Evaluation of Prothrombin Time, International Normalized Ratio, and Activated Partial Thromboplastin Time in Hepatitis B Virus Patients, Sudan2020 -2021

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Abstract:

Background: Hepatitis B virus is a growing problem worldwide leading to increasing incidences of life-threatening complications of which liver diseases play a major part of it.

Materials and Methods: To evaluate these effects in Sudanese patients, a cross-sectional study was carried out in Wad Medani teaching hospital from October 2020 to February 2021 for assessing the effect of the disease on the coagulation profile. Patients were randomly selected according to inclusion and exclusion criteria. 2.5ml of blood samples were collected from each patient in tri-sodium citrate containers, and then semi-automated Coatron M4 was used to estimate Prothrombin Time (PT) and Activated Partial Thromboplastin Time (APTT) after calibrated and controlled setting according to the manufacturer's instructions. SPSS (version 25) was used to analysed data.

Results: A total of 100 hepatitis B virus patients were included in the study, with males representing (79%) and females were (21%), their age were between 18 to 60 years. The study results showed PT prolonged in (98%) of the study population and only (2%) were normal. regarding the APTT (99%) of them had prolonged results (more than the upper normal), and the remaining (1%) was normal. Also, the study results showed there were slight statistically insignificant differences in coagulation profile in hepatitis B virus patients according to gender, age group, family history, and duration of the disease.

Conclusion: hepatitis B infection had a great effect on Coagulation profiles. Therefore, it is recommended on the basis of these results to request PT, INR, and APTT as routine works up for hepatitis B virus patients.

Keywords: Hepatitis, PT, APTT, Coagulation profile, Sudan, and INR.

تقييم معاملات تخثر الدم: زمن البروثرومبين، والنسبة المعيارية الدولية وزمن الثرمبوبلاستين الجزئي المنشط لمرضى فيروس التهاب الكبد الوبائي باء، السودان ٢٠٢٠-٢٠٢١

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الخلاصة:

المقدمة: إن فيروس التهاب الكبد الوبائي باء معضلة صحية متنامية في جميع أنحاء العالم تؤدي إلى تزايد حالات المضاعفات التي تهدد الحياة والتي تشكل أمراض الكبد جزءا كبيرا منها..

المواد والطرق:–أجريت هذه الدراسة في مستشفى ود مدني التعليمي في الفترة من تشرين الأول/أكتوبر 2020 إلى شباط/فبراير 2021 لتقييم أثر التهاب الكبد الوبائي باء على **معاملات تخثر الدم لدى المرضى السودانيين**. وتم اختيار المشاركين عشوائياً وفقاً لمعايير الإدماج والاستبعاد. تم جمع 2.5 مل من عينات الدم من كل مريض في حاويات سيترات ثلاثية الصوديوم، ثم استخدم جهازالكواترون لاختبار زمن البروثرومبين وزمن الثروموبلاستين الجزئي المنشط بعد وضع المعايرة والمراقبة وفقا لتعليمات الصانع. واستُخدمت النسخة 25من برنامج الحزمة الإحصائية للعلوم الاجتماعية في تحليل البيانات.

النتائج: أُدرج في الدراسة ما مجموعه 100 من مرضى فيروس التهاب الكبد الوبائي من الفئة باء، ويمثل الذكور (79 في المائة) والإناث (21 في المائة)، وتتراوح أعمارهن بين 18 و60 سنة. وأظهرت نتائج الدراسة أن زمن البروثرومبين كان مطولا في
(98 في المائة) من الذين شملتهم الدراسة.
وأن نتائج زمن البروثرومبين كان طبيعية في (2 في المائة) فقط من المشاركين. بينما نتائج زمن الثرمبوبلاستين **الجزئي المنشط كانت** مطولة (أعلى من الحد الطبيعي الأعلى) في (99 في المائة) من المشاركين، وكانت طبيعية في النسبة المتبقية (1 في المائة) من المشاركين. كما أظهرت نتائج الدراسة وجود فروق طفيفة من الناحية الإحصائية في معاملات التجلط لدى مرضى فيروس التهاب الكبد الوبائي باء حسب نوع الجنس، والفئة العمرية، والتاريخ المرضي للأسرة، ومدة المرض.

