Evalution of anti-HAV antibodies (IgM) among children aged 1-5 years in Diyala governorate

Ansam Dawod Salman ' Zainab Amer' Shyamaa Mohammed

Diyala University, College of science
Lecturer in Biology Department/ College of Sciences/Diyala University
Assistant Lecturer in Biotechnology Department/ College of Sciences/Diyala University
Biology Department/ College of Sciences/Diyala University
Corresponding Auther: Ansam Dawod Salman

Abstract: With improvement in economic and living conditions of the communities, the age of acquiring hepatitis A virus (HAV) infection is shifting from early childhood to adolescence and young adulthood. Such epidemiological shift leads to an increased incidence of symptomatic HAV infection, including heightened risk of liver failure. The present study was conducted in Diayala province for the period from 15/10/2017 to 10/4/2018. It included; 80 children patients were attended Al-Batool Hospital. 19 of patients were males and 61 were females. The age range was 1 years to 5 years. The study aimed to determining the rate of HAV-specific IgM antibodies in children aged from 1 to 5 years. Serum specimens were collected for each children. Detection of HAV-specific IgM was done by Enzyme Rapid test. The results of the present study showed that the 9 children (11.25%) were anti-HAV IgM positive and 71 (88.75%) were anti-HAV IgM negative. There was higher seropositivity of HAV between age group (1-2) years (23.07%) for anti-HAV IgM with other age groups. In this study regarding gender, the rapid test positivity rate was in females (4.91%) was lower than that in males (31.57%). In conclusion, am recommended that working similar studies need to be done in other parts of the Diayla governorate. Additionally, the present study recommended that Hepatitis A virus vaccination should be considered carefully in Iraq and other developing countries, because of the high seropositivity to HAV in our region.

Key world: hepatitis A virus (HAV), IgM.

Date of Submission:28-06-2018 Date of acceptance: 12-07-2018

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I. Introduction:

HAV is a single-stranded RNA virus. Infectivity occurs primarily through fecal-oral transmission and, after ingestion and absorption, the virus replicates in the liver and is excreted in bile. HAV enters into the hepatocyte by specific receptors located on the plasma membrane. Viral RNA is encoated after uptake and binds to ribosomes, stimulating the synthesis of viral proteins, and replication of the viral genome occurs by RNA polymerase. The virus is then secreted into the biliary tree and excreted in feces, where high concentrations of HAV are detected [land2]. An immunologic response occurs within the liver, leading to portal and periportal lymphocytic infiltration, and potentiates liver damage. A limited number of cases have been reported from blood transfusions, and vertical transmission from mother to fetus.

Transmission of HAV is the highest during the anicteric prodrome of 14-21 days, when fecal and serum virus concentrations are high. The incubation period is typically 2-6 weeks, with an average of 28 days. Fecal viral excretion may persist for up to 3 weeks. Immunoglobulins to HAV (IgM anti-HAVantibodies) can first be detected in serum 5-10 days after exposure $^{(3\text{and }4)}$.

HAV is spread from person to person through contact with the feces (stool) of people who are infected, which can easily happen if someone does not wash his or her hands properly. You can also get hepatitis A from food, water, or objects contaminated with HAV $^{(5)}$.

Areas of high endemicity include most of Africa, Asia, Central and South America. Conditions which contribute to the propagation of the virus among young children in these areas include household crowding, poor levels of sanitation and inadequate water supplies. Iraq considered as highly endemic area⁽⁶⁾. In communities that have intermediate rates of hepatitis A the disease occurs among children, adolescents and young adults, in contrast the communities that have high rates of hepatitis A, in which the majority of cases occur among children less than 15 years of age. With improvement in socio-economic conditions and its consequences, early childhood exposure to the virus has decreased ⁽⁷⁾.

Hepatitis A vaccine is an inactivated (killed) vaccine. You will need **2 doses** for long-lasting protection. These doses should be given at least 6 months apart.

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Children are routinely vaccinated between their first and second birthdays (12 through 23 months of age). Older children and adolescents can get the vaccine after 23 months. Adults who have not been vaccinated previously and want to be protected against hepatitis A can also get the vaccine ⁽⁸⁾.

Aim of the study:

The present study was conducted to achieve the following aims.

- 1. The diagnostic of HAV in serum specimens of human by Rapid test.
- 2. The effects of certain demographic factors on the detection rate by these laboratory technique.

II. Materials and Methods:

Laboratory diagnostic kit:

The following laboratory kit was employed throughout the study as showing in table (3.2).

Table (3.2): Laboratory diagnostic kit used in the study

Item	Company and country of origin
Hepatitis A virus IgM Rapid test device (serum)	Anon Biopharm(China)

3.2. Methods:

3.2.1. Samples collection:

The present study was conducted in Diayala province for the period from 15 / 10 / 2017 to 10 / 4 / 2018. It included; 80 children patients were attended Al-Batool Hospital. 19 of patients were males and 61 were females. The age range was 1 years to 5 years.

Collection of serum specimens

From each individual in this study, 5 ml of blood was drawn by vein puncture using disposable syringes. The blood was placed in plastic disposable tubes; it was left to stand at room temperature (20 25°C) to allow it to clot, then the sera was separated by centrifugation 10000 rpm for 5 minutes and stored at -20°C till examination. The specimens were transferred to the Microbiology Laboratory in Biology Department/Collage of Sciences/Diyala University for detection of Anti-Hepatitis (IgM) in serum specimens by rapid test. All sera and reagents were allowed to stand at room temperature before use in the test.

