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Research Article

Assessment on liver function biomarkers in HIV positive pregnant and Non-pregnant women on Antiretroviral therapy in Rivers State, Nigeria

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Abstract

Background: The use of Antiretroviral Therapy (ART) has greatly improved the health and lifespan of people living with HIV, however, hepatic dysfunction has been associated with HIV. This study investigated the liver function biomarkers of women living with HIV and who are on ART.

Methods: A cross-sectional study was conducted on HIV-positive pregnant women and HIV-positive non-pregnant women in Rivers State, Nigeria. A total of 330 women between 15-60 years participated in this study. HIV-negative pregnant and non-pregnant women served as a control to the test subjects. Sociodemographic data were collected using a well-structured questionnaire. Blood samples were collected for biochemical assay of the liver function biomarkers. The subjects were on Tenofovir-Lamivudine-Efavirenz (TLE) antiretroviral therapy. Data were analyzed statistically on IBM SPSS Version 25 using student's *t* - test, ANOVA and compared using the Post hoc test.

Results: The results obtained showed a significant increase at $p \leq 0.05$ in the ALP, ALT and AST levels of both HIV-positive pregnant and non-pregnant women when compared to the control group. The serum TP level of HIV-positive pregnant women decreased in comparison to the control group. However, the decrease was not statistically significant at $p \leq 0.05$. A statistically significant increase in the total protein level was observed in the HIV-positive non-pregnant women when compared to the control group. The albumin level in HIV-positive pregnant women had a statistically insignificant increase in comparison to the control group. HIV-positive non-pregnant women had a decreased level of Albumin at $p \leq 0.05$ in comparison to the control group.

Conclusion: The findings of the study suggest that the HIV-positive pregnant and non-pregnant women on ART are prone to adverse alterations in their liver function biomarkers in comparison to the control groups, hence they are vulnerable to liver dysfunction. Thus proper care and regular investigations should be carried out on HIV-positive women on ART.

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Introduction

The use of Antiretroviral therapy has significantly improved the health and lifecycle of People Living with HIV (PLWH) [1,2]. Albeit, abnormal liver function biomarkers levels, liver dysfunction, and HIV have been closely linked to each other [3]. Reports have shown a greater percentage of HIV infection and liver dysfunction in women [4]. The physiology, comorbidities, and biopsychosocial issues that contribute to altered rates of liver dysfunction and fibrosis in HIV-positive patients are recently been understood [5].

ALT, AST, ALP, Total protein, and Albumin are liver function biomarkers that have been employed in the laboratory test for investigation of the integrity of the liver [6,7]. ALT, ALP and ALT are used in the evaluation of a range of diseases, including Liver diseases [8].

Albumin which is synthesized in the liver experiences a fall when there is any liver disease, however, if the liver function is normal and serum albumin is low, it may reflect malnutrition or protein loss [6].

This study aimed to investigate the impact of ART on the liver cells in HIV-positive pregnant women and HIV-positive non-pregnant women on ART receiving care at Rivers State University Teaching Hospital, Port Harcourt, Nigeria. The information derived from this study will aid improve the care given to HIV-positive women as it pertains to the integrity of the liver cells, thus enhance healthier living in HIV-positive women.

Methods

Study site

The study was conducted at the Rivers State University Teaching Hospital formerly known as Braithwaite Memorial Specialist Hospital (abbreviated as BMSH), a government-owned hospital, named after Eldred Curwen Braithwaite, a British doctor and a pioneer of surgery, located in Old GRA, Rivers State.

Study design

Ethical consideration and informed consent: The ethical approval for this research was obtained from the Rivers State University Teaching Hospital ethics committee with REC No: RSUTH/REC/2020023.

Study population/size

This was across–sectional study designed to assess the liver function in HIV-positive women (pregnant and non-pregnant) on ART attending HIV outpatient clinic at Rivers State University teaching hospital, Port Harcourt, Nigeria. The study was carried out between October (2020) – February (2021). The study participants comprised a total of 330 subjects aged 15-60 years out of which 83 were pregnant and 82 nonpregnant HIV-positive subjects who have been receiving ART for over 6 months. 84 pregnant females and 81 nonpregnant HIV-negative subjects regarded as the control group were also recruited for the study. They were apparently healthy individuals recruited among staff and students.

