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Research Article

Seroprevalence and Associated Risk Factors of Hepatitis E Virus Infection among Pregnant Women in Akure, Ondo State, Nigeria

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About Article

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ABSTRACT

Hepatitis E virus (HEV) infection, an emerging viral disease, can lead to a high rate of fulminant hepatitis, liver failure, and an increased risk of mortality for pregnant women and fetus. This study aimed at determining the prevalence of HEV infection among pregnant women attending antenatal clinic at selected Government Hospitals in Akure city, Ondo State, in order to provide a comprehensive information of the infection on this particular group of people in the city. A questionnaire was administered to the participants who indicated their consent to participate in the study to generate socio-demographical data of the participants and the level of awareness of the infection. A total of 199 blood samples was collected from the recruited apparently healthy pregnant women with no history of immunosuppressive diseases. Plasma of the collected blood was subjected to the analysis of Hepatitis E virus IgM antibody using capture and colloidal gold immunochromatographic double antibody assay and the positive samples were subjected to assessment of liver enzymes using standard assays. The study revealed a prevalence rate of 3.5% of Hepatitis E virus infection among the pregnant women in the study area. Out of the 3.5% that were positive for HEV, the highest percentage frequency of occurrence of the infection (12.50%) was found in the age group > 45 years while the least percentage frequency of occurrence of HEV infection (7.14%) was observed in age group 15-19 years. Washing of hands without soap and water after toilet use, drinking of untreated well water, and eating of left-over foods were found to be risk factors that predisposed the pregnant women to HEV infection. The high level of AST and ALT in the pregnant women is an indication of liver stress. This study showed the prevalence rate of HEV among pregnant women in Akure metropolis, the level of awareness of the infection among recruited pregnant women and factors that can predispose pregnant women to the infection. There is therefore the need for appropriate health authorities to educate the public and especially pregnant women on HEV infection and factors that can predispose them to the infection.

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1. INTRODUCTION

Hepatitis E virus (HEV) infection is an emerging viral disease affecting both humans and animals. It causes liver inflammation. Hepatitis E virus infection is of significant public health concern in many developing and developed countries. Approximately 2 billion people reside in areas where hepatitis E is endemic (Raji *et al.*, 2023). According to the World Health Organization (WHO), there are approximately 20 million HEV infections each year, resulting in 3.3 million symptomatic cases (Songtanin *et al.*, 2023). Eight genotypes of HEV have been identified, of which genotypes 1-4 are the major human-pathogenic genotypes. Genotypes 1 (HEV-1) and 2 (HEV-2) exclusively infect humans and are typically transmitted through contaminated drinking water in developing countries, leading to large outbreaks with high morbidity and mortality rates (Schemmerer *et al.*, 2022). In contrast, genotypes 3 (HEV-3) and 4 (HEV-4) are zoonotic, infecting humans as well as other mammalian species, with swine serving as the main reservoir. Infections with HEV-3 and HEV-4 are commonly associated with the consumption of raw or undercooked meat (e.g., pork and game) or shellfish, resulting in sporadic cases worldwide (Ahmed & Nasheri, 2023). Hepatitis E is primarily transmitted through the consumption of contaminated water or food, making it an important concern in areas with poor sanitation and lack of access to potable drinking water. Infection with the virus can lead to both acute or severe inflammation of the liver resulting in symptoms such as jaundice, abdominal pain, fatigue, and nausea. The infection however is usually self-limiting and resolves within 2– 6 weeks (Ehi Airiohuodion *et al.*, 2022).

2. LITERATURE REVIEW

Although, all age groups can be infected with HEV however according to Qian *et al.* (2023) the infection poses greater risks in pregnant women. They are at a higher risk of experiencing severe complications when infected with the virus, especially during the first trimester (Ajayi *et al.*, 2022). Hepatitis E infection during pregnancy can lead to a higher rate of fulminant hepatitis, liver failure, and an increased risk of mortality for both the mother and the unborn child. Chronic infections elevate viral load and prolonged viremia during pregnancy which contribute to severe liver injury or liver failure in the mother and vertical transmission from mothers to foetus ranging from 30 to 100% (Galiana *et al.*, 2008; Ajayi *et al.*, 2022). Therefore, understanding of the specific impact of Hepatitis E virus infection on pregnant women is important for healthcare professionals as it can help in the development of preventive measures and strategies for managing the disease in this vulnerable population.

