International Archives of Medical Research 16:1 (2024) 11-17

ORIGINAL RESEARCH

Six-month Follow-up of Hepatitis A Antibodies in Newborn Infants in Diyarbakir Province and Its Region

Received Date: 23.03.2024, Accepted Date: 16.04.2024 **DOI: 10.56484/iamr.1457184**

Hakan ONUR[1a*], Arzu RAHMANALİ ONUR[2b]

[1] Memorial Private Diyarbakir Hospital, Department of Pediatrics, Diyarbakir, Türkiye
[2] University of Health Scienses, Diyarbakir Gazi Yaşargil Health Research Center, Deparment of Clinical Microbiology, Diyarbakir, Türkiye
[a]*ORCID: 0000-0003-4972-3878*, [b]*ORCID: 0000-0002-7669-9570*

Abstract

Objective: *In this study, we aimed to determine the frequency and persistence of maternally transmitted anti-HAV antibodies in healthy newborn infants of people living in Diyarbakır province, which is located in a high HAV endemicity area, and its neighbourhood.*

Material and Method: *In our study, infants born between October 1999 and January 2000 in the Gynaecology and Obstetrics Clinic of Diyarbakır Dicle University and registered in the Department of Paediatrics were followed up for 12 months. Blood samples were collected from all babies at birth, third and sixth months.*

Results: *A total of 112 healthy newborns were included in the study. Of the infants, 62 (55.3%) were boys and 50 (44.7%) were girls. Demographic and medical information was obtained from the parents. Seropositivity was 90.2% at birth, 87.5% at 1 month, 39.3% at 6 months and 28.6% at one year.*

Conclusion: *It was emphasised that our region has a high endemicity for HAV and in order to reduce HAV infection, socioeconomic level should be increased, hygiene should be given importance and immunisation with vaccine should be performed when appropriate conditions are met.*

Keywords: *Hepatitis A, newborn, hepatitis A vaccine, Diyarbakır*

Corresponding author: E-mail: drhonur@hotmail.com

Introduction

Hepatitis A virus (HAV) infection is among the most common viral infections in the world. It is still endemic in some regions of developing countries such as our country but can sometimes cause increases. Hepatitis A infection without symptoms, especially in childhood, causes some difficulties in diagnosis. The seroprevalence of hepatitis A infection can be affected by various factors such as geographical location, sociodemographic characteristics, and age distribution[1,2]. Due to improvements in hygiene with increasing socioeconomic conditions, the incidence of HAV infections continues to decrease in many developing countries, including Turkey[1,3].

In terms of passive immunity, transplacental transfer of maternal IgG antibodies may protect against infectious diseases during infancy. However, the presence of maternal antibodies at the time of vaccination may reduce the vaccine-induced immune response[4-6]. The duration of passive immunity provided by maternal antibodies varies among populations. Therefore, seroepidemiological studies provide important data regarding the decrease in maternal antibodies in a specific population.

Seroepidemiological studies have shown that maternally derived anti-HAV antibody titers remain high during the first six months of life but decline significantly after 12 months. With data indicating that only 6.1 percent of 18-month-old babies in Turkey have maternal antibodies, two-thirds of babies older than 12 months have a high risk of contracting HAV infection.

In our study, we aim to evaluate the persistence of naturally acquired maternal antibodies against HAV in a group of newborns aged between 0 and 6 months.

Materials and Methods

In our study, babies born in Diyarbakır Dicle University Gynecology and Obstetrics Clinic and registered in the Department of Child Health and Diseases were followed up for 12 months. Verbal and written consent was obtained from the families for the study. Blood samples were taken from all babies at birth and the third, 6th, and 12th months.

Infants born prematurely, experiencing a complicated delivery, or whose mothers had acute hepatitis, along with those unable to attend scheduled follow-ups, were excluded from the study cohort. Additionally, infants whose parents declined participation were also excluded.

ELISA

Blood samples for the detection of anti-HAV antibodies were taken at birth, at the first month, and at the sixth month. After cleaning the babies' forearms with antiseptic solutions, blood samples were taken from superficial veins in sterile straight tubes in 3 ml quantities. After the

serums were separated, they were brought to the ELISA unit of the Central Laboratory of Dicle University Faculty of Medicine Research Hospital on the same day.