الخاتمة: كان للإصابة بالتهاب الكبد الوبائي باء تأثير كبير على معاملات التجلط. وعلى أساس هذه النتائج يوصى بأن يطلب اجراء تحليل معاملات التجلط بصورة روتينية لمرضى فيروس التهاب الكبد الوبائي باء.

الكلمات المفتاحية: التهاب الكبد، التهاب الكبد الوبائي باء، زمن البروثرومبين، النسبة المعيارية الدولية، زمن الثرمبوبلاستين الجزئي المنشط، معاملات تخثر الدم.

Introduction:

individual could either be carrier without having clinical signs or active with apparent signs and disease status

Hepatitis is a condition causes the liver to become inflamed and is indicated by the presence of inflammatory cells in the liver tissues, which can result in fibrosis or cirrhosis.(1) However, Hepatitis B virus (HBV) infection represents a global public health problem with over 2 billion people worldwide being exposed to the virus continues to cause more than one million deaths annually.(2) Individual could either be carrier without having clinical signs or active with apparent signs and disease status (2)(3).There is a wide range of clinical presentations associated with chronic HBV infection, including asymptomatic infections to severe diseases. carrier state to chronic hepatitis, fibrosis, and in the worst cases liver cirrhosis and hepatocellular carcinoma (HCC).(3) Hemostasis is the stoppage of bleeding or is a process of blood clot formation at the site of vessel injury, primary hemostasis (for platelets plug formation), and secondary hemostasis (for activation of coagulation cascade) (4). The prothrombin time (PT) was developed by Armond Quick in 1935 in order to measure the extrinsic coagulation system. The prothrombin time (PT) is performed by adding a crude preparation of Tissue factor (TF) to citrate-anticoagulated plasma, recalcification of the plasma, and measurement of the clotting time.(5) A deficiency of factor(s) of the extrinsic coagulation pathway may result in prolonged prothrombin time e.g. VII(most likely to be deficient), II, V, X, and/or

fibrinogen.(6) The assay of coagulation factor deficiencies depends on the type of thromboplastin used because each thromboplastin has a different sensitivity.(7) Once the international sensitivity index (ISI) of the thromboplastin is assigned, the results can be reported as the international normalized ratio (INR).(8) The INR has two major advantages: it allows comparison between results obtained from different laboratories and it will enable investigators to standardize anticoagulant therapy in clinical trials and scientific publications.(9)The Activated Partial Thromboplastin Time (APTT) involves factors of both the intrinsic and common pathways. The APTT may be prolonged as a result of a deficiency of one or more of these factors or of the presence of inhibitors that affect the functions of the factor(s) or the phospholipid reagents.(10) In patients with hepatitis B virus, The progressive loss of hepatic parenchymal cells as a result of increased apoptosis caused by Oxidative stress secondary to increased production of reactive molecules leads to hepatic dysfunction, and among all these complications, hemostatic alterations have been considered for a long time, the principal cause of bleeding in cirrhotic patients.(11) The loss of hepatic function following HBV infection could arise from hepatic inflammation caused by HBX (hepatitis B Virus X protein) which is a major viral trans activator and induces many pro-inflammatory cytokines including interleukine-18 (1L-18).(12) Many Previous Studies have reported prolongation of PT and APTT following viral hepatitis with dysfibrinogenemia and thrombocytopenia. This is because of the loss of the integrity of both the extrinsic pathway of coagulation and the factors of the common pathway.(13) And the degree of prolongation of this clotting time has been suggested to correlate to the degree of deficiency or inhibition of extrinsic or common pathway clotting factors, according to the severity of the liver disease.(13) The present study aimed to measure the prothrombin time, international normalized ratio, and activated partial thromboplastin time in hepatitis B virus patients, in Gezira state, Sudan. Limited studies were performed to evaluate the Prothrombin Time, International Normalized Ratio, and Activated Partial Thromboplastin Time in Hepatitis B Virus Patients in Sudan, moreover this study expected to fill such gap in the literature.

Materials and Methods

Study design:

This was a cross-sectional hospital-based study. Aimed to measure prothrombin time, international normalized ratio, and activated partial thromboplastin time in hepatitis B virus patients, in Gezira state, Sudan.