In Akure, Ondo State, Nigeria, there is a dearth of information on the knowledge and awareness about HEV and no documented report on the seroprevalence of HEV among pregnant women in the city. This study therefore was carried out in order to know the impact of the infection among pregnant women in the city and environs, the extent of the awareness of the infection among this category of people, the risks factors associated with the infection in order to safeguard the health of expectant mothers and their babies in the community sampled.

3. METHODOLOGY

3.1. Study area

Akure is the largest city and the capital of the Ondo State, which is located in south-western Nigeria. It is located at 5°19 East of the Meridian and 7°25 North of the Equator, respectively. It is about 700 km (430 mi) Southwest of Abuja and 311 km (193 mi) North of Lagos State. It is predominantly a civil service town. The mean annual rainfall is about 1350 mm with the distribution spanning between March and November, relative humidity is about 80% with temperature range between 23°C and 30°C.

3.2. Study population

The target population for the study was pregnant women between the ages of 15-45 years old who attend antenatal clinic at the State Specialist Hospital, Oke-Aro, Akure and Ondo State Primary Healthcare Clinics. A total of one hundred and ninety-nine pregnant women (199) were recruited for the study.

3.3. Ethical consideration and consent

Ethical approval was sought from the Ondo State Health Research Ethics Committee (OSHREC 26/02/2024/631) and from the University of Medical Sciences Teaching Hospital ethical review committee (Insert certificate number if you have it) before embarking on this study. All participants were informed about the study's objectives and purpose and those that gave their consent through the filling of the informed consent form to participate in the study were recruited.

3.4. Data collection

Prior to sample collection, a well-structured questionnaire was administered to the participants to generate their socio-demographic data. Each questionnaire was assigned a distinct participant identification number (PIDN). Participants' biodata, such as age, gender, occupation, symptoms (abdominal discomfort, bloating, nausea, heartburn, etc.), risk factors, and kind of medicine used were included in the questionnaire. All completed questionnaires were reviewed for completeness.

3.5. Sample collection and laboratory analysis

Five milliliters (5ml) of venous blood were collected by venepuncture from each participant into a well labelled 5ml Ethylenediamine tetra-acetic acid (EDTA) bottle. The collected blood sample was mixed properly to avoid clotting of the samples. The sample containers were labelled properly with the date of collection of subjects. To protect and uphold confidentiality in this study, both the collected blood samples and questionnaires filled by the participants were assigned numbers rather than names. The collected blood samples were centrifuged at 3000rpm for 5min to separate the plasma from red blood cells.

3.6. Hepatitis e diagnostic method

The plasma of the blood samples collected from the recruited pregnant women was used for the diagnosis of HEV infection by detecting the presence of HEV-IgM antibody in the plasma using capture and colloidal gold immunochromatographic

double antibody assay as described by the manufacturer of the kit (Micropoint HEV Kit, China).

3.7. Biochemical assays for liver enzymes

Biochemical assays for the evaluation of the liver enzymes present in the blood samples of the pregnant women positive for HEV-IgM antibody were carried out using standard methods as described by Denova-Gutierrez *et al.* (2021). The enzymes evaluated include Aspartate aminotransferase (AST), Alanine aminotransferase (ALT), TBIL, DBIL, ALP, AST, ALT, GGT, TP and Albumin (ALB).

3.8. Statistical analysis

Data were expressed in percentages to calculate prevalence and frequency for categorical variables. Risk factors were linked to seroprevalence of Hepatitis E virus infection also using percentages.

4. RESULTS AND DISCUSSION

4.1. The overall seroprevalence of hepatitis E virus in the pregnant women recruited for the study in akure metropolis

Out of the 199 blood samples collected and analyzed, 7(3.5%) were positive and 192 (96.5%) were negative for Hepatitis E virus (Figure 1).

