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Differences in serum Lipopolysaccharide level among Hepatitis B virus, Hepatitis C virus (and liver cirrhotic patients

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Abstract

Bacterial translocation (BT) into the systemic circulation has been documented to be associated with liver dis-ease progression. However, BT has not completely defined in patients with Hepatitis B virus (HBV), Hepatitis C virus (HCV), and Liver cirrhosis. This study aims to study the differences in the lipopolysaccharide (LPS) level as a marker for BT among patients with HBV, HCV, and liver cirrhosis and their effect on disease severity. Across-sectional study was conducted. A total of 89 blood samples was collected from patients with HBV, HCV, and liver cirrhosis. Serum samples was used to measure the level of LPS automatically by ELISA Techniques. SPSS, version 22 software (IBM Corp., NY, and USA), was used to analyze data . Out of all samples analysed pa-tients, 45 (50.56%) patients were infected with HBV, 21 (23.59%) infected with HCV and 23 (25.84%) with liver cirrhosis., The mean age of the patients with HBV, HCV, and Liver cirrhosis were 39.84 ±16.823 years, 42.76 ±15.59 years, and 49.87 ±15.9 years, respectively, Additionally, there were statistically signif-icant difference in the mean age of patients with HBV infection and liver cirrhosis p-value less than 0.05. The mean LPS level was significantly lower in patients with HBV compared to those with HCV and liver cirrhosis. conclusion: The signif-icant positive correlation of LPS with ALP (ALP results was collected from reports of patients) may support the role of this parameter in disease pathogenesis. The highest mean LPS level found in liver cirrhotic patients may reflect the impact of LPS in pathogenesis.

Keywords: Viral hepatitis; HBV; HC; liver cirrhosis; Bacterial translocation; Lipopolysaccharide (LPS).

1.Introduction

Viral hepatitis is a important public health issue [1]. The WHO predicts that in 2019, 296 million individuals had chronic hepatitis B virus (HBV) infections, with 1.5 million new cases occurring annually[2] Hepatitis C virus (HCV) infects 1.5 million individuals worldwide each year, and 58 million people have chronic HCV infection, with a prevalence of 0.8 (0.6-1.0) percent in the general population worldwide [3].Chronic inflammation of liver caused by HBV and HCV can lead to scarring of the tissues (fibrosis), irreversible scarring (cirrhosis), and hepatocellular carcinoma (HCC) [4] Globally, among individuals with cirrhosis 42% had HBV infection and 21% had HCV infection [5].

The liver plays a critical role as an immunological barrier against pathogens in the circulation [6]. When liver is inflamed or damage due to viral infection, these immune activities of liver become less effective, allowing more gut-derived germs to enter the bloodstream and increasing the risk of developing systemic bacterial infection [7].

An essential factor in the emergence of infection in liver cirrhosis is bacterial translocation (BT). In healthy individuals, the migration and colonization of bacteria and/or bacterial products from the gut to mesenteric lymph nodes is a regulated process.BT occurs through three mechanisms, impaired host defense, bacterial overgrowth, and disruption of the mucosal barrier. Impairment of BT in cirrhosis is promoted by increased intestinal permeability, bacterial overgrowth, and defects in the gut-associated lymphatic tissue [8][9][0]. Microbial components like lipopolysaccharide and peptidoglycan are hypothesized to both contribute to and accelerate the progression of liver disease [7]

Increased microbial translocation has been found with HBV and HCV [11] [12]. Understanding the relationship between bacterial translocation and the severity of hepatitis infection may help to understand how translocation affects chronic liver illnesses. This might result in new treatment targets for preventing infections and other cirrhosis consequences[13]

The aims of this current study is to investigate the differences in serum level of LPS among patients with HBV and HCV infections, Study the association of the of LPS level with the disease severity.

2.Methods

Across-sectional study was carried out in College of Applied Medical Sciences, University of Kerbala, from the period of October 2022 to february2023. Out of 89 blood samples was collected from

patients with HBV, HCV, liver cirrhosis whom they referred to Karbala health departmental/ AL-Hussain Medical city, Women Obstetrics and Gynecology Hospital, and Karbala Center for Diseases and surgery of Digestive system and liver. Serum samples was used to measure the level of LPS, automatically by ELISA Techniques.

