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Prevalence of Human Parvovirus B19 Infection among Apparently Healthy Pregnant Women Attending Selected Antenatal Care Units, Khartoum State, Sudan

Wafaa Mohammed Abdalla[1], Marwa Hussein Abdalgabar[2], Hind Haidar Ahmed[1]*, Samar Mohammed Saeed[1], Ahmed Bakheeet Abd Alla[3] and Tagwa Salah Ahmed[3,4]

[1]Department of Microbiology, College of Medical Laboratory Science, Sudan University of Science and Technology, Khartoum – Sudan.
[2]Department of Microbiology, College of Medical laboratory Science, Omdurman Ahlia University, Khartoum- Sudan.
[3]Department of Parasitology and Medical Entomology, College of Medical Laboratory Science, Sudan University of Science and Technology, Khartoum – Sudan.
[4]Department of Clinical Laboratory Science, College of Applied Medical Science, Jouf University, Saudi Arabia.
*Corresponding Author E-mail:Hindhaidar011@gmail.com

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Fetuses and pregnant women are at risk of serious complications as a result of human parvovirus B19 infection, including non-immune hydrops, intrauterine fetal death, and fetal death. This study was conducted to determine the seroprevalence of Parvovirus B19 immunoglobulin G (IgG) and immunoglobulin M (IgM) antibodies among apparently healthy pregnant women and to identify the risk factors associated with B19 infection. Blood samples were collected from consenting pregnant women who were attending antenatal care units in Khartoum state, Sudan, from the period between September 2018 and August 2019. ?The enzyme-linked immunosorbent assay (ELISA) technique was used to detect Parvovirus B19 IgG and IgM antibodies in the sera of pregnant women. A structured questionnaire was administered to collect data on sociodemographic characteristics and potential risk factors. A total of 93 pregnant women were enrolled and screened for Parvovirus B19 IgM and IgG antibodies; their ages ranged from 15 to 45 years old. Overall, 8 (8.6%) were positive for Parvovirus B19 IgM antibodies, and 19 (20.4%) had IgG antibodies. Those aged 25-35 years had the highest prevalence of IgM antibodies 7 (7.5%) and IgG antibodies (13 (14%) there were a significant relations between B19 IgM, IgG seropositivity and age (p-value = 0.028,0.034) respectively, but no significant association between Parvovirus B19 seropositivity and gravidity, history of blood transfusion, or history of miscarriages (p>0.05). The prevalence of the B19 virus was low among apparently healthy pregnant women in Khartoum State, Sudan.

Keywords: ELISA; Parvovirus B19 IgG; Parvovirus B19 IgM; Pregnant Women; Sudan.

Parvovirus B19 (B19) is a small, non-enveloped DNA virus belonging to the genus Erythrovirus within the Parvoviridae family. [1] Infection with Parvovirus B19 occurs frequently around the globe, particularly in late winter or early spring, and typically spreads through respiratory droplets, including products like clotting factors. [2,3,4] Most individuals who contract B19V are

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asymptomatic or present with mild, non-specific symptoms resembling a cold, which are rarely associated with the virus.[5]

About 5% of expectant mothers become infected with parvovirus B19, yet most have a normal pregnancy outcome. The likelihood of infection increases during outbreaks. [6] Maternal viremia reaches its highest point roughly a week post-infection when the mother becomes infected. In about half of those infected, symptoms like erythema infectiosum, mild fever, joint pain, and headaches develop 10 to 14 days after the initial infection. The risk of transmitting the virus to the fetus is estimated to be around 25% when IgM antibodies first appear, likely during the peak viral load in the mother (around day 7). [7, 8]

Parvovirus B19 transmission is a significant contributor to non-immune fetal loss, spontaneous abortion, and intrauterine fetal deaths in pregnant women.[9] Research has indicated that miscarriages and intrauterine fetal deaths are more prevalent during the second trimester. [10, 11] Approximately 50% of women are at risk of B19 infection, which can lead to severe fetal complications such as anemia, spontaneous abortion, and hydrops fetalis.[12] The rate of maternal transmission of parvovirus B19 to the fetus ranges from 17% to 33%.[13] Detecting B19 infection early can assist in pinpointing individuals at risk and enhance the likelihood of fetal survival. Information regarding the occurrence of B19 infection in Sudan is quite sparse. Consequently, this research was conducted to offer additional insights into the serological detection of B19 infection.

MATERIALS AND METHODS

Study design

This study was conducted as a descriptive cross-sectional study from September 2018 to August 2019. The participants in the study were pregnant women visiting various Antenatal Care Units in Khartoum State.