Setting and duration: The study was carried out in Wad Medani teaching hospital, Gezira state from October 2020 to June 2021.

Study population and sampling technique:

Hepatitis B virus patients attending the Wad Medani teaching hospital, according to inclusion and exclusion criteria, 100 samples of hepatitis B virus patients in Wad Medani teaching hospital by randomized sampling technique.

Inclusion and Exclusion criteria:

Patients with hepatitis B virus in both sexes aged group above 18 years were included. Patients with chronic hypertension and diabetes, individuals from a liver surgical operation, any patients under warfarin or heparin treatment within the last three months prior to collection day, alcohol intake individuals, and Patients with Age group>65 years were excluded.

Samples and data collection:

data was collected by using a questionnaire, which was designed to include all the needed information. Blood sample collection was withdrawn by clean vein puncture technique, 1.25 ml of venous blood was dispensed in 0.11 mol/l tri sodium citrate anticoagulant container, and blood was mixed by mixers machines, and the container was labeled. Then semi-automated Coatron M4 was used to estimate PT and APTT after calibrating and controlling the setting according to the manufacturer's instructions.

Data analysis:

Data was analyzed by using Statistical Package for Social Sciences (SPSS) computer program (version 25).

Ethical consideration:

Ethical clearance was taken from the Ministry of Health, Gezira state. Ethical permission for this study was obtained from the agreement of the Wad Medani teaching hospital. Furthermore, Informed consent was obtained from participants before the collection of samples. Also, the specimens and information were collected from the individuals in privacy. Each individual included in this study was given his/her test results.

Results

A total of 100 patients with hepatitis B virus were included in this study, among them 79% were males and 21% were females. This study included 100 patient's hepatitis B virus. The study presented 28% of patients in age between (18-28 years), 30% between (29-39 years), and 42% (more than 39 years). As shown in figure (1). In this study, 82% have no family history and 18% of patients have a family history. This study included 100 patient's hepatitis B virus. The study presented (8%) of patients in 6 months, (9%) in8 months, (9%)in 10 months, (25%) in 1 year,(20%) in 2 years, (16%) in 3 years, (12%) in 4 years, and (1%) in 5 years. In this study 92% were carriers and 8% of patients have diseases. As shown in Figure (1). In this study 2% with normal PT and 98%with prolonged PT. As shown in Figure (2). In this study 2%with normal INR and 98% with high INR. As shown in Figure (2). In this study 1% with normal APTT and 99% with prolonged APTT. As shown in Figure (2).

In this study, there were no significant differences in the mean values of PT between males and females (23.8 vs 23.4). However, the APTT and INR mean values were slightly increased in the females. Moreover, the chronic disease status was associated with higher mean values in all the tests. Old age (> 39) was also associated with higher values for APTT and INR while the opposite was observed when it comes to PT values with patients less than 28 years having slightly higher mean values (24.4) than patients over 39 years (23.5). (Table 1, 2, 3).