IgM and IgG antibodies against the Hepatitis A virus were evaluated using the enzyme immunoassay method. For this purpose, EQUIPAR Diagnostic HA IgM, HAV Ab III. Belt kits (Equipar srl, Via Power Ferrari Sarenno (va) Italy) were used. The test demonstrated high levels of sensitivity and specificity.. The fresh serum samples taken were stored at 2-8 oC and studied with the Tecan Minilyser ELISA device in the ELISA unit on the same day. All processes were performed at room temperature (20-250C). We conducted all analyses following the manufacturer's instructions, utilizing commercial kits and systems.Sample and threshold values for hepatitis A were determined using the average adsorbance value of the calibrator. Individuals whose diagnostic findings for Hepatitis A surpassed a value of >1.1 S/CO were deemed positive. Our study focused on qualitative analysis of antibodies to HAV, without conducting additional quantitative evaluation.

Statistical analysis

Descriptive and comparative statistical assessments were carried out using the Social Sciences Statistical Package for Windows (SPSS). Nominal variables were delineated by quantity and percentage values.. Quantitative variables are presented in the tables as mean ± std. (standard deviation) and categorical variables are given as n (%). In comparing four dependent repeated measurements with each other, The Cochran's Q test was evaluated based on Monte Carlo simulation outcomes, and the Dun's Test was employed for the Post hoc analysis. Variables were assessed with a 95% confidence interval, and significance was determined for p-values below 0.05.

Results

The study comprised 112 healthy newborns with a mean gestational age of 38.7 ± 0.6 weeks. Of these, 62 (55.2%) were boys and 50 (44.7%) were girls. 38.2% of the fathers and 16.8% of the mothers of the babies included in the study were high school or university graduates. 20.2% of the babies were their mothers' first child. 41.2% of the babies lived in the city center (76.8% in Diyarbakır city center, 23.2% in the surrounding province centers), 58.8% in rural areas (71.2% in the rural areas of Diyarbakır province, 28.8% in the rural areas of the surrounding provinces) (Table 1).

Table 1. *Demographic characteristics of babies.*

	mean ± SD.
Gestational age (weeks)	38.7±0.6
	n (%)
Gender	
Male	62 (55.2)
Female	50 (44.7)
Mother's Educational Status	
Illiterate	43 (38.4)
Elementary School	40 (35.7)
High School	13 (11,6)
University and above	6 (5.3)
Father's Educational Status	
Illiterate	32 (28.6)
Elementary School	37 (33.0)
High School	32 (28.6)
University and above	11 (9.8)
Diyarbakır Province	82 (73.2)
City Center	35 (42.7)
Countryside	47 (57.3)
Surrounding Cities	30 (26.8)
City Center	7 (23.3)
Countryside	23 (76.7)

SD.: Standard Deviation

112 selected for the study successfully completed it.Anti-HAV antibodies persistence is set out in table 2.

Anti-HAV IgG positivity in newborns tested at birth was detected in 90.2% (n=101). The acceleration in the decrease in this positivity until the sixth month became more evident in the first year. Anti-HAV-IgG (+) was detected in 44 (39.3%) babies in the sixth month and in 32 (28.6%) babies in the 12th month (Table 2).

Table 1. *Changes in HAV serology in infants*

Time (n)	**Anti-HAV IgG (+) n (%)**	**Anti-HAV IgG (-) n (%)**	**p**
0. Day (n=112)	101 (90.2)	11 (9.8)	
1. Month (n=112)	98 (87.5)	14 (12.5)	**<0.001**
6. Month (n=112)	44 (39.3)	68 (60.7)	
12. Month (n=112)	32 (28.6)	80 (71.4)	

Cochran's Q test (Monte Carlo), Post Hoc Test: Dun's Test

Anti-HAV IgM antibodies (7.5%) were detected as positive in a male baby who was anti-HAV IgG negative in the neonatal period and two female babies who were anti-HAV IgG positive

until the sixth month but became negative in the sixth month, and in the laboratory examinations, the babies' ALT and AST values were found to be high. No clinical icterus was observed in these babies.

Based on these results, it was concluded that the babies had subclinical HAV infection. The seropositivity rate in the sixth month was significantly lower than those detected in previous months ($p<0.001$) (Table 2) (Figure 1). This value did not become seropositive in any of the anti-HAV IgG seronegative babies during the follow-up period.