Ethical considerations

This study was approved by the Ethics Committee of the Scientific Research Committee of the College of Medical Laboratory Sciences, Sudan University of Science and Technology, with ethical approval (Ethical No. SRC-MLS-011-19). Informed consent was obtained from all participants before their inclusion. All participants were informed about the study objectives and confidentiality measures.

Study population

Apparently healthy pregnant women, ranging in age from 15 to 45 years and at various trimesters of pregnancy, were included in this study. Before sample collection, clinical information, demographic information, and data on possible risk factors were collected from the women using structured questionnaires.

Sample size and sampling technique

A ninety three (n=93) blood specimens were collected from pregnant women using non probability convenience sampling techniques.

Specimen collection

Five milliliters (5 ml) of venous blood was aseptically collected from each subject. Serum was separated into Eppendorf tubes after centrifugation, then stored at "20°C till needed for analysis.

ELISA procedure

Anti-parvovirus B19 antibodies of the IgM and IgG classes were detected using an enzyme-linked immunosorbent assay (ELISA) (EUROIMMUN, Germany).

The reagents were diluted 1 to 20 times. The calibrator, positive control, negative control, and diluted specimen were then applied to the 96-wells microtiter plate in their respective wells. After covering the plate with protective foil, it was incubated for 60 minutes at 37°C ±1°C. The plate was washed three times with 300 ìL of working strength wash buffer. All liquid from the microplates was disposed of by tapping it on absorbent paper with the opening facing downwards to eliminate all residual wash buffer. In each of the microplate well, 100 ìL of enzyme conjugate was applied, and the plate was incubated for 30 minutes at room temperature (+ 18°C to +25°C) for IgM detection. In addition, 100 ìL of enzyme conjugate was applied to each microplate well, and the plate was incubated at room temperature (+ 18°C to +25°C) for 30 minutes for IgG detection. Then the plate was washed as described above.

Each well received 100 ìl of chromogen/substrate solution, which was incubated for 15 minutes at room temperature (+18°C to +25°C) and away from direct sunlight. Then, the 100 ìl of stop solution was applied to each microplate well, and after 30

minutes, a photometric measurement of the color intensity was taken at a wavelength of 450 nm.

Reading and interpretation of the results

The ratio of the extinction value of the control or patient sample was calculated over the extinction value of the calibrator to test the results semi-quantitatively. To measure the ratio, the following formula was used:

Extinction of the calibrator = ratio Extinction of the control or patient sample

Interpretation of the results:

Ratio <0.8= negative

Ratioe"0.8 to <1.1= borderline

Ratioe"1.1= positive

Statistical analysis

The Statistical Package for Social Science (SPSS) version 16.0 was used to analyze the collected data. The data were expressed as percentages and ranges as appropriate. Comparisons were made using the Chi-square test. The results were reported with P d" 0.05 as the accepted level of significance.

RESULTS

In this study, 93 gestating women between the ages of 15 and 45 who were apparently healthy participated. Most of them (50.8%) were between 20 and 35 years old. According to gestational age, 37 (38.7%) and 40 (43%) were in the second and third trimesters, respectively, while 17 (18.3%) were in the first trimester, as shown in Table 1.

According to Figure 1, Parvovirus B19 antibodies identified 19 (20.4%) IgG positives and 8 (8.6%) IgM positives.

Pregnant women aged 20 to 35 years had the highest prevalence of anti-parvovirus antibodies, whereas those aged 35 and older had the lowest prevalence of anti-parvovirus antibodies (0%) and (5), respectively. There was also a correlation between age and both IgG antibodies (p = 0.028) and anti-parvovirus antibodies (p = 0.028).

Table 1. Demographic data of the study population (Authors' own data)

Age groups/ years	Frequency	Percentage
< 25	12	12.9%
25-35	50	53.8%
> 35	31	33.3%
Total	93	100%
Trimester	Frequency	Percentage
First	17	18.3
Second	40	43.0
Third	36	38.7
Total	93	100%

