ORIGINAL ARTICLE

HEPATITIS E VIRUS PREVALENCE AND ASSOCIATED RISK FACTORS AMONG PREGNANT WOMEN IN MEDICAL CENTER OF SAMANDIN (OUAGADOUGOU, BURKINA FASO)

| Somda Hortense Poda Rouamba [1,2]* | Jean Bienvenue Ouoba [1] | Kuan Abdoulaye Traoré [1] | Bruno L. Ouoba [3] | Asséta Kagambéga [1] | Pierre Roques [4,5,6] | and | Nicolas Barro [1] |

[1]Université Joseph KI-ZERBO | Laboratoire de Biologie Moléculaire et d'Epidémiologie et de Surveillance des Bactéries et Virus transmis par les Aliments (LaBESTA) | Département de Biochimie-Microbiologie | Ecole Doctorale Sciences et Technologie (EDST) | 03 B.P. 7021 Ouagadougou 03 | Burkina Faso |
[2] Centre médical urbain Samandin | Burkina-Faso |
[3] Laboratoire National d'Elevage (LNE) | Burkina Faso |
[4] IDMIT Département / IBFJ | CEA, Fontenay-aux-Roses | France |
[5] Immunology of Viral Infections and Autoimmune Diseases (IMVA) | U1184, INSERM | Fontenay-aux-Roses | France |
[6] UMR1184, IMVA | Univ Paris-Sud, Saclay | France |

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ABSTRACT

Background: Hepatitis E virus (HEV) infection in pregnant women during the third trimester is commonly associated to fatal outcome risk. In Burkina Faso, previous epidemiological studies showed pregnant women was marginally exposed/protected to/from this virus (10.6 to 11.6% seroprevalence) in 2010-2014. **Objectives:** This study aim to update our knowledge of HEV risk factor in this population in 2017. **Methods**: From July to December 2017, 90 pregnant women were recruited at the Samandin Medical Center. Demographic data were retrieved and Anti-HEV IgM and IgG detection tests were performed using commercial ELISA kits (Wantai, Beijing, China). **Results:** HEV IgG antibodies were detected in 55.6% (50/90; 95% CI: [45.3-65.8]) of the pregnant women. The seroprevalence seems to increase with age from 33% (<20 years of age) to 71.4% (30–36 years) p = 0.0501. There was no significant association between anti-HEV antibody seroprevalence rate with other risk factors. **Conclusion:** In 2017, we found a significant higher HEV IgG prevalence than the one observed among pregnant women in 2012. Identification of the source of this increase thus deserves new study. More importantly, a recall of hygiene rules before and during pregnancy is urgently needed to avoid numerous at risk new HEV infection in this population as we have no idea of the current origin (food, water, HEV genotype involved) of this increase of HEV exposition.

Keywords: *Hepatitis E virus, pregnant women, co-infection, risk factors, Burkina Faso.*

1. INTRODUCTION

Hepatitis E (HE) is a liver disease caused by infection with the hepatitis E virus (HEV). According to the recent updated World Health Organization (WHO) data, every year there are over 3 million of symptomatic cases of hepatitis E and 44, 000 hepatitis E-related deaths in the world [1,2]. HE is usually by fecal-oral route acquired through consumption of contaminated food or water [3]. HEV is a non-enveloped, single-stranded positive sense RNA virus. It belongs in genus of *Orthohepevirus*. Only the specie *Orthohepevirus A* contains seven genotypes (HEV-1 to HEV-7) and a putative new genotype 8 that can infect humans and/or a wide variety of mammals [4,5].

Hepatitis E is generally characterized by an acute self-limited or asymptomatic course in the host with a rapid viral clearance [6]. However, these infections may have extra-hepatic manifestations and lead to chronic hepatitis in immunocompromised patients [7]. HEV infection can be severe in patients with pre-existing liver disease or receiving organ transplantation, and in pregnant women in developing countries. Thus, studies from various developing countries have shown a high incidence of HEV infection in pregnancy [9]. Mortality rates up to 25% have been described, which is much higher than general population [8,9]. It is more important in the third trimester and has been reported to be associated with 81% cases of fulminant hepatitis with obstetric complications, such as premature rupture of membranes and intrauterine growth restriction [10-12]. In addition, neonatal death in 56% of newborns and stillbirth were reported [13,14].

