Original Research Article

GUESSTIMATING ANAEMIA IN HEPATIS B AND HEPATITIS C PATIENTS RECEIVING CARE IN UNIVERSITY OF PORT HARCOURT TEACHING HOSPITAL (UPTH).

ABSTRACT

Hepatitis B and C, often known as HBV and HCV, are blood-borne diseases that have a high incidence of illness and mortality. Hematological parameters are crucial in identifying cirrhosis and hepatocellular carcinoma in these patients because these illnesses are responsible for liver ailments. In this study, anaemia in Hepatitis B and Hepatitis C patients undergoing treatment at the University of Port Harcourt Teaching Hospital (UPTH) will be estimated using a quick diagnostic test. A cross-sectional hospital research will be used to screen for hepatitis B and C patients on routine screening by rapid diagnostic test. There were 85 participants in the trial, of whom 20 had Hepatitis C, 40 had Hepatitis B, and 25 were in the control group (healthy subjects). The patients' blood was obtained for testing (study group and control group). The study group in the HBV positive significantly differed from the control group. At $p<0.05$, PLT (222.13± 60.5) and (257.3± 84.7), LYM (37.11 ± 9.48) and (45.34 ± 10.4), and NEUT (55.21 ± 10) and (46.24 ± 9.64) respectively for both study and control groups showed statistically significant differences. There was no discernible difference between the HCV positive study group and the HCV control participants in the haematological measures at $p<0.05$ (control). Variations in cell morphology were seen in both HBV- and HCV-positive participants as well as control subjects. 9 of the 31 HBV positive study participants displayed microcytic hypochromic film appearance, while the microcytic normochromic film appearance was seen in 31. A microcytic normochromic film appearance was seen in the 12 HCV positive study group, while a microcytic hypochromic film appearance was seen in the 8 groups. All of the control subjects, with the exception of 1, had a normocytic normochromic film. The study has shown that persons with HBV and HCV infections may exhibit microcytosis, hypochromia, anisocytosis, and poikilocytosis on film readings despite having normal results for haematological indicators.

KEYWORDS: Guesstimation**,** Anaemia, Hepatitis B virus (HBV) and Hepatitis C virus (HCV)

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INTRODUCTION

Owing to the fact that the hepatitis B virus (HBV) infects over 350 million people while the hepatitis C virus (HCV) affects 150 million people worldwide, the number of people with viral hepatitis is alarming and has become a significant public health concern (WHO, 2008; WHO, 2011). Hepatitis B virus (HBV) and hepatitis C virus (HCV) infections are widespread in Nigeria and sub-Saharan Africa, with estimates of the numbers of patients reaching around 35 million and 75 million, respectively (Modi and Feld, 2007; Madhava, *et al.*, 2002).

Blood-borne illnesses with a high morbidity and fatality rate include hepatitis B and C. The hepatitis B and C viruses are members of the Hepadnaviridae and Flaviviridae families, respectively (Washington *et al*., 2006; Cox *et al*., 2005).

Infections with the hepatitis B virus are a serious health issue that are more common in injecting drug users (IDUs). Age, gender, sexual activity, shared syringe use, length of addiction, incarceration, tattoos, prior history of surgery, dental surgeries, blood transfusion, jaundice, kind of illicit drug use, and degree of education are some of the risk factors associated with the hepatitis B virus (Ajugwo *et al.*, 2017).

When the hepatitis B virus infects the liver of hominoids, including humans, it results in hepatitis, also known as serum hepatitis at first. They produce liver conditions, which varies in severity depending on the individual (Barker *et al*., 1996; Ganem and Prince, 2004) Asymptomatic infection caused by the hepatitis C virus might subsequently progress to chronic hepatitis, which is typically brought on by either impaired liver function or increased pressure in the liver circulation (Wrong *et al*., 1995). Reused syringes, needles, multi-use drug bottles, and infusion bags all contribute to the cross-contamination that results in the spread of the hepatitis C virus (Alter, 2007).