2 .1 Statistical Analysis

Data was analyzed using SPSS, version 22 software (IBM Corp., NY, USA).Mean, Standard Deviation, and cross-tabulation, Bivariate correlations, the Least Significant Difference (LSD) test and the Analysis of Variance (ANOVA) test was calculated. To evaluate the categorical variables, the chi-square test was applied. P value <0.05 was used to determine the statistical significance.

3.Results

Eighty-nine patients were enrolled in this study. Out of 45 (50.56%) of patients were infected with HBV, 21 (23.59%) infected with HCV and 23 (25.84%) with liver cirrhosis, The mean age of the patients with HBV, HCV, and Liver cirrhosis were 39.84 ±16.823, 42.76 ±15.59, and 49.87 ±15.9, respectively, Additionally, in the mean ages of patients was differ significantly between HBV infection and liver cirrhosis. A higher mean of age was found in cirrhotic liver patients, as shown in Table (1), The distribution of patients according to age was shown in Table (2).

Virus types	Mean age	N (%)	SD	Multiple Comparisons LSD			
				(I) virus types	(J) virus types	Mean Difference (I-J)	Sig.
HBV	*39.84*	*45 (50.56)*	*16.823*	*HBV*	*HCV*	*-2.917*	*.500*
					Liver cirrhosis	*-10.025**	*.019*
HCV	*42.76*		*15.595*	*HCV*	*HBV*	*2.917*	*.500*

		21 (23.5)			*Liver cirrhosis*	*-7.108*	*.152*
Liver cirrhosis	*49.87*	*23 (25.8)*	*15.907*	*Liver cirrhosis*	*HBV*	*10.025**	*.019*
					HCV	*7.108*	*.152*
ANOVA test P-value	*0.061*						

*. The mean difference is significant at the 0.05 level;SD: Standard Deviation; LSD: Least Significant Difference

Table 1. Mean age of patients

Table 2. Distribution of Patients According to Age Categories

		Type of Disease N (%)			Total
		HBV	HCV	Liver cirrhosis	
Age Categories	≤40 years	*27(60)*	*11(52.3)*	*6(26.08)*	*44(49.43)*
	>40 years	*18(40)*	*10(47.61)*	*17(73.91)*	*45(50.56)*
Total		*45(100)*	*21(100)*	*23(100)*	*89(100)*
P-value		*0.029*			

The Sex distribution of patients were shown in Table (3) with Male/ Female ratio (16/29, 0.5:1), (9/12,0.75:1), (13/10,1.3:1), For HBV, HCV, and liver cirrhosis, respectively.

Table 3. Distribution of patients with HBV, HCV, and liver cirrhosis according to Sex

Type of Disease N (%)

Sex	HBV	HCV	Liver cirrhosis	Total
Male	*16(35.55)*	*9(42.8)*	*13(56.52)*	*38(42.69)*
Female	*29(64.44)*	*12(57.1)*	*10(43.47)*	*51(57.30)*
Total	*45(100)*	*21(100)*	*23(100)*	*89(100)*
P- Value	*0.255*			

The current study revealed the presence of significantly lower mean of LPS in patients with HBV in comparison to HCV and liver cirrhotic patients, as show in table (4).

Table 4. Differences in LPS level among HBV, HCV and Liver cirrhotic patients

	Type of disease	Mean	SD	Min	Max	Post Hoc	Kruskal-Wallis test *P-value*
LPS	HBV	105.89	110.02	2	476	b	.001*
	HCV	232.29	131.03	18	514	a	
	Liver cirrhosis	299.64	133.49	9	619	a	

* Kruskal-Wallis test

*Significant at 0.05 level, Similar letter (a) means non-significant differences

This study showed that there was significant positive correlation of LPS with Alkaline phosphatase. No significant correlation was found with ALT, AST, TSB, PT, and Albumin, as shown in Table (5)

Table 5. Correlation between LPS Markers of liver damage

	ALT	AST	ALP	TSB	PT	Alb

LPS	Pearson Correlation	.007	.084	.395**	.173	.125	.028
	Sig. (2-tailed)	.962	.550	.006	.244	.579	.953
	N	55	53	47	47	22	7