HEV Seroprevalence in pregnant women living in Burkina Faso ranged from 10.6 - 11.6% [15,16]. The pregnant women of Ouagadougou are undoubtedly exposed to this virus because of the poverty, promiscuity, unsanitary conditions and unhealthy lifestyle. In addition, the precise reasons for increased susceptibility to HEV infection during pregnancy and

associated severe disease are still not resolved. The aims of this study were to assess the exposition to Hepatitis E virus (IgG) and recent infection level (IgM) among pregnant women attending the Medical Center of Samandin (Ouagadougou, Burkina Faso), and to evaluate the risk factors associated with HEV infection.

2. MATERIALS AND METHODS

2.1 Study site

This is a cross-sectional study conducted at the Samandin medical center (Ouagadougou, Burkina Faso), during the period of July to December 2017. Blood samples (5 ml) were collected in dry tubes without anticoagulant. The sample was centrifuged at 3000g for 10 min at 25°C. The sera were collected in cryotubes and stored at -20 ± 5°C until serological analysis. Specific IgG and IgM antibody for HEV (anti-HEV) were measured by ELISA method using the Wantaï anti-HEV IgM ELISA and Wantaï anti-HEV IgG ELISA (Wantai, Beijing, China). The results were scored as positive or negative according to the standard procedures recommended by the manufacturer. The biological risk factors associated with HEV infection were determined by testing of HIV, HCV and HBV using the following reagents respectively: HIV RDT (Alere Medical Co., Ltd, Japan), HCV-TDR (STANDARD DIAGNOSTIC, INC, Korea) and HBsAg ELISA kits (AccuDia; USA).

2.2 Patients

Ninety pregnant women from 16 to 49 years, attending prenatal clinics, as part of prenatal testing were recruited in this study. After recruitment, the study protocol was explained to each woman and written informed consent was obtained. Women who agreed to participate received a pre-tested questionnaire to obtain information on demographic characteristics and risk factors, including age, gender, place of residence, education level, job, food regime (outside), water source, medical history and diet.

2.3 Statistics analysis:

Logistic regression analyses were carried out to determine which variables (age, level of education, locality, profession, dietary habits, miscarriage and source of water supply) were significantly associated with detection of HEV antibodies. Logistic regression was performed using R software version 2.13.0. P <0.05 was considered significant. The lower and upper limits of the 95% confidence interval (CI) were also calculated.

2.4 Ethical considerations

Ethical approval was obtained from national Ethic committee for the Health Research of the Ministry of Health (Deliberation N° 2014-12-138). Study participants received information about the study, after which they signed the written informed consent

3. RESULTS

Socio-demographic characteristics of study participants

Regarding the level of education, 33.3% of participants have completed primary, secondary or higher education; 66.7% were illiterate. Of ninety (90) pregnant women, 47.8% were official workers and 52.2% were household worker (Table 1). Concerning the source of drinking water, 35.6% of the pregnant women used taps water, supplied by the national office of water and sanitation (ONEA) and 64.4% used fountains water. 83.3% of study population resided in the urban area and only 16.7% lived in the peripheries of Ouagadougou (Table 1).

According to their activities, the pregnant women had a diversified diet out of home consisting of fruit, vegetables, meat and others (rice and maize cake). Fruits and vegetables are eaten according to the season.

Table 1: Socio-demographic characteristics of the women in the study.

CHARACTERISTICS	NUMBER	PERCENTAGE (%)
Age		
<20	9	10
[20-26[	31	34.4
[26-30[	17	18.9
[30-36[	28	31.1
>36	5	5.6
Level of education		
Unschooled	60	66.7
Schooled	30	33.3

Profession		
Household worker	47	52.2
Official workers	43	47.8
Place of residence (city)		
Center	75	83.3
Periphery	15	16.7
Food regime (outside)		
Fruits and vegetables	19	44.4
Pork meats	31	17.8
Others	40	3.3
Source of water supply		
Tap	32	35.6
Fountain	58	64.4
Number of pregnancies		
1 to 2	54	60
3 to 4	26	28.9
5 to 6	10	11.1
History of abortion		
No	82	90
Yes	8	10
History of miscarriage		
Yes	9	8.9
No	81	91.1

