The Role of Hepatitis E Virus Infection among Patients with Sporadic Acute Viral Hepatitis in Diyala Province

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Abstract:

Objective: To explore the role of hepatitis E virus (HEV) infection in sporadic cases of acute viral hepatitis in Diyala province.

Materials and Methods: 163 patients with clinically and biochemically evident acute viral hepatitis. 96 (58.9%) were males with mean age (27 ± 18) years, and 67 females (41.1%) with mean age (24.3 ± 15.6) years. They were referred from outpatient clinic of Baquba General Hospital and other district hospitals in Diyala province during the period from October 2003 to August 004. Patient's sera were submitted for the detection of anti-HEV IgM, anti-hepatitis A virus (HAV) IgM, anti-hepatitis C virus (HCV) antibody, and hepatitis B surface antigen (HBsAg), using Enzyme linked immunosorbant assays (ELISA), and determination of liver function tests using standard biochemical procedures.

Results: The Total positivity rate of seromarkers was 69.3 %, while 30.7% of sera were negative for all viral markers. The rate of anti-HEV IgM was (30.1%) which was significantly higher ($P < 0.001$) than anti-HCV antibody (11.6%), HBsAg (6.1%), but insignificantly higher than anti-HAV IgM (21.5%). The rate of coinfection of HEV with HAV was 3.1%, with HBsAg was 1.2 % and with HCV was 0.6%. The anti-HEV IgM positivity rate was significantly higher ($P< 0.05$) among the age group (10-49) years compared to other age groups. There was insignificant difference in the infectivity rate between males and females (31.3% vs 28.4%). Similarly there was insignificant effect of residence (rural, urban) on the anti-HEV IgM positivity rate (26% and 33.3%) respectively. The source of water supply has insignificant effect ($P>0.05$) on the anti-HEV IgM positivity rate. The difference in the mean of liver function tests among patients with different types of viral hepatitis was statistically insignificant.

Conclusion: Infection by HEV constitutes the highest proportion of sporadic acute viral hepatitis in Diyala province.

Key wards: HEV infection, Diyala province, Acute viral hepatitis.

Introduction:

Infections due to hepatitis A, hepatitis B, hepatitis C, and hepatitis E viruses are the major causes of viral hepatitis and are associated with significant morbidity and mortality in developing countries [1,2,3]. However, in approximately 10% of acute hepatitis, non of these types can be diagnosed that are collectively described as non-A, non-E hepatitis (NANE) [4,5]. HBV and HCV have received more attention due to their wide distribution and high prevalence and also to the severe long-term consequences of chronic infections with these agents [6,7,8,9].

HAV is a feco-orally transmitted virus that is highly endemic in many developing countries in which infection is acquired mainly during the childhood in whom the majority of infections were asymptomatic [10,11]. However, recent reports have pointout to the age-shifting at infection toward older ages [12,13]. Of note, the incidence of the clinical disease increases by age reaching more than 80% in those above 10 years of age [14,15].

Hepatitis E virus infection is an enterically transmitted acute viral hepatitis that is clinically similar to other forms of viral hepatitis except in pregnant women, in whom the illness is particularly severe with high mortality rates [16,17]. Infection by HEV is a disease of developing nations with improper sewage disposal and unsafe water supply that mainly affects young adults [18,19]. HEV is responsible for both waterborne outbreaks and

sporadic cases of acute hepatitis [20,21]. Asymptomatic and anicteric infections may occur[22].

Jiang et al. (2001) [23] reported that the rate of acute HAV, HBV, HCV, and HEV among patients with sporadic acute hepatitis were 29%, 18.1%, 3.9%, 49% respectively. Furthermore, they found that the total positivity rate was 72.4% and 5.8 % of patients were coinfected with two hepatitis viruses. In another study on children aged 5-16 years with acute viral hepatitis, the anti-HAV IgM and anti-HEV IgM were found to be 17% and 7%respectively, with significant correlation between ALT levels and anti-HEV IgM or anti-HEV IgM positivity rate [10]. The seroprevalence of anti-HAV IgM and anti-HEV IgM among patients with sporadic acute viral hepatitis was22.2% and 33% respectively, with significantly higher liver enzymes in both viral infections [24]. Among pregnant women with sporadic acute viral hepatitis, the rate of HAV, HBV, HCV, HDV coinfection, HEV were 1.5%, 15%, 1.7%, 1.5%, 49.6% respectively, and 30.7% was due to NANE [25].