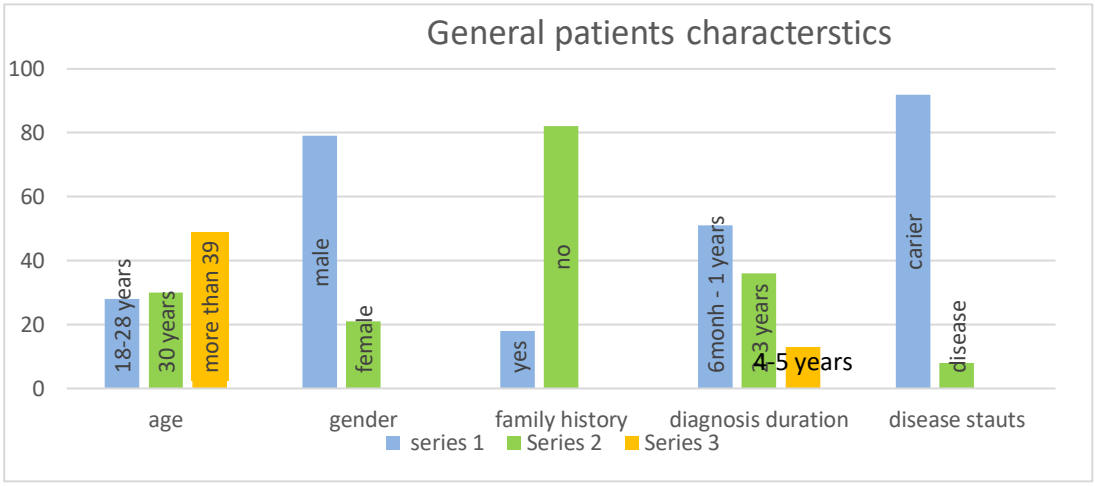

Figure (1): General characteristics of hepatitis B patients.

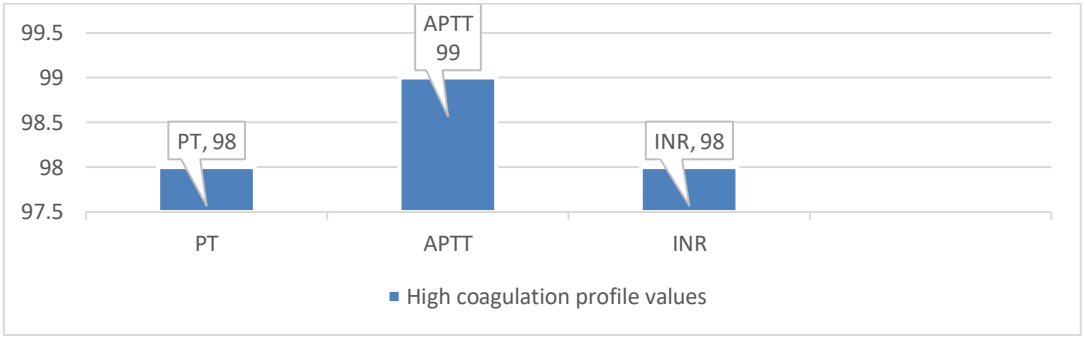

Figure (2): percentage of hepatitis B patients with high coagulation profiles results

Table (1): Comparison of PT with different variables in hepatitis B virus patients

			Mean	SD	Sig
PT	Age group	18 – 28	24.436	6.3843	0.646
		29 – 39	23.433	6.4124	
		>39	23.507	6.1432	
	Gender	male	23.828	6.1121	0.309
		female	23.433	7.4264	
	Duration	6month	21.737	5.1035	0.814
		8month	27.656	9.4962	
		10month	24.678	6.3910	
		1 years	23.468	5.2693	
		2 years	21.930	6.5124	
		3 years	23.619	6.3580	
		4 years	25.058	6.3258	
		5 years	5.700	6.3719	

	Family history	Yes	23.261	6.9450	0.234
		No	23.851	6.2799	
	Diagnosis	chronic	23.916	6.4585	0.674
		Acute	21.775	5.2087	

Table (2): Comparison of INR with different variables in hepatitis B virus patients

			Mean	SD	Sig
INR	Age group	18 – 28	1.889	0.6014	0.244
		29 – 39	1.810	0.5714	
		>39	2.224	2.7214	
	Gender	male	1.837	0.5412	0.309
		female	2.634	3.8260	
	Duration	6month	1.663	0.4406	0.318
		8month	2.167	0.8500	
		10month	1.889	0.5776	
		1 years	1.804	0.4486	
		2 years	1.657	0.5757	
		3 years	2.906	4.3524	
		4 years	1.975	0.5895	
		5 years	2.000	1.8157	
	Family history	Yes	2.772	4.1202	0.503
		No	1.838	0.5546	
	Diagnosis	chronic	2.037	1.8865	0.674
		Acute	1.650	0.4536	

Table (3): Comparison of APTT with different variables in hepatitis B virus patients