Prevalence of infectious disease markers among

In total, of the 90 pregnant women tested, 1.1% (1/90, CI 95% [0.0-3.3]) were positive for anti-HCV antibodies, 8.9% (8/90 IC95% [3.0-14.8]) were positive for HBsAg, and 2.2% (2/90, CI 95% [0.0-5.3]) were positive for anti-HIV antibodies (Table 2). The proportion of women exposed to HEV was very high, with an overall prevalence of anti-HEV IgG antibodies of 55.6% (50/90), CI 95% [45.3– 65.8]) for this study population. None of the sample was positive for IgM anti-HEV.

Table 2: Prevalence of HEV, HBV, HCV and HIV among the pregnant women in the study

	Negative			**Positive**		
	Number	Percentage	CI95	Number	Percentage	CI95%
IgG-HEV (ELISA)	40	44.4	[34.2 – 54.7]	50	55.6	[45.3– 65.8]
IgM-HEV (ELISA	0	0	-	0	0	0
HBsAg (ELISA)	82	91.1	[85.2 – 97.0]	8	8.9	[3.0 – 14.8]
HIV (RDT)	88	97.8	[94.7 – 100.0]	2	2.2	[0.0 – 5.3]
HCV (RDT)	89	98.9	[96.7 – 100.0]	1	1.1	[0.0 – 3.3]

Population stratification and risk factor analysis

The overall prevalence rate of antibodies to HEV was highest (71.4%) among pregnant women [30-36[year group (p = 0.0501), followed by (60%) in >36 years of age (p = 0.341), and (58.8%) in [26-30[years group (p = 0.223), then (45.2%) in [20-26[year group (p = 0.529) and (33.3%) in <25 years of age. Neither the geographical location (downtown or periphery) nor the water source (tap or public fountain) appear to be risk factors for HEV in this study. Others factors which did not show statistical evidence include employment status and educational level. Although, none any of these risk factors were statistical evidence of infection association, on the nine pregnant women have been victim of miscarriage, eight women were carried HEV antibodies. Moreover, on eight women being an abortion history, five were positives to the HEV antibodies. The two HIV positive women arbored IgG anti-HEV. Of eight pregnant HBsAg-carriers women, four women were HEV IgG positive.

Table 3: HEV and Associated Risk Factors

Associated Risk Factors	**Prevalence (%)**	**Odds-ratio**	**p-Value**
Age			
<20	33.3 (3/9)	ref	ref
[20-26[	45.2 (14/31)	1.7 [0.4; 8.9]	0.5296
[26-30[	58.8 (10/17)	2.86 [0.6; 17.5]	0.2232
[30-36[	71.4 (20/28)	5.0 [1.1; 28.7]	0.0501
>36	60.0 (3/5)	3.0 [0.3; 34.8]	0.3414

Level of education			
Unschooled	60 (32/60)	1.31 [0.5 – 3.2]	0.548
schooled	53.3 (18/30)	Ref	
Profession			
Household worker	53.2 (25/47)	Ref	
Official workers	58.1 (25/43)	1.2 [0.5 – 2.8]	0.637
Place of residence (city)			
Center	54,6 (41/75)	Ref	
Periphery	60.0 (9/15)	1.2 [0.4 – 4.0]	0.704
Food regime (outside)			
Pork meat	54.8 (17/31)	Ref	
Fruits and vegetables	57.9 (11/19)	1.0 [0.4 – 2.6]	0.989
None of them	55.0 (22/40)	1.1 [0.4 – 3.7]	0.833
Water source			
Tap	50.0 (16/32)	Ref	
Public fountain	58.6 (34/58)	1.4 [0.6 – 3.4]	0.431
Number of pregnancies			
1 to 2	51.9 (28/54)	Ref	
3 to 4	57.7 (15/26)	1.7 [0.5 – 3.3]	0.624
5 to 6	70.0 (7/10)	2.2 [0.5 – 10.9]	0.297
History of miscarriage			
No	51.9 (42/81)	Ref	
Yes	88.9 (8/9)	7.4 [1.3 – 100.0]	0.033
History of abortion			
Yes	62.5 (5/8)	1.4 [0.3 – 7.0]	0.679
No	54.9 (45/82)	Ref	
HBsAg ELISA			
Positive	50 (4/8)	Ref	
Negative	56.1 (46/82)	1,3 [0,3 ; 5,7]	1
HCV RDT			
Positive	0 (0/1)	Ref	
Negative	56.2 (50/89)	NA	0.444
HIV RDT			
Positive	100 (2/2)	NA	
Negative	54.5 (48/88)	Ref	0.5