Extrahepatic abnormalities, which are widespread, frequently accompany the hepatic disease brought on by the hepatitis B and C virus (Ikram *et al*., 2004). Hematological abnormalities are one of the most frequently identified extrahepatic abnormalities identified at the time of diagnosis (Fasola *et al*., 2009). Determining hematological parameters is crucial in patients with cirrhosis, hepatocellular carcinoma and other liver illnesses caused by the hepatitis B and C viruses. The current study's objective was to evaluate the variation in hematological parameters in hepatitis B and C patients with that of healthy individuals.

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2. MATERIALS AND METHODS

2.1 Study Design

This cross-sectional study was conducted in a hospital setting at the Port Harcourt Health Center (University of Port Harcourt Teaching Hospital). The intended scope of the work includes determining the following haematological parameters: mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), packed cell volume (PCV), red cell count (RBC), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), and to examine peripheral blood smear to assess the incidence

of anemia in hepatitis. The total and differential white cell counts, as well as the platelet count, will be taken into account.

2.2 Study Area

The city of Port Harcourt in Rivers State served as the site for this investigation. Nigeria's southern region includes the state of Rivers. This study was conducted at the University of Port Harcourt Teaching Hospital (UPTH).

2.3 Study Population

85 participants in total, ranging in age from 20 to 60, made up the study's population. 60 subjects served as the case study group, and 25 subjects served as the control group.

2.4 Data Collection

The following demographic and clinical data was collected using a specially designed questionnaire: name, age, knowledge of Hepatitis B and C, history of Hepatitis B and C, routes of transmission, knowledge of vaccinations and type of vaccines taken, sexual activity and behavior (multiple sex partners), use of drugs, history of blood transfusion, and sharing of sharp objects.

22.5 Sample Collection

After pre-test counseling and explanations, venous blood was taken from the patients' antecubital fossa using a vacutainer in accordance with Cheesebrough's (2010), each individual had 5ml of blood drawn aseptically using the venipuncture procedure and placed it in EDTA anticoagulant sample bottles.

2.5 Methodology

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2.5.1 Determination of Rapid Diagnostic Tests for Hepatitis B and C Surface Antigens

Method: Rapid Diagnostic Kit Method

Following the leaf insert as prescribed by the manufacturer, before testing, the sample and test kits were brought to room temperature. The cassette was removed from the foil-wrapped pouch Placing on a spotless, level surface. Three drops of serum was added using a pipette. The results were read after 15 minutes. Results obtained more than 20 minutes later were deemed invalid. In the test region, if a colorful line appears, it signifies the result was successful; if not, it means the result was unsuccessful.

2.5.2 Automated estimation of the whole blood count (Sysmex KX-21N Haematology Analyzer)

Method: **Automation of the Sysmex XE-2100 Haematology Automated Analyzer.**

The roller mixer was used to mix all of the blood samples that had been collected and placed in EDTA vials. The device was then serially updated with the laboratory number for each blood sample. Also, before using the sample probe to aspirate the blood samples, the samples were gently shaken by hand (Orifice). At a chilly temperature, the machine was turned on and allowed to boot up. The equipment appeared to be ready for operation when a green light appeared. A new sample was tested twice, and the first result served as the control for the day. The results were printed out by the instrument and noted.

2.5.3 Determination of Peripheral Blood Film

Method: Making of a Thin Film

On one end of a clean grease-free slide, a drop of blood was inserted. At an angle of 450, the spreader was brought into contact with the drop of blood and allowed to spread uniformly. To

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obtain a thin film, the spreader was moved forward gently and steadily. The slide was labeled and left to dry naturally.

Staining and Viewing of the Thin Film

Leishman stain was applied to the blood film, and it was left on for two minutes. pH 6.8 buffered water twice the volume was added and let to sit for 10 minutes. Water from the faucet was used to remove the discoloration. The slide's back was cleaned, then it was set on the draining rack to let the smear air dry. An oil immersion and X100 objective were used to see the slide.