			Mean	SD	Sig
APTT	Age group	18 – 28	54.971	6.6224	0.273
		29 – 39	57.612	6.8274	
		>39	56.700	6.8843	
	Gender	male	56.165	6.2867	0.604
		female	57.714	8.5463	
	Duration	6month	53.850	5.0850	0.177
		8month	56.056	7.4209	
		10month	59.022	7.1391	
		1 year	56.076	6.8933	
		2 years	59.165	7.3792	
		3 years	55.400	6.9269	
		4 years	54.233	5.2536	
		5 years	60.100	6.8042	
	Family history	Yes	56.456	8.4483	0.638
		No	56.498	6.4502	
	Diagnosis	chronic	56.779	6.9625	0.767
		Acute	53.163	3.2880	

Discussion

Liver diseases including Hepatitis B characterized by variable hemostasis defects that affect primary homeostasis, fibrinolysis, and coagulation. The extent of

coagulation abnormalities depends upon the severity of the disease. (14)

A total of 100 hepatitis B virus patients were enrolled in this study according to inclusion and exclusion criteria, (79%) of them were males and (21%) were females, with ages varying from 18-60 years.

Furthermore, (18%) of patients have a family history of infection, whereas (82%) of them were not reported to have a family history of infection. (Figure 1)

The present study revealed that (92%) of patients are carriers of the Hepatitis B virus and 8% have the disease.

(PT, INR, and APTT) are prolonged in patients with hepatitis B virus due to Vit K deficiency and liver damage mediated by the effect of tumor necrosis factor and IL-18 among other related factors. While PT was normal in (2%) of patients and prolonged in (98%) of them. INR is normal in (2%) of participants and high in (98%). in addition, APTT is found to be normal in (1%) and prolonged in (99%) of patients (Figure 2). These findings correlate well with another study done by Fazaa et al. to evaluate PT and APTT in Iraq that found all the results were significant in the disease group when compared with the control group.(15) Moreover, the HBV-DNA load and blood clotting test have a positive association P value (>0.05).(15)

Present study results also revealed that there is no significant association between (PT, INR, and APTT) with gender, age group, family history, duration, and diagnosis P value (>0.05)

(Tables: 4-1 4-2 4-3). In 2014 a study was done in Nigeria, which included 115 patients and 50 healthy control. It assessed the effect of viral hepatitis A, B, and C on several liver parameters and showed slightly different mean PT values (22.3 in the Umuahia study vs 22.6 in the current study) and lower level of mean APTT values compared to the current study (42.5 vs 56.4). (12) These differences in APTT could be due to the difference in the underline methodology and reagents used to assess the APTT values in these studies. (16) However, further studies are needed to confirm his hypothesis.

Similarly in 2020 in a study at Affiliated Hospital in Chongqing, China. Wang et al measured PT and APTT in hepatitis B patients. It included (266) cases which unlike our study were composed of only 53% males vs 70% in our current study (Figure 1). Results showed the mean PT was 14.83±1.55 sec (P value <0.05). The mean APTT was 37.25±6.59 sec (P value <0.05).(17) The dissimilarity between these results and the current study could be attributed to the added effect of hepatocellular carcinoma in this group of patients.

In addition, in 2021 a study at Emergency County Clinical Hospital Sibiu, Romania. Vecerzan et al also measured PT and APTT in chronic liver disease patients. It Included 59 patients and 62 control. 19 of them were diagnosed with hepatitis and 40 with cirrhosis. The coagulation profile was analyzed by an automated coagulation analyzer. The result showed a mean PT of 12.30±5.5 sec (P value <0.001). The mean APTT was 30.90 ±8.8 sec (P value <0.022) with a mean INR of 1.01. (18) it is worth noting that only 8% of the included patient sample had hepatitis B virus which could explain the overall difference in mean values of coagulation profile with our study that demonstrates higher coagulation values in chronic hepatitis B patients as noted in table (1.2. and 3) . This study is limited by the lack of a control group, however, it still provides valuable insight into the effect of the hepatitis B virus on the coagulation profile in Sudanese patients, as it is the first published Sudanese study covering this specific objective as far as we know.

Conclusion

Most of the study population have prolonged PT, INR, and APTT, moreover, the study results showed there were slight

statistically insignificant differences in the coagulation profile of hepatitis B virus patients according to gender, age group, family history, and duration of the disease. Therefore, it is recommended on the basis of these results to request PT, INR, and APTT as routine workups for hepatitis B virus patients.

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