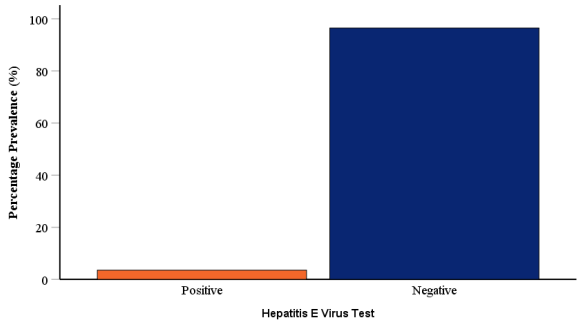

Figure 1. Overall seroprevalence of hepatitis e virus in the pregnant women recruited for the study in akure metropolis

4.2. Level of awareness of hev infection among the pregnant women recruited for the before the survey

Out of 199 pregnant women recruited for the study, 45(22.6%) of them indicated awareness of HEV infection before the survey while 154(77.4%) of them had not heard about HEV hepatitis before the survey (Figure 2).

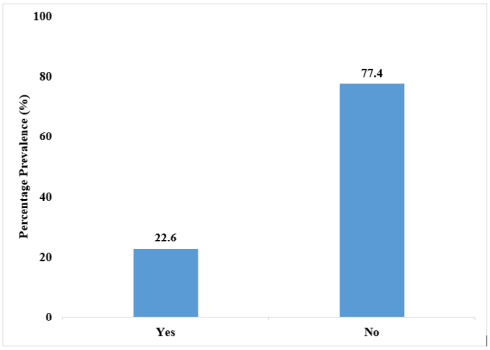

Figure 2. Seroprevalence of hepatitis e virus among the pregnant women (n=199) recruited for the study in akure metropolis based on their hepatitis awareness before the survey.

4.3. Percentage frequency of occurrence of hev in pregnant women positive for the infection based on age group

Out of the 7 pregnant women that were positive for HEV among the pregnant women that were recruited for the study, the highest percentage occurrence of HEV infection (12.50%) was observed in the age group >45 years while the age group with the least frequency of occurrence was 25-29 years (1.93%) (Table 1).

Table 1. Percentage occurrence of HEV infection among the recruited pregnant women in akure based on age group

Age group (years)	No recruited	No. positive (%)	No. negative (%)
15-19	14	1 (7.14)	13 (92.86)
20-24	23	0 (0.00)	23 (100.00)
25-29	52	1 (1.92)	51 (98.08)
30-34	58	2 (3.45)	56 (96.55)
35-39	32	2 (6.25)	30 (93.75)
40-44	12	0 (0.00)	12 (100.00)
>45	8	1 (12.50)	7 (87.50)
Total	**199**	**7 (3.52)**	**192 (96.48)**

4.4. Percentage frequency of occurrence of hev among pregnant women positive for the infection based on educational status

In respect to educational status of the recruited pregnant women that were positive for HEV, the highest frequency of occurrence of HEV (11.76%) was observed in those who do not have formal education, followed by those who had only primary education (28.57%) while least (2.53%) was observed among those who had secondary education and tertiary education, respectively (Table 2).

Table 2. Percentage frequency of occurrence of HEV infection among the recruited pregnant women in akure based on their educational status

Educational background	No recruited	No. positive (%)	No. negative (%)
No formal education	17	2 (11.76)	15 (88.24)
Primary	24	1 (4.17)	23 (95.83)
Secondary	79	2 (2.53)	77 (97.47)
Tertiary	79	2 (2.53)	77 (97.47)
Total	**199**	**7 (3.52)**	**192 (96.48)**

4.5. Percentage frequency of occurrence of HEV among pregnant women positive for the infection based on occupation status

Out of the 199 pregnant women that were recruited for the study, traders had the highest frequency of occurrence of HEV infection (11.11%), followed by the unemployed (3.70%). The least frequency of occurrence of 2.70% was observed among the civil servant group (Table 3).

Table 3. Percentage occurrence of hev infection among the recruited pregnant women in akure based on their occupational status

Occupational Status	No recruited	No. positive (%)	No. negative (%)
Civil/public servant	38	1 (2.70)	37 (97.37)
Self-employed	56	0 (0.00)	56 (100.00)
Unemployed	54	2 (3.70)	52 (96.30)
Farmer	13	0 (0.00)	13 (100.00)
Trader	36	4 (11.11)	32 (88.89)
Others	2	0 (0.00)	2 (100.00)
Total	199	7 (3.48)	192 (96.52)

4.6. Percentage frequency of occurrence of HEV among pregnant women positive for HEV based on trimesters

Out of the 199 participants whose blood samples was collected, 19.60% was negative and 1.01% was positive in their first trimester, pregnant women in their second trimester, 46.73% were negative and 1.51% was positive (Table 4).