Sample size determination [9]

The sample size for this study was determined by the procedure of (Habib, et al. [10]; Wang, et al. [11,12]) thus:

Considering liver function biomarkers levels among HIV patients on ART of 68.3% at 0.05 error margin the following statistical formula was used to determine the sample size for this study

Where P = Proportion/Prevalance/mean ± SD

e = error margin/precision = 0.05

Z = constant (1.96)

$$N = Z^2 \frac{P(I-P)}{e^2} \quad P = 68.3\% \rightarrow 0.683$$

Z=1.96

$$Z = (1.96)^2 \frac{0.683(1-0.683)}{(0.05)^2}$$

$$N = 3.8416 \text{ x } \frac{0.683(0.317)}{0.0025}$$

N = 3.8416 x 86.6044

N =332.699

≅ = 333 minimum sample size

Exclusion criteria

Individuals with AIDS or any other chronic disease condition such as Patients with a prior diagnosis of diabetes mellitus, hypertension, acute or chronic kidney disease, taking nephrotoxic drugs, pregnant women, and patients with hepatitis B and C were excluded from the study.

Inclusion criteria

The HIV subjects on ART were subjects who have been on TLD-ART regimens corresponding with the Nigerian National guidelines for ART for a minimum of six months.

Data collection and analysis

Administration of questionnaires: Data on the participants' socio-demographic characteristics and medical history were collected using a structured questionnaire. Demographic data included sex, age, residence, level of education, alcohol consumption, marital and employment status [13].

Laboratory procedures

Five millimeters (5ml) of blood was collected aseptically by venepuncture with vacutainer needles and syringes and dispensed into lithium heparin bottles, centrifuged at 5,000 rpm for 5min and the serum separated and stored at 20°c until assayed for ALT, AST, ALP, TP, and Albumin.

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Serum ALT, AST, ALP, Total protein, and Albumin levels were analyzed using the Randox kits from Randox Laboratories Ltd, United Kingdom following the manufacturer's instructions.

Data management and statistical analysis

Codes were assigned to each participant so as to observe strict confidentiality. The results were entered in a secured logbook. The questionnaires and logbooks were treated with strict confidentiality. Data were then entered into Microsoft excel version 10, cross-checked for errors, and then transferred into the Statistical Package for Social Sciences (SPSS) version 25 for analysis. The differences between group means were compared using the analysis of variance (ANOVA). Fisher's Least Significant difference multiple comparisons were used for post hoc analysis for significant ANOVA comparison. Categorical data were compared using the chi-square test. Pearson's correlation was used to correlate variables. Statistical significance was set at $p < 0.05$.

Results

Sociodemographic data

The socio-demographic data were collected with the aid of a well-structured questionnaire and has been reported by Odinga, et al. [13] in a Sociodemographic study of HIV-positive pregnant and HIV-positive nonpregnant women on ''HAART'' in a tertiary hospital in Port Harcourt.

The results obtained showed a statistically significant increase at $p \leq 0.05$ in the liver ALP (19.53 ± 3.887), ALT (4.1205 ± 2.32378) and AST (7.1446 ± 3.40778) levels of HIV-positive pregnant women when compared to the control group; HIV-negative pregnant women: liver ALP (16.76 ± 3.484), ALT (3.8929 ± 2.87078), AST (7.0000 ± 3.39028). Also, the levels of the liver function enzymes ALP, ALT, and AST in HIV-positive non-pregnant women showed an increase; liver ALP (20.51± 4.047), ALT (4.2250 ± 2.30561), AST (7.7561 ± 3.4303) when compared to those of the control group; HIV-negative non-pregnant women; ALP (17.96± 5.321), ALT (3.8293 ± 1.69097), AST (7.2000 ± 3.36174). The serum TP level of HIV-positive pregnant women decreased in comparison to the control group: HIV-negative pregnant women. However, the decrease was not statistically significant at $p \leq 0.05$. A statistically significant increase in the total protein level was observed in the HIV-positive non-pregnant women when compared to the control group of HIV-negative non-pregnant women. The albumin level in HIV-positive pregnant women had a statistically insignificant increase in comparison to the control group (HIV-negative pregnant women). The HIV-positive non-pregnant women had a decreased statistically significant level of Albumin at $p \leq 0.05$ in comparison to the control group (HIV-negative non-pregnant women).