2.6 Statistical Analysis

Simple percentage calculations were used to statistically assess the data collected.

3 Results

A total of 85 patients were screened, 32 of whom were men and 52 of whom were women. The analysis's findings revealed that 20 people tested positive for Hepatitis C and 40 respondents tested positive for Hepatitis B. Using Microsoft Excel and a 95% confidence interval, an independent t-test was performed on the outcomes of the analysis. Findings are presented using Mean Standard Deviation. The specifics are provided on Table.

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Table 1 Total Number of Screened Subjects for Hepatitis B, Hepatitis C And Controls in Relation to Gender.

	Hepatitis B	Hepatitis C	Control	Total
MALE	14	9	9	32
FEMALE	26	11	16	52
TOTAL	40	20	25	85

The average values of the haematological parameters in the Hepatitis B positive subjects and the control subjects are contrasted. PLT, LYM, and NEUT all showed statistically significant differences (222.13± 60.5 g/dl, 37.11± 9.48 g/dl, and 55.21± 10 g/dl, respectively) at p 0.05. however, there was no discernible change in the other factors.

Table 2: Comparison of Mean ± SD Haematological Parameters between HBV Positive Vs Control Subjects

	PCV %	RBC (×10^6 /µl)	HB (g/dl)	MCV (fl)	MCH (pg)	MCHC (g/d)	PLT	WBC	NEUT	LYM	MXD
HBV positive n=40	36.23 ± 5.01	4.93 ± 0.70	11.78 ± 1.62	84.17 ± 11.27	27.35 ± 3.71	32.51 ± 0.83	222.13± 60.5	7.39 ± 2.01	55.21 ± 10	37.11 ± 9.48	7.47 ± 2.79
Control n=25	36.68 ± 4.71	4.21 ± 0.55	11.83 ± 1.77	88.42 ± 9.29	28.40 ± 3.26	32.14 ± 1.20	257.3± 84.7	8.49 ± 3.85	46.24 ± 9.64	45.34 ± 10.4	7.42 ± 2.72
P-value	0.713	0.242	0.907	0.105	0.237	0.180	0.033	0.658	0.0007	0.02	0.943

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Remark	NS	NS	NS	NS	NS	NS	S	NS	S	S	NS

Key

NS = not significant

S = significant

Average levels of red cell indices between male and female subjects with hepatitis C. At p0.05, PCV and HB exhibited a statistically significant difference, but the other variables did not.

Table 3 Comparison of Mean ± SD Red Cell Indices between Male and Female Hepatitis C Positive Patients

	PCV %	**RBC (×10^6/µl)**	**HB (g/dl)**	**MCV (fl)**	**MCH (pg)**	**MCHC (g/d)**
Male n=9	38.33 ± 3.12	4.43 ± 0.57	12.43 ± 0.96	88.78 ± 8.36	28.60 ± 2.47	32.54 ± 1.21
Female n=11	33.27 ± 2.86	3.92 ± 0.58	10.81 ± 0.76	87.94 ± 12.37	28.16 ± 3.42	32.53 ± 0.99
P-value	0.002	0.066	0.0009	0.860	0.740	0.973
Remark	S	NS	S	NS	NS	NS

Key

NS = not significant

S = significant

Examination of the frequency of cell mutations in those who have Hepatitis B and C in comparison to the control group. 9 HBV positive participants had microcytic hypochromic film

appearance, compared to 31 HBV positive subjects who had microcytic normochromic film appearance. The microcytic normochromic film appearance was seen in 12 HCV positive participants while the microcytic hypochromic film appearance was seen in 8. While 11 HBV and 3 HCV positive participants displayed anisocytosis, 8 HBV positive subjects displayed poikilocytosis. All of the control subjects, with the exception of 1, had a normocytic normochromic film look.