Table 4. Percentage Occurrence of HEV Infection among the Recruited Pregnant Women in Akure based on Trimesters

Trimesters	No recruited	No. positive (%)	No. negative (%)
First Trimester	41	2 (4.88)	39 (95.12)
Second Trimester	96	3 (3.13)	93 (96.88)
Third Trimester	62	2 (3.23)	60 (96.77)
Total	**199**	**7 (3.52)**	**192 (96.48)**

4.7. Percentage frequency of occurrence of HEV among pregnant women positive for the infection HEV based on number of pregnancies

Out of the 199 pregnant women that were recruited for the study of which 51 were pregnant for the first time, 88 were pregnant for the second time, 48 for the third time and 12 for the fourth time, 1.96% , 1.14%, 8.33% and 8.33% were seropositive for HEV respectively (Table 5). Showing that the highest frequency of occurrence of HEV was observed in multigravida 3 and 4.

Table 5. Percentage occurrence of HEV infection among the recruited pregnant women in akure based on number of pregnancies

Number of pregnancies	No recruited	No. positive (%)	No. negative (%)
First pregnancy	51	1 (1.96)	50 (98.04)
Second pregnancy	88	1 (1.14)	87 (98.86)
Third pregnancy	48	4 (8.33)	44 (91.67)
Fourth pregnancy	12	1 (8.33)	11 (91.67)
Total	**199**	**7 (3.52)**	**192 (96.48)**

4.8. Percentage frequency of occurrence of HEV among recruited pregnant women in akure based on the type of water they drink

Out of the 199 pregnant women that were recruited for the study, pregnant women who drink well water had the highest frequency of occurrence of HEV infection (4.50%), followed by those who borehole water (2.98%). Those who drink from sources of water like river, stream and rainwater were negative for HEV (Table 6).

Table 6. Percentage occurrence of HEV infection among the recruited pregnant women in akure based on the source of drinking water

Source of water	No recruited	No. positive (%)	No. negative (%)
Well water	111	5 (4.50)	106 (95.50)
Borehole	67	2 (2.98)	65 (97.01)
Stream water	10	0 (0.00)	10 (100.00)
River water	2	0 (0.00)	2 (100.00)
Rainwater	9	0 (0.00)	9 (100.00)
Total	**199**	**7 (3.52)**	**192 (96.48)**

4.9. Percentage frequency of occurrence of HEV among recruited pregnant women in akure based on whether they boil their water before drinking

Out of the 199 pregnant women that were recruited for the study, 54 (27.14%) admitted to boiling their water before drinking. However out of these number, only 3(5.56%) were positive for HEV. On the other hand, out of the 145 (72.86%) who do not boil their water before drinking, 4(2.76%) were positive for HEV (Table 7).

Table 7. Percentage occurrence of HEV infection among the recruited pregnant women in akure based on whether they boil their water before drinking or not

Boiling of water before drinking	No recruited (%)	No. positive (%)	No. negative (%)
Yes	54(27.14)	3(5.56)	51(94.44)
No	145(72.86)	4(2.76)	141(97.24)
Total	**199(100.00)**	**7(3.52)**	**192(96.48)**

4.10. Percentage frequency of occurrence of HEV among recruited pregnant women in akure based on whether they wash fruits with water before consumption

Out of the 199 pregnant women that were recruited for the study, 191 (95.98%) reported that they wash fruits with water before consumption. Out of these number, only 6 (3.14%) were positive for HEV. On the other hand, only 1(12.50%) of those who do not wash their fruit before consumption was positive for HEV (Table 8).