Discussion

This study investigated the impact of ART on the liver in HIV-positive pregnant women and HIV-positive non-pregnant women on ART receiving care at Rivers State University Teaching Hospital, Port Harcourt, Nigeria. A statistically significant difference in the liver ALP level was observed in the HIV-positive pregnant women and HIV-negative pregnant women. The liver ALP level in the HIV pregnant women increased significantly at a 95% confidence interval (19.53 ± 3.887) when compared to the HIV-negative pregnant women (16.76 ± 3.484). The HIV-positive non-pregnant women had a statistically significant increase in the liver ALP level (20.51 ± 4.047) when compared to the HIV-negative non-pregnant women (17.96 ± 5.321). HIV-positive pregnant women had an increased liver ALP level while the HIV-positive non-pregnant women had a decreased liver ALP level. Dey, et al. [14] reported an increase in ALP levels of PLWH when compared to the control group. This agrees with the findings of this study for HIV-positive pregnant women. An increase in liver enzymes in HIV-positive patients as reported by Sterling, et al. [15] is common, however, while the absence of Protease Inhibitors use is associated with elevated AST and ALT levels, an increase in ALP levels is associated with hepatic steatosis (Diabetes Mellitus and Body Mass Index (BMI). Pregnancy in women has been associated with an increase in weight, leading to an increased BMI [16]. The liver ALP result revealed an increased level of liver ALP in HIV-positive pregnant women. A significant association with severe liver outcomes among those with the greatest exposure to stavudine (Zerit), didanosine, or tenofovir disoproxil fumarate (TDF) [17]. The AST serum levels of HIV-positive pregnant women (7.1446 ± 3.40778) were seen to increase statistically at $p \leq 0.05$ when compared to the HIV-negative pregnant women (7.0000 ± 3.39028). Also, the HIV-positive non-pregnant women had a statistically significant elevated AST level (7.756 ± 3.4303) at $p \leq 0.05$ when compared to the HIV-negative non-pregnant women (7.2000 ± 3.36174). (Neto and Lobo [18]; Poles et al. [19]) their studies reported that 90% of those infected with HIV had increased AST and ALT levels, which are indicative of liver dysfunction. The ALT levels as revealed in Table 1 showed a significant increase at $P \leq 0.05$ in HIV-positive pregnant women when compared to HIV-negative pregnant women. Also, the ALT level of HIV-positive non-pregnant women showed a statistically significant elevation in comparison to the HIV-negative non-pregnant women. Elevated liver enzymes have been reported to be associated with factors such as viral hepatitis, fatty liver infiltration, alcohol consumption, HIV cholangiopathy, and medication [20]. An elevation in the liver enzyme ALT has been frequently observed in HIV patients even without viral hepatitis infection [21]. The intake of some drugs and substances synergistically can hamper the functionality of the liver and kidney [22]. The study of (Ryom, et al. [17]; Schiano, et al. [23]; Vispo, et al. [24]) observed that HIV patients exposed to antiretroviral drugs stavudine (Zerit), didanosine, or tenofovir disoproxil fumarate (TDF) has been associated with severe liver outcomes. This study reported a statistically significant elevation at $p \leq 0.05$ in the level of ALT in all HIV-positive groups (pregnant and non-pregnant) in comparison to their control groups (HIV-negative pregnant and non-pregnant). The findings are in consonance with past findings. Abnormal levels in the liver function test parameters; Liver ALP, AST, ALT, TP, Albumin and Bilirubin are potentially treatable as chronic liver diseases [25,26]. Elevated liver enzymes have been reported to be often indicative of inflammation or damage

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Table 1: Liver function biomarkers levels of Study subjects.

