 2521-3512)*, *29*(1), pp.33-40.

Kanda, T., Goto, T., Hirotsu, Y., Moriyama, M. and Omata, M., 2019. Molecular mechanisms driving progression of liver cirrhosis towards hepatocellular carcinoma in chronic hepatitis B and C infections: a review. *International journal of molecular sciences*, *20*(6), p.1358.

Kateera, F., Walker, T.D., Mutesa, L., Mutabazi, V., Musabeyesu, E., Mukabatsinda, C., Bihizimana, P., Kyamanywa, P., Karenzi, B. and Orikiiriza, J.T., 2015. Hepatitis B

and C seroprevalence among health care workers in a tertiary hospital in Rwanda. *Transactions of the Royal Society of Tropical Medicine and Hygiene*, *109*(3), pp.203-208.

Khodare, A., Gupta, E., Rani, N., Singh, G., Aggarwal, K., Sharma, M. and Sarin, S.K., 2020. Performance evaluation of Xpert HBV viral load (VL) assay: Point-of-care molecular test to strengthen and decentralize management of chronic hepatitis B (CHB) infection. *medRxiv*.

Latham, N.H., Pedrana, A., Doyle, J.S., Howell, J., Williams, B., Higgs, P., Thompson, A.J. and Hellard, M.E., 2019. Community-based, point-of-care hepatitis C testing: perspectives and preferences of people who inject drugs. *Journal of viral hepatitis*, *26*(7), pp.919-922.

Merza, M.A., 2020. Seroprevalence and risk factors of hepatitis B and C viruses among diabetes mellitus patients in Duhok province, Iraqi Kurdistan. *Journal of Family Medicine and Primary Care*, *9*(2), p.642.

Mohamed, Z., Mbwambo, J., Rwegasha, J., Mgina, N., Doulla, B., Mwakale, P., Tuaillon, E., Chevaliez, S., Shimakawa, Y., Taylor-Robinson, S.D. and Thursz, M.R., 2020. In-field evaluation of Xpert® HCV viral load Fingerstick assay in people who inject drugs in Tanzania. *Liver International*, *40*(3), pp.514-521.

Moradi, G., Piroozi, B., Alinia, C., Safari, H., Mohamadi, P., ZAVAREH, F.A., Ramezani, R., Gouya, M.M. and Nabavi, M., 2019. Incidence, mortality, and burden of hepatitis B and C and geographical distribution in Iran during 2008-2015. *Iranian Journal of Public Health*, *48*(Supple 1), pp.10-19.

Pardee, M., 2019. Diagnosis and Management of Hepatitis B and C. *Nursing Clinics*, *54*(2), pp.277-284.

Police, S.M.C., Boua-Akélélo, N.P., Mofini, E., Elowa, B., Kalebanga, A.T.Y., Bessanguem, B., Simaléko, M.M., Guérendo, P., Diemer, H. and Kobendo, J.R.M., 2020. Prevalence of HBsAg and Antibodies to Hepatitis C Virus among Female Sex Workers in Bangui. *Open Journal of Gastroenterology*, *10*(06), p.144.

Popping, S., Verwijs, R., Cuypers, L., Claassen, M.A., van den Berk, G.E., De Weggheleire, A., Arends, J.E., Boerekamps, A., Molenkamp, R., Koopmans, M.P. and Verbon, A., 2020. Transmission of NS5A-Inhibitor Resistance-Associated Substitutions Among Men Who Have Sex With Men Recently Infected with Hepatitis C Virus Genotype 1a. *Clinical Infectious Diseases*.

Rezaei, N., Asadi-Lari, M., Sheidaei, A., Gohari, K., Parsaeian, M., Khademioureh, S., Maghsoudlu, M., Kafiabad, S.A., Zadsar, M., Motevalian, S.A. and Delavari, F., 2020. Epidemiology of Hepatitis B in Iran from 2000 to 2016: A Systematic Review and Meta-Regression Analysis. *Archives of Iranian Medicine*, *23*(3), pp.189-196.

Rufai, T., Mutocheluh, M., Kwarteng, K. and Dogbe, E., 2014. The prevalence of hepatitis B virus E antigen among Ghanaian blood donors. *Pan African Medical Journal*, *17*(1).

Said, B.A. and Hassan, R.A.R., 2019.Risk factors for hepatitis b virus among blood donors in Baghdad, Iraq. International Journal of Advanced Research in Medicine,1(2), pp. 62-67

Saludes, V., Antuori, A., Lazarus, J.V., Folch, C., González-Gómez, S., González, N., Ibáñez, N., Colom, J., Matas, L., Casabona, J. and Martró, E., 2020. Evaluation of the Xpert HCV VL Fingerstick point-of-care assay and dried blood spot HCV-RNA testing as simplified diagnostic strategies among people who inject drugs in Catalonia, Spain. *International Journal of Drug Policy*, *80*, p.102734.

Taher, C.A. and Saleh, G.N., 2020. Prevalence and Characterization of Hepatitis B and Hepatitis C Infection among Blood Donors in Erbil. *Cihan University-Erbil Scientific Journal*, *4*(1), pp.45-51.

Tozun, N., Ozdogan, O., Cakaloglu, Y., Idilman, R., Karasu, Z., Akarca, U.L.U.S., Kaymakoglu, S. and Ergonul, O., 2015. Seroprevalence of hepatitis B and C virus infections and risk factors in Turkey: a fieldwork TURHEP study. *Clinical Microbiology and Infection*, *21*(11), pp.1020-1026.

van den Brand, F.F., van der Veen, K.S., de Boer, Y.S., van Gerven, N.M., Lissenberg-Witte, B.I., Beuers, U., van Erpecum, K.J., van Buuren, H.R., den Ouden, J.W., Brouwer, J.T. and Vrolijk, J.M., 2019. Increased mortality among patients with vs without cirrhosis and autoimmune hepatitis. *Clinical Gastroenterology and Hepatology*, *17*(5), pp.940-947.

Villar, L.M., Amado, L.A., De Almeida, A.J., De Paula, V.S., Lewis-Ximenez, L.L. and Lampe, E., 2014. Low prevalence of hepatitis B and C virus markers among children and adolescents. *BioMed Research International*, *2014*.

Villar, L.M., Cruz, H.M., Barbosa, J.R., Bezerra, C.S., Portilho, M.M. and de Paula Scalioni, L., 2015. Update on hepatitis B and C virus diagnosis. *World journal of virology*, *4*(4), p.323.

World Health Organization, 2016. *Global health sector strategy on viral hepatitis 2016-2021. Towards ending viral hepatitis* (No. WHO/HIV/2016.06). World Health Organization.

Yakaryilmaz, F., Alp Gurbuz, O., Guliter, S., Mert, A., Songur, Y., Karakan, T. and Keles, H., 2006. Prevalence of occult hepatitis B and hepatitis C virus infections in Turkish hemodialysis patients. *Renal failure*, *28*(8), pp.729-735.

Ziaee, M., Ebrahimzadeh, A., Azarkar, Z., Namaei, M.H., Saburi, A., Fereidouni, M., Bijari, B., Karamian, M. and Sharifzadeh, G., 2016. Seroprevalence and risk factors for hepatitis B in an adult population: the first report from Birjand, South Khorasan, Iran. *Hepatitis monthly*, *16*(9).