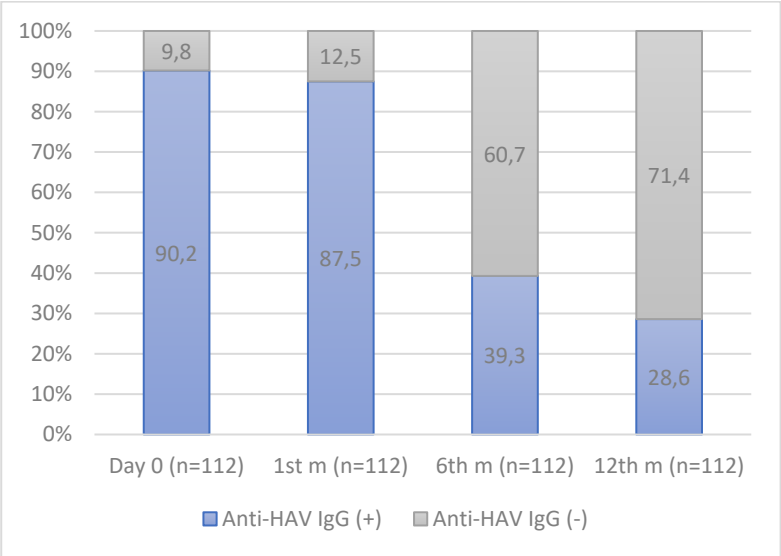

Figure 1. *Changes in anti-HAV-IgG positivity.*

Discussion

Newborns in developing countries are at risk for hepatitis. HAV infection has a high incidence, especially in regions where sanitation conditions are inadequate, and there are no appropriate vaccination programs. Since HAV infection in children is generally asymptomatic, regular seroprevalence studies can provide more clear information to ascertain disease occurrence .

In recent years, there has been no homogeneous distribution among seroepidemiological data following rapidly changing sanitation conditions, awareness about infection, and widespread vaccination programs.

In the context of HAV infections, the occurrence of maternal antibodies and the duration of their decline are pivotal in determining the optimal timing for vaccination across diverse populations[4,7].

The hepatitis A vaccine effectively interrupts virus transmission, resulting in a notable decrease in hepatitis A infection rates within the general populace. While immunizing infants represents an ideal preventive measure, in regions with moderate to high endemicity, a significant

proportion of women of childbearing age possess anti-HAV antibodies, which are passed on to their offspring during pregnancy. For instance, research by Alabaz et al.[8] in Turkey revealed that 138 out of 147 newborns and their mothers were hepatitis A seropositive. Similarly, Kanra et al.[9] demonstrated HAV seroprevalence rates of 70.2% in infants under one year old and 73.2% in women aged 30 or younger. These studies underscored the high incidence of maternal antibody transmission[10,11]. High HAV seropositivity was expected because the patients we included in our study consisted of people living in Diyarbakır city center and rural areas, where sanitation conditions and access to primary health care were difficult at the time of the studyPrior research indicates that maternal antibodies targeting HAV may endure in infants for as long as one year, and may even persist until the second year in moderately endemic regions such as Turkey[10,12]. However, although HAV IgG (+) was previously detected in two babies during the study period, it was observed that their antibodies became negative after the sixth month, and HAV-IgM (+) was detected. This result may be evidence that the protection of maternal antibodies is short-lived, especially in children living in high-endemicity regions.

Alabaz et al. observed HAV seroprevalence rates of 93.9%, 90.5%, 84.4%, 62.6%, 36.1%, and 13.6% at 3, 6, 9, 12, 15, and 18 months, respectively. Meanwhile, Lieberman et al. documented HAV seropositivity rates in 4, 6, and 12-month-old infants as 100%, 95%, and 39%, respectively. In our study, it was found that antibodies became negative at a higher rate in the first six months of postnatal follow-up compared to other studies. This difference in HAV seropositivity rates can be explained by socioeconomic status, sanitation conditions, geographical location, and changes in HAV epidemiology[13]. While more than 90% of HAV infections are asymptomatic in childhood, up to 70-80% of them are symptomatic in adults[1,2]. We assumed that most mothers were exposed to HAV pre-pregnancy and that during pregnancy, antibodies had been transmitted to the fetüs.

