



Seroprevalence of parvovirus B19 infection in patients with beta thalassemia major in Fayoum University Hospital

Mohamed E. Al Ghwass*, Shahera M. El Shafei, Wael S. Mohamed, Basant S. Mohamed

Fayoum University, Egypt

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KEYWORDS

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Abstract *Background and objectives:* Patients with beta thalassemia major are at higher risk of acquiring reticulocytopenia, and cessation of erythropoiesis after exposure to parvovirus B19. The aim of the study was to detect the seroprevalence of parvovirus B19 infection in children with transfusion dependent beta-thalassemia major through the detection of its specific IgM and IgG in their sera by Enzyme-linked Immunosorbent Assay (ELISA) in the Pediatric Department of Fayoum University Hospital.

Materials and methods: A cross sectional descriptive study was carried out from July to September 2013, in the Pediatric Department of Fayoum University Hospital in Egypt. A total of fifty-five children with beta-thalassemia major aged from 2 to 16 years were enrolled in the study. All patients were subjected to history taking, examination and investigations including; Complete blood count, serum creatinine, blood urea, hepatitis B surface antigen (HBsAg), anti hepatitis C virus antibody (anti HCV), reticulocytic count, anti B19 IgM and IgG.

Results: Anti-B19 IgM antibodies (recent infection) were detected in 14.5% (8 patients), while anti B19 IgG antibodies (prior infection) were detected in 18.2% (10 patients) of transfusion dependent thalassemic patients. There was a significant statistical correlation between the history of other siblings with beta thalassemia major, the hepatitis C virus and hepatitis B virus infection regarding absent, prior and recent infection.

Conclusion: Parvovirus B19 infection is detected in high rates among children with beta thalassemia major. Measures to avoid iatrogenic and nosocomial transmission have to be implemented including screening of donated blood for B19 especially blood given to patients with hematological disorders. Also data from this study support the need for introduction of an approved B19 vaccine that primarily protects children with thalassemia major against that infection.

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Abbreviations: B19, parvovirus B19; IgM, Immunoglobulin M; IgG, Immunoglobulin G; ELISA, Enzyme-linked Immunosorbent Assay; HBsAg, hepatitis B surface antigen; Anti HCV, anti hepatitis C antibody

* Corresponding author at: 31A Tatweer Fifth Settlement, New Cairo, Cairo, Egypt.

E-mail address: mghwass@gmail.com (M.E. Al Ghwass).

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Introduction

In Egypt, it was estimated that 1.000/1.5 million per year live births will suffer from thalassemia.¹ Patients with beta thalassemia major, are at higher risk of acquiring aplastic crisis, sudden worsening of anemia, reticulocytopenia, and cessation of erythropoiesis of the bone marrow after exposure to parvovirus B19.² Also B19 infection results in a severe drop of hemoglobin values and anemia which could be life-threatening.³ Transmission of B19 occurs via respiratory droplets, contaminated blood, organ transplantation and vertical transmission from mother to fetus.⁴

The aim of this study was to detect the seroprevalence of parvovirus B19 infection in multitransfused beta-thalassemia major patients at Pediatric Department of Fayoum University Hospital.

Materials and methods

A cross sectional descriptive study was carried out from July to September 2013, in the Pediatric Department of Fayoum University Hospital in Egypt. A total of 55 children (26 female and 29 male) aged from 2 to 16 years with mean age 6.2 ± 13.25 years were enrolled in the study; all of them have transfusion dependent beta thalassemia major and are receiving medical care at Pediatric Department of Fayoum University Hospital. All patients were under a hypertransfusion regimen and treated with regular transfusion of packed red blood cells. Exclusion criteria were patients with other hemoglobinopathies such as sickle cell anemia and anemia due to other causes.

All the included subjects were treated according to the Helsinki Declaration of biomedical ethics,⁵ informed consent was obtained from the parents, and the Ethics Committee of the Medical Faculty of Fayoum University approved the study protocol.

Data collection was carried out using a field pre-tested interviewing questionnaire covering the following elements:

- History of thalassemia; complications and frequency of blood transfusion along the course of the disease.
- History of B19 infections manifestations; characteristic "slapped cheeks" rash, joint manifestations, previous or recent episodes of transient aplastic crisis "TAC" (severe pallor, tachycardia, tachypnea $\pm$ a rash).
- History of vaccination especially to hepatitis B virus.

A fully detailed clinical examination for identifying recent B19 infections and the manifestations of the chronic hematologic conditions was done, documented in the form of check-lists for B19 fulfilling all useful items of personal data, history and clinical examination. Collection of all lab data from patients' files e.g. CBC, kidney function (urea and creatinine), liver functions (ALT and AST) and hepatitis B and C virus status.

Sample collection and preparation

Ten milliliters of blood was drawn from the antecubital fossa of each subject after obtaining informed consent. The 10 ml

of blood was divided into two aliquots, 3 ml was added into EDTA anticoagulated bottle and was used for basic complete blood count (CBC), while the remaining 7 ml was added into a plain specimen container allowed to clot. After clotting, blood samples in the plain tubes were centrifuged at 3000 rpm for 10 min and the resulting sera were collected into separate cryo tubes for biochemistry and serological analysis of B19 IgM and IgG antibodies. The serum samples were used immediately or stored at -20°C until tested.

Determination of IgG and IgM antibodies against parvovirus B19 by in vitro diagnostic Enzyme immunoassay

Quantitative immunoenzymatic determination of IgG and IgM antibodies against B19 based on the ELISA (Enzyme-linked Immunosorbent Assay) technique, using one of the commercially available ELISA kits (RIDASCREEN® B19 IgG kits) manufactured by R-Biofarm, Germany, with test sensitivity 91.3% and specificity 100%.

- According to the results of parvovirus B19 IgG and IgM antibodies by ELISA, patients were categorized into:
No infection if both Ig M & Ig G were negative;
prior infection if IgG +ve and IgM -ve;
recent infection if IgM +ve and IgG -ve or +ve.

Statistical analysis

Data were coded and entered using the statistical package SPSS version 15. The data were summarized using descriptive statistics: mean, standard deviation, median, minimum and maximum values for quantitative variables and number and percentage for qualitative values. Statistical differences between groups were tested using Chi Square test for qualitative variables, independent sample test and ANOVA (analysis of variance) with postHoc Scheffe test for quantitative normally distributed variables while nonparametric Mann-Whitney test and Kruskal-Wallis test were used for quantitative variables which aren't normally distributed. *P*-values less than or equal to 0.05 were considered statistically significant.

Results

A total of 55 children were enrolled in the study: 22 patients (40%) aged < 5 years, 26 patients (47.2%) aged 5–10 years and 7 patients (12.8%) aged > 10 years. According to the results of parvovirus B19 IgG and IgM antibodies by ELISA, it was found that 37 patients (67.3%) had no infection (both IgM & IgG were negative), 10 patients (18.2%) had prior infection (IgG +ve & IgM -ve) and 8 patients (14.5%) had recent infection (IgM +ve & IgG -ve or +ve). As regards the clinical manifestations of parvovirus B19; transient aplastic crises were found in 2 patients (3.6%) also 2 patients (3.6%) had a history of arthralgia and no one had a rash while 96.4% were asymptomatic. History of vaccination against hepatitis B was irrelevant. As regard to number of blood transfusions: Of children who received less than 10 blood transfusion (6 patients), 2 of them had no infection, 2 had prior infection and 2 had recent infection. Of children who received more than 10 blood transfusions (49 patients), 35 of them had

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