Legend: « **ref** » = reference modality; **NA:** Not Available

4. DISCUSSION

This study found very high overall seroprevalence (55.6%; OR: 9.49 [5.23-17.73], p<0.007) of HEV antibody among pregnant women than in the previous study, suggesting a possible high exposure to enterically and zoonotic transmitted Hepatitis E Virus. Indeed, potential existence of new source of human contamination by direct contact with infected animals or consumption of infected meat that was not cooked properly, might explain this high prevalence of HEV antibodies among the pregnant [17]. Previous reports from sub-saharian Africa demonstrated prevalence of 10.6 - 11.6% in Burkina [15,16], 12.2% [18] in Ghana, 14.1% in Gabon [19], These prevalence's are less than that found in Khartoum (61.2%) [20] and Egypt (84.3%) [21] and 33.6% in India [22]. These differences between studies could be related to the hygiene, access to safe water sources, and sanitary sewage disposal systems in these communities [12].

Considering IgM anti-HEV antibodies, the sero-prevalence from this study was 0.0%. Absence of recent infection (IgM positive) observed, also registered by Kafando *et al*, could be explained by the fact that our investigation was performed on asymptomatic patients [16].

Prevalence increased with age and the highest prevalence has been recorded in the age group [30-36[, (71.4%; p = 0.0501). This is consistent with the age distribution of HEV infection in developing countries, where HEV infection is more prevalent among young adults [16,23].

In this study, anti-HEV reactivity among pregnant women living in rural areas appear to be higher than that those living in the center areas. This could be explained by low standards of living and lack of potable water supply [24]. These includes, the absence of continued sanitation systems, and the poor water management and hygiene. Moreover, of the

90 pregnant women, 58% used fountain water for drinking, washing vegetables and fruit, washing kitchen utensils, etc, which was emphasized as a risk factor for HEV in other studies [16,18]. The results of the present study have shown that there is no significant association between level of education and HEV seropositivity. In contrast, other studies showed that unschooled women are more prone to be infected by HEV than schooled pregnant women, which may indicate that the relationship between sanitation system and HEV prevalence might be balanced by hygiene practice [25].

HEV associated risk factors for abortion among pregnant women, was reported in a study conducted in Khartoum state [20,6]. Our study however did not find a significant association with history of abortion and miscarriage ($p>0.05$).

In addition, this study is unable to make inferences on HEV/HIV coinfection since only two of the participants was HIV positive. Other studies concluded that HEV may be a significant risk to pregnant women co-infected with HIV, which indicated a specific risk for HEV acquisition, with an increased viral load of HIV-1 [19]. Thus, High prevalence HEV was reported in HIV-infected patients compared to control population in Ghana (35.6% and 45.3%) [25,26], Cameroon (14.2%) [26], Italy (6.7%) [27], Spain (9.2%) [28] and 10.4% [29]. A wider assessment of the prevalence of HEV in HIV-positive people would be worthwhile because this coexistence may cause a high risk of morbidity and mortality for pregnant women.

A total of four pregnant women on eight women carriers HBsAg were HEV IgG positive. Kafando *et al.,* (2016) [16], indicated in their study that the risk of transition to chronicity and viral reactivation is very high for this group of vulnerable women (HEV/HBsAg) [16], because of their immune depression induced by the pregnancy.

5. CONCLUSION

Our study indicates that the prevalence of IgG anti-HEV in pregnant women in Ouagadougou was higher than in the previous study. Outcome to HEV infection in pregnant women is not the same in HEV genotype 1 and HEV genotype 3 or 4. Thus identification of the HEV genotype circulating in the human population need to be determined. Because it seems that it may induce subclinical infection, and we believe that the risk of HEV genotype 1 is unlikely.

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