It was found that in a population not vaccinated and with no previous history of HAV infection , HAV IgG seropositivity rates decreased to 39.3% in 6-month-old children and 28.6% in the first year. In particular, in our research, the effect of sociodemographic characteristics on rates of seropositivity could not be adequately evaluated due to the similar middle/high socioeconomic status between the study groups. Furthermore, there is a lack of documentation regarding the impact of maternal antibodies on cell-mediated immune responses to HAV. In this investigation, the discovery of 90.2% seropositivity for HAV IgG in newborns suggests that mothers may have been exposed to HAV infection and passed on their antibodies to their offspring. Therefore, the decreasing trend in these seropositivity rates in children during the 6-month follow-up most likely reflects the existence of anti-HAV antibodies acquired from the mother, and the low protection rates

indicate that the vaccination program after the sixth month is accurate.

Maternal antibodies have been shown to inhibit the effectiveness of all inactivated vaccines, live attenuated vaccines, and most DNA vaccines[7,14]. Hence, the timing of primary vaccination is influenced by various factors related to the infant's developing immune system, including the necessity to provide protection before potential exposure to diseases, the potential interference of passively acquired maternal antibodies with vaccine-induced immunity, and other considerations (4,7). By the age of 12 months, passively transferred antibodies from the mother can be detected in only a small minority of infants, and it decreases more dramatically from 6 months onwards[15].

Although the hepatitis vaccine was not yet administered between 2000 and 2012, a significant decrease in seropositivity was observed. This decrease is probably due to improvements in infrastructure and sanitation conditions in Turkey[16,17]. In the subsequent period, vaccination also played a role in this decline. Research undertaken in Turkey before widespread vaccination recommendations reported varying rates due to socioeconomic differences between regions. With the acceptance of widespread vaccination after 2012, it is observed that the differences between regions in Turkey have decreased significantly[18,19]. However, vaccination at an early age has an important place in reducing HAV infection, which causes high morbidity and mortality in adulthood.

The decline in maternal antibodies may create a vulnerable period during which the infant becomes susceptible to relevant infections[4,7,20]. According to Brinkhof et al., the half-life of antibodies is estimated at 40 days, with 95% of infants projected to lose their passive immunity by around 13.2 months[21,22]. Similarly, Alabaz et al. found that roughly two-thirds of infants are susceptible to HAV infection at approximately 12 months of age[8]. Consistent with these findings, our study indicates that one out of every two children becomes susceptible to HAV infection starting from the 6th month. . We think that babies become susceptible to HAV in their first year in terms of decreased antibodies in the mother.

Our study has some inevitable shortcomings. Given that our reserach was undertaken in a medium-high endemicity region for HAV is the most important reason for high HAV-IgG positivity in mothers and therefore in babies. National vaccination programs for HAV in Turkey were started after our study and after 2012. Hence, the data gathered in our study depict the pre-vaccination era among a cohort of unvaccinated children, thus aiding in the identification of the ideal vaccination schedule. The fact that the HAV vaccine was not part of the widespread vaccination program at the time of the study caused the HAV seroprevalence in adults and newborns to have significant differences from current results.

In conclusion, although the transfer of maternal anti-HAV IgG antibodies in the placenta seems protective in the first six months of the baby, it has been determined that one in every two babies is vulnerable to HAV in the following period. Furthermore, given the rapid decline in HAV occurrence among adults as against previous years, the transmission of maternal antibodies is expected to decrease significantly. For this reason, we think that it is appropriate to start widespread vaccination programs for HAV in early infancy.

Acknowledgment

Some portions of this article constitute Dr. Hakan Onur's Graduation thesis for the requirement of the degree in Specialization in Pediatrics at Dicle University, Medical Faculty, Diyarbakir, Türkiye