3.2.4. Detection of Hepatitis A virus IgM:

3.2.4.1. Hepatitis A virus IgM Rapid test device (serum):

This test was performed using commercially available kit (Anon Biopharm(China)).

Interpretation of results:

Positive: Two lines appear. One colored line should be in the control line region (C) and another apparent colored line should be in the test line region (T).

Negative: One colored line appears in the control line region (C). No line appears in the test line region (T). **Results:**

Using the rapid test for detection of Hepatitis A- specific IgM antibodies in sera children specimens, the age was 1-5 years. Additionally, none of the subjects remembered to have a history of vaccination, the results showed that 9 (11.25%) of specimens were positive for IgM antibodies and only 71 (88.75%) were negative for IgM antibodies. table (1).

Table (1): Results of Rapid test for detection of IgM antibodies to Hepatitis A Virus in children

	Results	Frequency (%) of IgM for HAV
Children	Positive	9 (11.25%)
n= 80	Negative	71 (88.75%)
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Results in table (2) revealed that the detection rate of Hepatitis A by Rapid IgM tests was highest in the age group (1-2) years for IgM as compared to other age groups.

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Result of IgM (HAV) Total groups (years) No. positive No. negative (%) (%) 20 (76.92) 1-2 6 (23.07) 26 2-3 3 (14.28) 18 (85.71) 21 3-4 0 (0.0) 19 (100) 19 4-5 0 (0.0) 14 (100.0) 14 9 Total 71 80

Table (2): Hepatitis A- specific IgM antibodies among the different age groups of children specimens

Regarding gender, the Rapid test positivity rate was in females (4.91%) was lower than that in males (31.57%), table (3)

Table (3): Distribution of Rapid test results according to sex
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Sex	Result of DAT		
	No. positive (%)	No. negative (%)	Total
Male	6 (31.57)	13 (68.42)	19
female	3 (4.91)	58 (95.08)	61
Total	9	71	80

III. Discussion:

HAV infection is one of health problems in Iraq encountering 41% of suspected cases acute viral hepatitis in general population during $2005^{(19)}$.

The finding in this study of 9 cases (11.25% of suspected cases) who were positive for IgM HAV testing. Since, worldwide studies had reported variable results, so the present results were disagree with the results of the study reported by Tareef, F. Raham and Assad, M. Abood ⁽¹⁵⁾. However, other workers (Muslim Dhaher,

Munther Kameel and Hekmat Kadhum) reported an infection rate 36.5% (20). Nevertheless, worldwide studies have yielded variable HAV infection rates among children less than 5 years old; for instance, in Irland (Connor et al., 2018)⁵ reported an infection rate of 50%, while in Glasgow (Li et al.,2015)²¹ reported an infection rate of 41.3%. These controversies in the results of the different studies maybe attributed to many reasons, the difference in geographical area, seasonal variation, sample size of the study, and the concentration of RV antigen in the collected specimens.

The results of present study revealed infection rate by HAV among age group 1-2 years (23.07%) detected by rapid test. These results were in agreement with those reported by other Iraqi researchers (Tareef F. Raham and Assad M. Abood, 2015; Muslim Dhaher, er al. 2015) $^{(11 \text{ and } 20)}$. Additionally, worldwide studies reported results agreement with present results in the infection rate by HAV increased among children less than 5 years of age and that children < 2 years were mostly infected $^{(1, 10, 14 \text{ and } 15)}$.

Male predilection in this study is (31.57%) and this finding is in agreement with many studies and reports from developing and developed countries. In multicenter study conducted in five different centers in India; males show predominance ⁽¹⁰⁾. In USA From 1996 through 2002, rates of acute, symptomatic hepatitis A have been higher among males than females, however, since 2006, overall rates have declined more among males than among females. In 2008, incidence among males was 0.9 cases per 100,000 populations, compared with 0.8 cases per 100,000 populations among females ⁽²⁴⁾. In Canada, the rates are higher for males than for females according to study on reported cases of hepatitis A from 1990 to 1999⁽²⁵⁾. In a previous study in Iraq and in Saudia Arabia the prevalence was almost the same in male and female^(22 and 23). According to this study and other studies and reports ⁽⁹⁾; incidence of acute, symptomatic hepatitis A varies by age.

IV. Conclusions:

In conclusion, the study shows low incidence of HAV among children with, this finding indicates the need for periodic screenings of prevalence of HA virus IgM antibody seroposetivity among all age groups to detect transition to older age groups. The need for implementing preventive programs including HAV vaccine is highly indicated if transition occurs.

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Recommendations:

The present study recommends that Hepatitis A virus vaccination should be considered carefully in Iraq and other developing countries, because of the high seropositivity to HAV in our region. Although, working similar studies need to be done in other parts of the Diayla governorate.

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Ansam Dawod Salman "Evalution of anti-HAV antibodies (IgM) among children aged 1-5 years in Diyala governorate ""IOSR Journal of Pharmacy and Biological Sciences (IOSR-JPBS) 13.4 (2018): 09-12