Table 4 Comparison of Cell Variations in HBV, HCV and Control Subjects

	Microcytic Normochromic	**Microcytic Hypochromic**	**Normocytic Normochromic**	**Normocytic hypochromic**	**Poikilo-Cytosis**	**Aniso-cytosis**
HBV	31 (72%)	9 (53%)	0	0	8 (100%)	11 (79%)
HCV	12 (28%)	8 (47%)	0	0	0	3 (21%)
Control	0	0	24 (100%)	1 (100%)	0	0
Total	**43**	**17**	**24**	**1**	**8**	**14**

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Discussion

The results of this study showed a substantial difference between HBV positive participants and control subjects in PLT, NEUT, and LYMP. In HBV positive subjects, the results revealed

higher neutrophil levels and reduced platelet and lymphocyte counts. As the body fights off the infection, a high neutrophil number is likely caused by a variety of physiological states and illnesses. Low levels of lymphocytes are a sign of viral infection, like the chronic hepatitis B infection. This contradicts the findings of Zuberi *et al*. (1997) research, which showed that lymphocyte proliferative stimulation causes an increase in lymphocytes during viral infection.

Additionally, it contradicts Al-(2012) Jaifry's assertion that lymphocyte production increases greatly when a virus is present because this is when antibodies are produced to neutralize the virus. The results of this study, which showed a high neutrophil count, are consistent with those of Badu (2016), who found a high neutrophil count in people who had tested positive for hepatitis B. In the aftermath of a viral infection in the local microenvironment, neutrophils—the first line of defense against a viral invasion—increase substantially, according to Lindemans *et al*. (2006) research.

With regards to gender, there was a significant difference in the PCV and HB concentration of Hepatitis C positive males and females with the males having slightly higher mean values (38.33 ± 3.12 for PCV and 12.43 ± 0.96 for HB) than females (33.27 ± 2.86 for PCV and 10.81 ± 0.76 for HB). Hepatitis B positive males also showed a significant difference in PCV (39.86 ± 5.26), HB (12.97 ± 1.65), MCV (90.89 ± 4.99) and MCH (29.44 ± 1.38) values having slightly higher mean values than females with mean PCV (34.27 ± 3.65), HB (11.13 ± 1.21), MCV (80.56 ± 3.84) and MCH (26.12 ± 3.96).

As numerous studies have demonstrated, men have larger red cell indices than women, particularly women who are menstruating, this is likely the result of the influence of sex

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hormones. This result is in line with that of Ibeh *et al*. (2016), who discovered decreased RBC, PCV, and HB in females.

In the blood film, 12 patients with HCV infection and 31 HBV positive participants both had microcytic normochromic cells. Nine HBV positive participants and eight HCV positive subjects both had microcytic hypochromic cells. Blood films from control subjects showed that the majority of the cells were normocytic, normochromic, while one subject also had hypochromic cells. Even when red cell indices are within the normal range in people with microcytic hypochromic look, this could be a sign of iron shortage. Inadequate iron intake from the diet may be a potential cause of microcytosis. This demonstrates that if patients are not appropriately watched, they may develop anemia.

This result is consistent with the research done by Badu (2016), who found hypochromia on peripheral blood films taken from HBV-positive individuals. Microcytosis and hypochromia may be explained by an increase in hepcidin, which the liver releases in response to interleukin 6 and is seen in inflammatory diseases. Hepcidin controls iron homeostasis, and when it is overexpressed in inflammatory disorders like hepatitis B and C, it prevents iron from entering the bloodstream and causes an iron deficit.

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Conclusion

Microscopy indicated anomalies in blood film, thus there is a potential to become anemic if the disease is not well monitored and controlled, even if the study found no significant difference in the haematological indices of Hepatitis participants, the film reading of these patients showed microcytosis, hypochromia, anisocytosis, and poikilocytosis.

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