References

1. **FitzSimons D, Hendrickx G, Vorsters A, Van Damme P.** Hepatitis A and E: Update on prevention and epidemiology. Vaccine. 2010 Jan;28(3):583–8.
2. **Jacobsen KH, Koopman JS.** Declining hepatitis A seroprevalence: a global review and analysis. Epidemiol Infect. 2004 Dec 16;132(6):1005–22.
3. **Ceyhan M, Yildirim I, Kurt N, Uysal G, Dikici B, Ecevit C, et al**. Differences in hepatitis A seroprevalence among geographical regions in Turkey: a need for regional vaccination recommendations. J Viral Hepat. 2008 Oct 27;15(s2):69–72.
4. **Gans HA, Maldonado YA.** Loss of Passively Acquired Maternal Antibodies in Highly Vaccinated Populations: An Emerging Need to Define the Ontogeny of Infant Immune Responses. J Infect Dis. 2013 Jul 1;208(1):1–3.
5. **Palfi M, Selbing A.** Placental Transport of Maternal Immunoglobulin G. Am J Reprod Immunol. 1998 Jan;39(1):24–6.
6. **Saji F.** Dynamics of immunoglobulins at the feto-maternal interface. Rev Reprod. 1999 May 1;4(2):81–9.
7. **Siegrist CA.** The Challenges of Vaccine Responses in Early Life: Selected Examples. J Comp Pathol. 2007 Jul;137:S4–9.
8. **Derya A, Necmi A, Emre A, Akgün Y.** Decline of maternal hepatitis a antibodies during the first 2 years of life in infants born in Turkey. Am J Trop Med Hyg. 2005 Aug;73(2):457–9.
9. **Kanra G, Tezcan S, Badur S,** Turkish National Study Team. Hepatitis A seroprevalence in a random sample of the Turkish population by simultaneous EPI cluster and comparison with surveys in Turkey. Turk J Pediatr. 2002;44(3):204–10.
10. **Vidor E**. Vaccination of Newborns against Hepatitis A in the Presence of Maternally Derived Antibodies. J Comp Pathol. 2007 Jul;137:S42–5.
11. **Lieberman JM, Chang S ju, Partridge S, Hollister JC, Kaplan KM, Jensen EH, et al.** Kinetics of maternal hepatitis and antibody decay in infants: implications for vaccine use. Pediatr Infect Dis J. 2002 Apr;21(4):347–8.
12. **De Silvestri A, Avanzini MA, Terulla V, Zucca S, Polatti F, Belloni C.** Decline of maternal

hepatitis A virus antibody levels in infants. Acta Paediatr. 2002;91(8):882–4.

13. **Wasley A.** Hepatitis A in the Era of Vaccination. Epidemiol Rev. 2006 Jun 1;28(1):101–11.
14. **Glezen W**. Effect of maternal antibodies on the infant immune response. Vaccine. 2003 Jul 28;21(24):3389–92.
15. **Bell BP, Negus S, Fiore AE, Plotnik J, Dhotre KB, Williams J, et al.** Immunogenicity of an Inactivated Hepatitis A Vaccine in Infants and Young Children. Pediatr Infect Dis J. 2007 Feb;26(2):116–22.
16. **Cortes-Martins H, Matos R, Moura S, Almeida L, Ferreira S, Manita C, et al.** Anti-HAV IgG seroprevalence in Lisbon region residents: Preliminary results from the National Serological Survey 2015–2016. J Clin Virol. 2016 Sep;82:S79–80.
17. **Arvas G, Kaya B, Berktaş M.** The Seroprevalance of Acute Hepatitis A in 0-18 Age Group Children who Applied to Igdır State Hospital. J Pediatr Infect. 2011 Dec 15;5(4):129–31.
18. **Yilmaz A.** Hepatitis A seroprevalence in Erzurum, Turkey. Ann Agric Environ Med. 2020 Sep 11;27(3):481–4.
19. **Alhan E, Kozanoglu B, Tumgor G, Celik U, Yaman A, Bozdemir N.** Epidemiological shift of hepatitis A in central Adana, Turkey. Turkish J Gastroenterol. 2015 Apr 22;25(1):6–8.
20. **Kovarik J, Siegrist C.** Optimization of vaccine responses in early life: The role of delivery systems and immunomodulators. Immunol Cell Biol. 1998 May;76(3):222–36.
21. **Brinkhof MWG, Mayorga O, Bock J, Heininger U, Herzog C.** Kinetics of maternally acquired anti-hepatitis A antibodies: Prediction of waning based on maternal or cord blood antibody levels. Vaccine. 2013 Mar;31(11):1490–5.
22. **Kalyoncu D, Urganci N.** Response to hepatitis A and B vaccination in patients with chronic hepatitis C: 8-year follow-up. Paediatr Int Child Health. 2012 Aug 12;32(3):136–9.