Table 8. Percentage occurrence of HEV infection among the recruited pregnant women in akure based on whether they wash fruits before consumption

Washing fruit before consumption	No recruited (%)	No. positive (%)	No. negative (%)
Yes	191(95.98)	6(3.14)	185(96.86)
No	8(4.02)	1(12.50)	7(87.50)
Total	**199(100.00)**	**7(3.52)**	**192(96.48)**

4.11. Percentage frequency of occurrence of HEV among recruited pregnant women in akure based on how often they eat vegetable salad

Out of the 199 pregnant women that were recruited for the study, for those who often eat vegetable, only 3(4.3%) was positive for HEV while 67(95.7%) was negative. For those who seldomly eat vegetable, 0(0.0%) was positive for HEV while 82(100.0%) was negative while for those who do not take vegetable salad, 4(8.5%) was positive and 43(91.5%) was negative (Table 9).

Table 9. Percentage occurrence of HEV infection among the recruited pregnant women in akure based on how often they take vegetable salad.

How often do you take vegetable salad?	No recruited (%)	No. positive (%)	No. negative (%)
Often	70(35.18)	3(4.29)	67(95.71)
Seldomly	82(41.21)	0(0.00)	82(100.00)
Not at all	47(23.62)	4(8.51)	43(91.49)
Total	**199(100.00)**	**7(3.52)**	**192(96.48)**

4.12. Percentage frequency of occurrence of HEV among recruited pregnant women in akure based on eating of left-over foods

Out of the 199 pregnant women that were recruited for the study, 98 (49.25%) reported that they always eat left over foods. Four (4.04%) out these women were observed to be positive for HEV and 95(95.96%) were negative while out of the 100 pregnant women who reported that they do not eat left over foods, 3(3.0%) were positive for HEV while 97(97.0%) were negative (Table 10).

Table 10. percentage occurrence of HEV infection among the recruited pregnant women in akure based on eating of left-over foods

Do you eat left over food?	No. recruited (%)	No. positive (%)	No. negative (%)
Yes	99(49.75)	4(4.04)	95(95.96)
No	100(50.25)	3(3.00)	97(97.00)
Total	**199(100.00)**	**7(3.52)**	**192(96.48)**

4.13. Percentage frequency of occurrence of HEV among recruited pregnant women in akure based on type of toilet they use

Out of the 199 pregnant women that were recruited for the study, 60 (30.15%) reported that they used pit toilet, 107 (53.77%) use water cistern, 18 (9.05%) using dung hill (shot put) while 14(7.04%) use potty. Three (5.00%) out of those using pit toilet were positive for HEV, 3(2.80%) of those using water cistern were positive for the virus and 1(7.14%) out of those using potty was positive for the virus (Table 11).

Table 11. Percentage occurrence of HEV infection among the recruited pregnant women in akure based on type of toilet

Type of toilet	No. recruited (%)	No. positive (%)	No. negative (%)
Pit toilet	60(30.15)	3(5.00)	57(95.00)
Water cistern	107(53.77)	3(2.80)	104(97.20)
Shot-put	18(9.05)	0(0.00)	18(100.00)
Potty	14(7.04)	1(7.14)	13(92.86)
Total	**199(100.00)**	**7(3.52)**	**192(96.48)**

4.14. Percentage Frequency of Occurrence of HEV among Recruited Pregnant Women in Akure based on Washing of Hands with Soap and Water after Toilet Use

Out of the 199 pregnant women that were recruited for the study, 91 (45.73%) reported that they wash their hands with soap and water after each toilet use, 97 (48.74%) wash their hands only with water while 11 (5.53%) use hand sanitizer. Out of those who was their hands with soap and water, 2(2.20%) were positive for HEV, out of those who wash their hands with only water, 5(5.15%) were positive for the virus while out of those who use hand sanitizer, none was positive for the virus (Table 12).

Table 12. Percentage Occurrence of HEV Infection among the Recruited Pregnant Women in Akure based on washing of Hands with Soap and Water after using the Toilet

Do you wash your hands after each toilet use	No recruited (%)	No. positive (%)	No. negative (%)
With soap	54(27.14)	3(5.56)	51(94.44)
Without soap	145(72.86)	4(2.76)	141(97.24)
With sanitizer	11(5.53)	0(0.00)	11(100.00)
Total	**199(100.00)**	**7(3.52)**	**192(96.48)**

4.15. Liver Enzyme Profiles of the HEV Positive Pregnant Women in Akure

The liver enzyme profiles of the 7 pregnant women positive for HEV revealed increase in AST value above the normal range (0 - 17 iU/L) in all the women (100%) and high level of ALT values in 4 (57.1%) of the women though the values were within the normal range (0 - 40 iU/L) (Table 13).