Materials & Methods:

The study group includes 163 patients with clinically evident acute viral hepatitis. They were referred from outpatient clinic of Baquba General Hospital and other district hospital in Diyala province, to the Public Health Laboratory to explore seromarkers of viral hepatitis. The males were 96 (58.9%) with mean age (27 ± 18) years, and 67 females (41.1%) with mean age (24.3 ± 15.6) years.

Five to ten milliliters of venous blood were collected aseptically from each patient. Blood samples were left at room temperature to clot. Sera were separated by centrifugation at 300 RPM, divided into aliquots (250 microliter each) and stored at –20 ^{0}C till use. Patient's sera were submitted for the (1) determination of liver function tests, including serum total bilirubin (direct and indirect), as well as determination of alanine aminotrasferase (ALT) activity, using commercial biochemical kits.{2} detection of viral markers including, anti-HEV IgM, anti-HAV IgM, anti-HCV antibody, hepatitis B surface antigen (HBsAg), using Enzyme linked immunosorbant assays (ELISA).

Results:

The rate of anti-HEV IgM was 49 (30.1%) which was significantly higher ($P < 0.001$) than anti-HCV antibody (11.7%), HBsAg (6.1%), but insignificantly higher than anti-HAV IgM (21.5%), table (1).

It is important to clarify that 5 (3.1%) sera were found to be positive for both anti-HEV IgM and anti-HAV IgM. Similarly, 2 (1.2 %) sera were positive for both anti-HEV IgM and HBsAg. Furthermore, 1 (0.6%) serum sample was positive for both anti-HEV IgM and anti-HCV antibody, table (2).

The anti-HEV IgM positivity rate was significantly higher ($P< 0.05$) among the age group (10-49) years as compared to other age groups, Table (3). The results also showed that the age group (10-49) years had 19.2 times more risk to have HEV infection compared to other age groups.

The anti-HEV IgM positivity rate in males was insignificantly higher ($P> 0.05$) in males as compared to females (31.3% *vs* 28.4%), table (4).

Table (1): Rates of positive viral markers among the patients.

Viral markers	No.	Infection rate	95% CI
Anti-HEV IgM	49	30.1	23.3-37.8
Anti-HAV IgM	35	21.5	15.6-28.7
Anti-HCV antibody	19	11.6	7.3-17.8
HBsAg + anti-HBc IgM	10	6.1	3.2-11.3
Non	50	30.7	

Table (2): Rates of viral coinfections among the patients.

Co-infection	N0.	%
Anti-HEV IgM + anti-HAV IgM	5	3.1
Anti-HEV IgM + HBsAg	2	1.2
Anti-HEV IgM + anti-HCV antibody	1	0.6

Table (3): Anti-HEV IgM positivity rate by age groups.

Age (years)	No.	Anti-HEV IgM positive		Odd ratio	95% CI
		No.	%		
< 5	18	1	5.6	Reference	
5-9	15	2	13.3	2.6	0.2-32.1
10-19	27	10	37	10	1.2-68.9
20-29	38	13	34.2	8.4	1.1-74.9
30-39	35	12	34.3	8.9	1.1-74.9
40-49	15	8	53.3	19.4	2-185.7
50 +	15	3	20	4.3	0.4-46

Table (4): Anti-HEV IgM positivity rate by gender.

Sex	No.	Anti-HEV IgM positive		Odd ratio	95% CI
		No.	%		
Male	96	30	31.3	1.1	0.6-2.2
Female	67	19	28.4	reference	

Similarly, there was insignificant (P> 0.05) effect of residence (rural, urban) on the anti-HEV IgM positivity rate (26% and 33.3%) respectively, table (5).The results also showed that the source of water supply has insignificant effect (P>0.05) on the anti-HEV IgM positivity rate, table (6).

Regarding the biochemical liver function test, the mean of total serum bilirubin was insignificantly higher (P= 0.46) in patients with HEV and HAV coinfection (10.1 unit/100 ml) as compared to patients with other viral markers. Similarly, although there was slight increase in the mean of direct serum bilirubin in patients with HEV infection (6.6 mg/ 100 ml) compared to other patient groups; however, this increment was statistically insignificant (P=0.48). The mean of indirect serum bilirubin were also showed insignificant difference (P=0.36) among the patient groups, although it is relatively higher among patients co-infected by HEV and HAV. The mean of serum alanine aminotransferase activity was insignificantly higher (P=0.08) among patients with HEV and HAV coinfection compared to patients with other viral markers, table (7).