AIT (alanine aminotransferase); AST (aspartate aminotransferase); ALP (Alkaline Phosphatase); TSB (Total serum bilirubin); PT (Prothrombin Time); Alb (albumin)

4. Discussion

This study aimed to investigate the importance of serum bacterial LPS presence in patients with HBV, HCV, and liver cirrhosis. Eighty nine patients were included. Significant differences in the mean ages of patients with HBV infection in comparison to cirrhotic liver patients. A higher mean was discovered in cirrhotic liver patients show in Table(1). This result agrees with previous study [14]. Higher mean found in liver cirrhotic patients might possibly due to life style and accumulated exposure to environmental factors together with drinking alcohol and being exposed to harmful substances, these factors all contribute to the progression of persistent liver inflammation which may result in cirrhosis [15].

Higher frequency of patients with HBV and HCV were found under 40 years of age whereas higher frequency of patients with liver cirrhosis were above 40 years of age show in Table 2. Similar results were documented in previous studies [16] [17] [18]. More than 75 % of patients in this study were suffer from chronic disease. Age regarded as a major risk factor for hepatitis virus infection. It was documented that infection during the early age of life increase liver cancer development and the possibility of persistent infection [19]. When HBV or HCV infections are persistent, older age has consistently been reported as an independent risk factor for the development of HCC [20]

In the current study, higher frequency of infection among female was found in case of HBV and HCV infection while lower frequency was found in case of cirrhosis show in Table 3. Higher ratio were

recorded in previous studies concerning HBV, HCV, and cirrhosis [21][22].Sex is considered as risk factor in different types of diseases. Actually, there are many differences between female and male in health and disease status. Longer age was seen in female than in men but usually female are not healthier. Sex differences in immune response has been documented. The immunological response to viral infections was found to be stronger in females. The course of liver disease appears to differ across the sexes, and numerous clinical trials have demonstrated that postmenopausal women and men with Chronic hepatitis caused by HBV and HCV progresses more quickly to cirrhosis [23].

The presence of significantly lower mean of LPS among HBV infected patients might possibly due to immune response manipulation by the virus show in Table4. It has been documented that during Chronic HBV infection, the virus or its component (HBeAg), manipulate a number of mechanisms that prevent IL-1ß from expressing and having a biological effect. Additionally, it prevents the NLRP3 inflammasome is activated by LPS. the generation of IL-1ß, and the activation of NK-B, all of which may encourage the development of chronic infection [24].Interestingly it has been demonstrated that persistent HCV infection causes a considerable elevation in LPS, which is a marker of liver failure. The etiology of the disease and recurrent liver damage are both impacted by the dysbiosis of the gut microbiota in chronic HBV infection [13]

Additionally, dysbiosis of gut microbiome and innate immunological response in the gut are linked to cirrhosis [25]. Pathogen-associated molecular patterns (PAMPs), for instance LPS, are the main cause of systemic inflammation in cirrhosis and are the result of bacterial translocation [26].

This study showed that there was significant positive correlation of LPS with ALP show in Table 5. ALP is considered as liver disease biomarker and is associated with biliary tract disease[27] .Additionally, ALP has been recognized as a standalone prognostic factor for recurrence in patients with (HCC). There has been evidence of a connection between liver pathology and the digestive system. [28]

(ALP) also has a crucial anti-inflammatory function. ALP cause detoxification of bacterial LPS which decreases its action [29]. ALP, a measure of nodular regenerative hyperplasia, suggests that, even in the absence of viral infection, microbial translocation and LPS-induced monocyte or Kupffer

cell activation cause hepatic fibrosis and portal hypertension. Hepatic fibrosis may be caused by increased microbial translocation and the subsequent immune activation, as well as both of these factors[11].

5. Conclusion

Higher mean age of patients with liver cirrhosis and higher frequencies of HBV, HCV infection within the age range of 20 -39 years and the presence of significant difference in the mean age of acute and chronic infection could reflect the impact of age on infection. Higher frequency of infection occurs among female in case of HBV and HCV infection while lower frequency occur in case of cirrhosis.

Significant decrease in LPS mean level was seen in patients with HBV infection in comparison to the two other groups which may reflect the role of this virus in manipulating the immune response to achieve persistent infection. Significant positive correlation of LPS with ALP may support the role of these parameter in disease pathogenesis.

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