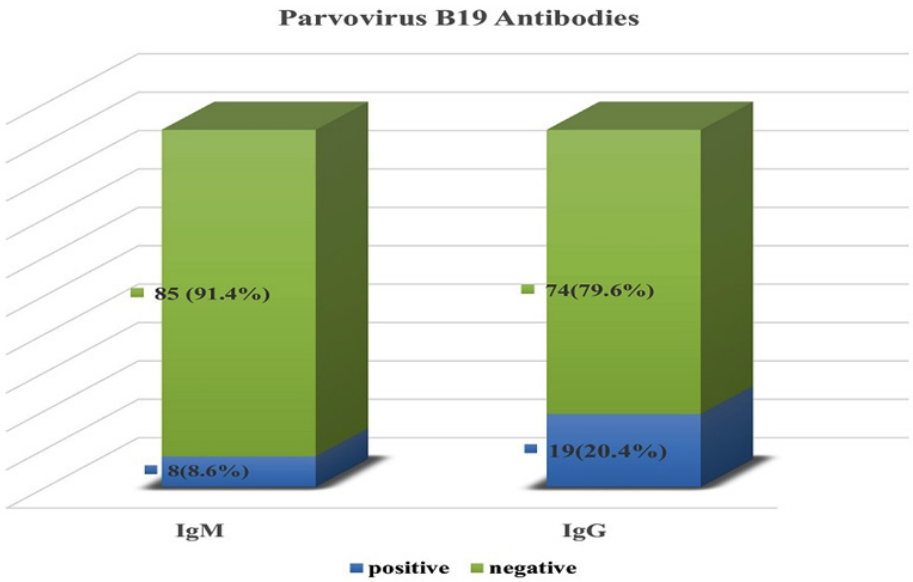

Fig. 1. Prevalence of Parvovirus B19 IgM and IgG antibodies among apparently healthy pregnant women (Authors' own data)

Table 2. The association between possible risk factors and IgM, IgG results among apparently healthy pregnant women (Authors' own data)

Variable		IgM		IgG	
		Positive	Negative	Positive	Negative
Age group/Years	<25	1(1.1%)	11(11.8%)	4 (4.3%)	8(8.6%)
	25-30	7 (7.5%)	43 (46.3%)	13 (14%)	37(39.8%)
	>30	0) 0%)	31 (33.3%)	2) 2.1%)	29(31.2%)
	Total	8(8.6 %)	85(91.4%)	19(20.4%)	74(79.6%)
	P. value	0.028	0.034		
Trimester	1st trimester	2 (2.1%)	15 (16.1%)	6) 6.4%)	11 (11.9%)
	2nd trimester	2 (2.1%)	38 (40.9%)	8)8.6%)	32) 34.4%)
	3ed trimester	4 (4.3%)	32 (34.4%)	2) 2.1%)	31 (33.3%)
	Total	8 (8.6%)	85 (91.4%)	19(20.4%)	74)79.6%)
	P. value	0.542	0.086		
Gravidity	Primigravida	2(2.1%)	25(26.9%	4 (4.3%)	23(24.7%)
	Multigravida	6(6.5%)	60(64.5%)	15(16.1%)	51(54.9%)
	Total	8(8.6 %)	85(91.4%)	19(20.4%)	74(79.6%)
	P. value	0.576	0.289		
History of miscarriage	Yes	2(2.1%)	18(19.3%)	3(3.2%)	17(18.4%)
	No	6(6.4%)	67(72.1%)	16(17.2%)	57(61.2%)
	Total	8(8.6 %)	85(91.4%)	19(20.4%)	74(79.6%)
	P. value	0.548	0.370		
History of blood transfusion	Yes	8(8.6%)	83(89.2%)	18(19.3%)	73(78.5%)
	No	0(0%)	2(2.2%)	1(5.3%)	1(1.1%)
	Total	8(8.6 %)	85(91.4%)	19(20.4%)	74(79.6%)
	P. value	0.835	0.369		

The majority of pregnant women with anti-parvovirus B19 IgM antibodies were in their third trimester (4, 4.3%), while those with IgG antibodies were primarily in the second trimester (8, 8.6%). There was no association between gestational age and IgM (p. value = 0.542) or IgG (p. value = 0.086) anti-parvovirus antibody seropositivity.

Anti-parvovirus B19 IgM antibodies were present in 6 (6.5%) and 15 (16.1%) of multigravida pregnant women; gravidity age showed no association with anti-parvovirus IgM (p = 0.576) or IgG antibodies (p.value = 0.289).

There was no association between history of miscarriage and seropositivity of anti-parvovirus IgM (p value = 0.548) or IgG antibodies (p. value = 0.370).

The majority of pregnant women have a history of receiving blood transfusions. Blood transfusion history and seropositivity to anti-parvovirus IgM (p.value = 0.835) and IgG antibodies (p.value = 0.369) did not associate with one another, as illustrated in Table 2.

DISCUSSION

The prevalence of parvovirus B19 IgM antibodies in this study was 8 (8.6%), which is interestingly similar to other studies conducted in Libya by Elnifro et al. (5.3%), [4] in Nigeria by Emiasegen et al. (13.2 %), [14] and in Iran by Keikha et al. (10.3 %).[15]

This research had a higher seropositive rate than a previous study conducted in Sudan by Adam et al. [(16)] which found that 0.2% of people were seropositive. This can be attributed to several factors: the large sample size, the significant time gap between the two studies (since 2008), the use of different types of ELISA kits, and the challenge of avoiding parvovirus B19 infection, which is often asymptomatic and commonly encountered during epidemics.