Study Group	ALP (IU/L)	ALT (IU/L)	AST (IU/L)	TP (g/l)	ALBUMIN (g/l)
Group 1: HNPW	$16.76^{a} \pm 3.484$	$3.8929^{a} \pm 2.87078$	$7.0000^{a} \pm 3.39028$	$71.99^{a} \pm 10.800$	$39.796^{a} \pm 6.9836$
Group 2: HNNPW	$17.96^{ac} \pm 5.321$	$3.8293^{ab} \pm 1.69097$	$7.2000^{ab} \pm 3.36174$	$69.48^{b} \pm 8.350$	$42.0631^{b} \pm 7.0949$
Group 3: HPNPW	$20.51^{b} \pm 4.047$	$4.2250^{c} \pm 2.30561$	$7.7561^{c} \pm 3.4303$	$73.90^{a} \pm 13.092$	$40.976^{c} \pm 6.9335$
Group 4: HPPW	$19.53^{bcd} \pm 3.887$	$4.1205^{d} \pm 2.32378$	$7.1446^{bd} \pm 3.40778$	$71.70^{abd} \pm 9.185$	$41.120^{abd} \pm 5.9128$

HNPW: HIV-Negative Pregnant Women; HNNPW: HIV-Negative Non-Pregnant Women; HPNPW: HIV-Positive Non-Pregnant Women; HPPW: HIV-Positive Pregnant Women

to cells in the liver, this is because inflamed or injured liver cells leak higher than normal amounts of certain. Chemicals, including liver enzymes into the bloodstream, elevating liver enzymes on blood tests [27,28].

A decrease in the albumin level has been reported to be a consequence of a dysfunctional liver because albumin is synthesized in the liver [6], howbeit, when the liver is functioning normally, it could be a sign of Malnutrition. Serum total protein and Albumin levels are used in the assessment of the nutritional status of patients [29]. A decrease in the serum albumin level in HIV-positive pregnant women was observed when compared to the HIV-negative pregnant women, however, the decrease was not significant at $p \leq 0.05$. The total protein of HIV-positive pregnant women decreased in comparison to the control group: HIV-negative pregnant women. However, the decrease was not statistically significant at $p \leq 0.05$. A statistically significant increase in the total protein level was observed in the HIV-positive non-pregnant women when compared to the control group of HIV-negative non-pregnant women. The albumin level in HIV-positive pregnant women had a statistically insignificant increase in comparison to the control group (HIV-negative pregnant women). The HIV-positive non-pregnant women had a decreased statistically significant level of Albumin at $p \leq 0.05$ in comparison to the control group (HIV-negative non-pregnant women). Total protein and albumin are used as liver function tests to support the diagnosis of liver disorders. HIV and hepatitis are associated with high total serum proteins [30]. A study reported that the density of albumin was significantly associated with HIV status and among HIV-positive patients [31] Serpa, et al. [32] in their study observed that HIV-positive patients on HAART had a significant in Albumin levels after 6-12 months and a reduction in total protein. Busher, et al. [33] reported that the normal serum total protein and albumin levels are 6-8g/dl and 3.5-5.0g/dl respectively.

In view of the findings from this study and the above literature, it is suggested that HIV-positive pregnant and HIV-positive non-pregnant women on ART are more prone to adverse alterations in their liver as shown in the test parameters in comparison to the control groups, hence appropriate care and regular investigations on the liver function is advised.

Conclusion

The liver function biomarkers tested in HIV-positive pregnant women and HIV-positive non-pregnant women showed a significant increase in the levels when compared to their control groups; HIV-negative pregnant women and HIV-negative non-pregnant women. The results and findings of this study suggest a potency of adverse alteration in the liver of HIV-positive women on ART as evidenced in the liver function biomarkers of HIV-positive women on ART, hence vulnerability to liver dysfunction. Thus proper care and regular investigations should be carried out on HIV-positive women on ART.

Authors contributions

Conceptualization: Conceived and designed the experiments: Odinga Tamuno-Boma and Azuonwu, Obioma. Analysis of data: Ihua, Nnenna, and Odinga Tamuno-Boma, Manuscript write-up: Odinga Tamuno-Boma, all authors provided help and financial contribution to the research, and all authors read and approved the final manuscript.

Ethical approval

The ethical committee of the Rivers State University Teaching Hospital ethically approved the research and all study procedures were under relevant guidelines: REC No: RSUTH/REC/2020023.

Informed consent

All the study participant duly gave their informed consent via a completed questionnaire form.

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