Table (5): Anti-HEV IgM positivity rate by residence.

Residence	No.	Anti-HEV IgM positive		Odd ratio	95% CI
		No.	%		
Rural	73	19	26	Reference	
Urban	90	30	33.3	1.4	0.7-2.8

Table (6): Anti-HEV IgM positivity rate by source of water supply.

Water supply	No.	Anti-HEV IgM positive		Odd ratio	95% CI
		No.	%		
River/Well	5	2	40	1.6	0.7-2.8
Municipality	158	47	29.7	Reference	

Table (7): Biochemical liver function tests among patient groups.

Positive Viral Markers	Mean ± SD			
	TSB (mg/dl)	Direct SB (mg/dl)	Indirect SB (mg/dl)	ALT (U/ml)
Non	5.8 ±2.45	3.2 ± 1.4	2.6± 2.71	67.3 ±38.5
Anti-HEV IgM	8.6 ± 6.92	6.6± 6.15	2 ± 1.81	87± 56.3
Anti- HAV IgM	6.1 ±1.6	4.4 ± 1.6	1.8 ± 0.45	115.3± 63.2
HBsAg	5.8± 2.9	4.6± 2.58	1.3± 0.87	124.1± 35.2
Anti-HEV IgM + Anti-HAV IgM	10.1± 7.2	6.4 ± 3.2	3.7 ± 4.0	165.3± 74.6
Anti-HEV + HBsAg	4.9± 0.48	3.6 ± 0.25	1.3 ± 0.74	43.7± 59.8
P value	**0.46**	**0.48**	**0.36**	**0.08**

Discussion:

The 49 patients appeared to be infected with HEV in the present study were include 41 patients with HEV infection alone plus 8 patients who had coinfection between HEV and other hepatitis viruses. The significantly higher infection rate of HEV is consistent with many previous reports conducted in endemic areas [23,24]. These results may affirm the notion that subclinical or inapparent infection in humans are do exist [22]. Those carriers may act as reservoirs for shedding the virus with their feces and consequently contaminate the water supply, which is believed to be the principle source of infection by HEV [18,26]. However, the insignificant difference between HEV and HAV infection is not unusual in a community like ours, since the ancient feco-orally transmitted HAV has been well documented to be prevalent in high rates in such communities [10].

Another result that draws attention is the considerable rate of negativity for all viral markers utilized in this study. These results may be either due to the low titer of viral markers that is beyond the sensitivity of the assays employed, or probably attributed to another hepatitis viruses (Non, A–Non, E) not included in this study. These results were consistent with previous reports [25,32].

Simultaneous infection by hepatitis E virus and other hepatotropic viruses was reported in the literature [23]. Therefore, the rate of coinfections recorded in the present study was not so remarkable.

The tendency of young adults for HEV infection (Odd ratio 19.2) was concordant with almost all studies carried out in this field, and actually this age specificity of HEV infection constitute one of the remarkable feature of this disease [21, 27]. The most reasonable explanation is that, because HEV is not ubiquitous in nature, as the excretion of virus in stools during acute infection is low and is often in a degraded form [27], beside that the virus is extremely labile to the unfavorable environmental conditions [19]. So, when infections occur in childhood are mostly asymptomatic or mild [28]. Consequently, the provoked immunity is short lived, and therefore reinfection is likely to occur during later life [16].

The results revealed an insignificantly higher infectivity rate in males compared to females. However, controversial reports regarding the infectivity rate between sexes have been found in the literature [21,29,30].

The anti-HEV IgM was insignificantly higher among urbans as compared to rurals. On the contrary, municipality as a source of water supply appeared to have lower risk over that of rivers/wells. A lot of controversial results have been surrounding this subject [28, 31,32]. Of note, outbreaks of HEV infection in rural areas have been reported that are caused by major contamination of water supply [22,33]. It seems that contamination of drinking water by the virus weather it is occur in rural or urban areas is the principal cause of HEV infection.

The insignificant difference in the mean of liver function tests in patients with different types of viral hepatitis. These results clearly support the notion that clinical picture and biochemical investigations are insufficient to differentiate among hepatitis caused by different hepatotropic viruses [2,3].

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