According to Ghazi, [17] the prevalence of IgM in Saudi Arabia was 25 %, which is lower than in the current study; this is likely due to seasonal demographics and geographic differences.

The prevalence of parvovirus B19 IgG antibodies found in this study was 20.4%, which matched other studies by Emiasegen et al. [14] in Nigeria (27.5%) and Keikha et al. [15] in Iran (21.8%).

In this study, the seropositivity rate was lower than in previous studies conducted in Sudan by Adam et al.[16] with a seropositivity rate of 61.4%, and in in two Saudis studies on pregnant women by Ghazi [17] and Johargy, [18] with a seropositivity rate of 46.6% and 50%, respectively. This could be due to the large sample sizes (500 and 1200, respectively) and demographic variations geographically and seasonally.

The study found that most participants (50.83%) were aged 20 to 35 years old. The prevalence of IgM and IgG antibodies increases with age, showing a strong association between the two, consistent with previous studies by Ghazi and Abraham.[19]

This study found women in their third trimester had the highest level of IgM antibodies (4.3%), followed by those in their first and second trimesters (2.1% for each). Unlike women in their second trimester had the highest level of IgG antibodies (8.6%), followed by those in their first and second trimesters (6.4% and 5.4%), respectively. This finding was in line with a prior study conducted in Nigeria by Emiasegen et al.[14] which may be because the majority of pregnant women in this study were in their third trimester. Furthermore, women in this study with seronegative IgG in the 1st and 2nd trimesters were 11 (11.9%) and 32 (34.4%), respectively. Consequently, those B19 IgG seronegative women might experience fetal loss or hydrops fetalis if infected with B19. Because there is a significant B19-associated risk of hydrops fetalis and/or fetal death if the mother is infected with B19V between 9 and 20 weeks of gestation. [4]

The prevalence of B19V antibodies was higher in multigravida pregnant women, with 6(6.5%) IgM and 15(16.1%) IgG positive, but there was no significant association. This finding agreed with Adam et al.[16] in Sudan, and there was no significant association between gravidity and seropositivity. In the present study the seropositivity of anti-parvovirus B19 IgG antibodies among pregnant women with a history of blood transfusion was 18(19.3 %). This was lower than the previous reported study done in Nigeria Emiasegen et al.[14] which found seropositivity to be 12(30.8 %). This could be attributed to the smaller sample size. No association found between miscarriage and the presence of anti-parvovirus B19 IgM and IgG antibodies.

CONCLUSION

In this, the prevalence of Parvovirus B19 IgM was lower, and IgG was slightly increased. Parvovirus seropositivity for IgM and IgG antibodies significantly associated with the age of pregnant women, whereas parvovirus B19 infection did not correlate with other factors (gravidity, history of miscarriage, history of blood transfusion). More accurate tests, such as antigen detection and molecular methods, must be used to confirm infection.

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Conflict of Interest

The author(s) do not have any conflict of interest.

Data availability

This work is a part of a full research article available in Sudan University of Science and Technology repository, https://repository.sustech.edu/handle/123456789/23836

Fighshare: Sero-Frequencey of parvovirus B19 among Apparently Healthy Pregnant Women Attending Selected Antenatal Care Units in Khartoum State https://doi.org/10.6084/m9.figshare.12629855.v1 (Abdelrahim and Salih, 2020).

Data are available under the terms of the Creative Commons Zero "No rights reserved" data waiver (CC0 1.0 Public domain dedication).

Ethics Statement

This study was approved by the Ethics Committee of the Scientific Research Committee

of the College of Medical Laboratory Sciences, Sudan University of Science and Technology, with ethical approval (Ethical No. SRC-MLS-011-19).

Informed consent statement

Informed consent was obtained from all participants before their inclusion. All participants were informed about the study objectives and confidentiality measures.

Clinical Trial Registration

This research does not involve any clinical trials.

Permission to reproduce material from other sources

Not Applicable.

Authors' contribution

Marwa Hussein Abdalgabar: Principal investigator; Wafaa Mohammed Abdalla: Study design, visualization, Conceptualization and supervisor; Hind Haidar Ahmed: Methodology, data analysis, reference writing, editing, and reviewing the revised the manuscript critically; Samar Mohammed Saeed: Methodology and data analysis; Ahmed Bakheeet Abd Alla: Writing the draft paper; Tagwa Salah Ahmed: Data analysis and interpretation.

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