Table 13. Liver Enzyme Profiles of Recruited Pregnant Women Positive for HEV

Liver enzyme	Patients Number						
	1	2	3	4	5	6	7
TBIL	0.8*	0.2	0.7	0.8*	0.2	0.3	0.8*
DBIL	0.2	0.4*	0.3	0.3	0.3	0.1	0.2
ALP	52	75	82	52	40	42	65
AST	26*	36*	36*	34*	36*	31*	32*
ALT	40*	40*	25	25	38*	35*	24
GGT	38	15	41	48*	42	48*	42
TP	6.5	7.2	7.5*	6	6.7	6.1	5

*Keys: TBIL (0.2 - 1.0 mg/dL), DBIL (0.0 - 0.4 mg/dL), ALP (35 - 105 iU/L), AST (0 - 17 iU/L), ALT (0 - 40 iU/L), GGT (7 - 50 iU/L), TP (6.0 - 8.0 g/dL), ALB (3.5 - 5.0 g/dL), * (above normal)*

4.16. Discussion

The prevalence of HEV infection, level of awareness of the infection and the predisposing risk factors among pregnant women in Akure metropolis was investigated in this study. The outcome of this study revealed a seroprevalence rate of 3.52% of HEV infection among pregnant women in the study area based on IgM-HEV antibodies present in the plasma of the pregnant women. This value was higher than the prevalent rate of 0.3% based HEV- IgM antibody among pregnant women in Ekiti State (Fasakin *et al.*, 2024) and 1.9% among pregnant women in Ibadan, Nigeria (Ike *et al.*, 2024a). However, it was lower than the prevalence rate of 4.3 % reported by Ike *et al.* (2024b) among pregnant women in Abeokuta, Nigeria. It was also lower to the estimated pooled prevalence of 42.6% (Junaid *et al.*, 2014), 28.0% (Nalunkuma *et al.*, 2021) and 13.3% (Okwara *et al.*, 2021) obtained from HEV seropositivity studies conducted in pregnant women in different regions of Nigeria. These differences could be attributed to sample size, and the differences in specificity and sensitivity of HEV detection that resulted from different laboratory diagnostic techniques, including the study location and environment. The discrepancies in seropositivity could also be related to socioeconomic status, urban and rural socio-infrastructural differences, such as poor housing, poor sewage and drainage infrastructures, and unhygienic conditions of the environment (flooding and poor sanitary practices) compounded by unwholesome waste disposal practices in various homes.

The high HEV seropositivity in pregnant women within the age range greater > 45 years in this study agrees with findings of Ehi Airiohuodion *et al.* (2022) who reported that HEV seropositivity was much higher among older pregnant women who were 46-50 years old. The high seropositivity of HEV in the older pregnant women may be explained by the cumulative risk of infection over a subject's lifetime. It may also be due to reduced immunity as people as they advance in age therefore leading to reduced immune response to HEV infection.

The high HEV seroprevalence among pregnant women who were traders 4(11.11%) in this study might be due to factors such as socio-behavioural practices such as eating and drinking indiscriminately from food vendors with zero hygiene level, poor sanitation and low socioeconomic status. The observation that the highest prevalence rate of HEV was found in pregnant women during their first trimester agrees with the findings of Ajayi *et al.* (2022).

5. CONCLUSIONS

In this study, the HEV seroprevalence among pregnant women was 3.52%. The level of awareness of HEV infection was low (22%) among the recruited subjects. Washing of hands without soap and water after toilet use, not washing fruits with water,

drinking of untreated well water, use of potty to defaecate and eating of left-over foods were found to be risk factors that predisposed the pregnant women to HEV infection though not statistically supported. HEV screening programmes for pregnant women should be included in State health guidelines for maternal and childcare. Public enlightenment program on Hepatitis E Virus infection and its mode of transmission should be embarked on by the appropriate authorities to help in reducing the magnitude of the infection.

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