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Short Article

Identification of First Patient With Rh Null Phenotype in Southeast Iran

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Background and Aims: $Rh_{null,}$ with an estimated incidence of one per 6,000,000 individuals, is an extremely rare disorder with an autosomal recessive pattern of inheritance that is more common in societies with a high rate of consanguinity.

Materials and Methods: In this study, we report the first case with $Rh_{null,}$ a blood group phenotype in southeast Iran, which was diagnosed during pretransfusion testing.

Results and Conclusions: A 21-year-old woman with a positive parents' consanguineous marriage was found to have an unusual reaction with all packed red blood cell units during routine pretransfusion cross-match testing in the hospital. The patient's serum was reacted with all screening and identification panel cells, suspected to have an alloantibody against a common antigen or multiple alloantibodies against her absence antigens. Further studies revealed negative results for C, c, E, and e, which are highly suspected of Rh_{null} phenotype. Confirmatory assessments were performed, including adsorption and elution studies and Rh phenotyping of patients, along with known positive and negative controls. Due to the blood requirement of the patient, we performed serological studies on the patient's family members and found that her sister also has a Rh_{null} phenotype. Blood transfusion from her sister's donated units was performed, and the pregnancy was ended without any complications. Finally, due to the rarity of the Rh_{null} phenotype, early identification of individuals and autologous or compatible allogeneic blood transfusion should be planned prior to selective or emergency surgeries.

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Introduction

Rh_{null} is an extremely rare autosomal recessive blood group phenotype with an estimated prevalence of one per 6,000,000 individuals [1, 2]. The Rh blood group system with about 50 different antigens is the most polymorphic human blood group and the most clinically significant blood group next to ABO [3]. The Rh blood group system is one of the most complex blood group systems with a significant effect on the erythrocyte structure [4]. Patients with Rh_{null} phenotype do not express any Rh antigens and therefore have red blood cells with somatotype phenotype and experience chronic hemolytic anemia [3]. In addition, patients with Rh_{null} syndrome have other erythrocyte abnormalities, including spherocytosis, increased osmotic fragility, elevated Na1/K1 ATPase activity, and others [3, 5]. A patient with a Rh_{null} phenotype may be Rh_{null}-amorph or Rh_{null}-regulator; both may produce an antibody against all Rh antigens, which is also known as anti-total Rh or anti-Rh29, which is a significant clinical and laboratory challenge [3, 4]. The main difference between these two forms of Rh_{null} is the molecular basis of the disorders [5]. Due to the rarity of the syndrome, patients with Rh_{null} are at a high risk of a lack of blood supplements in emergencies like surgery. Therefore early identification and registration of patients with rare blood groups is of special importance. Due to this requirement, an International Society for Blood Transfusion (ISBT) working party report on rare blood donors was established to facilitate blood availability for patients with rare blood groups [6]. Due to the importance of this issue in the present study, we reported the first family with Rh_{null} phenotype in southeast Iran.

Materials and Methods

The patient was referred to the immuno-hematology laboratory of Zahedan Blood Transfusion Organization for compatibility testing. Initially, blood grouping was performed on the patient's fresh samples using anti-A, anti-B, and anti-AB negative, A-cell, and B-cell. In the next step, antibody screening with a homemade 3-cell panel was performed to determine the presence of any unexpected antibody in the patient's serum. After observation of a positive reaction on the antibody screening step, antibody identification was performed using the Iranian Blood Transfusion Organization (IBTO) homemade antibody identification 11-cell panel. Finally, red blood cell phenotyping of the patient was performed by anti-C, anti-c, anti-E, and anti-e antibodies.

Due to a lack of specific assays in the immunohematology laboratory of the Zahedan Transfusion Organization, the patient's sample was sent to the immunohematology reference laboratory of the IBTO in Tehran. Confirmatory tests were performed for the index cases in the immunohematology reference laboratory, as previously described in detail [7]. The study was proved by the ethical committee of the Blood Transfusion Research Center, High Institute for Research and Education in Transfusion Medicine, Zahedan, Iran.

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Results and Discussion

In her first delivery, a 21-year-old woman with positive parents' consanguineous marriages was referred to the immunohematology laboratory of Zahedan Blood Transfusion Organization for pretransfusion testing. In the hospital cross-match of the patient, they found a strong reaction (+4) of the patient's serum with all available red blood cell units. Therefore, the patient's sample was sent to the immuno-hematology laboratory of Zahedan Blood Transfusion Organization for compatibility testing. In blood grouping, the following reactions were observed: Anti-A, anti-B, and anti-AB negative, A-cell, and B-cell positive (4+), which were compatible with O negative blood group. Antibody screening with a homemade 3-cell panel revealed a positive reaction with all panel cells, which was highly suspected to be an antibody against a common panel cell antigen or multiple alloantibodies against the patient's absence antigens. Further studies on the Iranian Blood Transfusion Organization (IBTO) homemade antibody identification panel (11 cell panel) revealed a widespread reaction with all panel cells. Following this reaction, further red blood cell phenotyping was performed by anti-C, anti-c, anti-E, and anti-e, which showed a negative reaction for all antibodies, highly suspected of Rh_{null} phenotype (Table 1).

Table 1. Characteristics of a family with Rh_{null} phenotype

Case	Anti-A	Anti-B	Anti-AB	Anti-D	Anti-C	Anti-c	Anti-E	Anti-e
Index case	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg
Son	Neg	Neg	Neg	Neg	Pos	Neg	Neg	Pos
Sister	Neg	Neg	Neg	Neg	Neg	Pos	Neg	Pos
Brother	Neg	Neg	Neg	Neg	Neg	Pos	Neg	Pos
Father	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg
Mother	Neg	Neg	Neg	Neg	Neg	Neg	Neg	Neg

Immunohematology reference laboratory confirmed the Rh_{null} phenotype of the patient. Due to the requirement of the patient for blood transfusion and the autosomal recessive inheritance pattern of Rh_{null}, we called her family members for blood donation. Her parents and sister were referred to the immunohematology laboratory of the Zahedan Transfusion Organization for further assessments (Table 1). She had a sister, and primary and confirmatory serological testing, including the Rh system phenotyping, revealed that she is also a Rh_{null} individual. Blood was collected from the patient's sister and transfused to the patient, and the pregnancy ended without complications.

Rh_{null} is an extremely rare condition that could be found in one per 6,000,000 individuals worldwide. Although the precise incidence of the syndrome is unknown, societies with a high rate of consanguineous marriages have a high rate of this condition. A few individuals with Rh_{null} syndrome have been reported in different counties, including Iran, India, Europe, the

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United States, and others [3, 7, 8]. Most patients with Rh_{null} syndrome have been identified randomly in pretransfusion testing, similar to the present case and previously reported Iranian case [3, 7].

In patients with Rh_{null} syndrome, all the Rh blood group system antigens are absent [3]. Patients with Rh_{null} syndrome could have a severe blood transfusion reaction following Rh-positive blood transfusion. Thus, appropriate management of patients, particularly during surgery, is an important issue. Patients with Rh_{null} phenotype can produce anti-Rh29, anti-Rh17, or a mixture of antibodies. The transfusion process in patients with these antibodies, particularly anti-Rh29, which is a total Rh antibody, is a significant challenge and only can give blood from an individual with a Rh_{null} phenotype, as observed in the present case [3, 4].

Identifying one case with Rh_{null} is a significant diagnostic clue for identifying other family members with the same phenotype. In the present study, randomly diagnosing one individual with Rh_{null} syndrome leads to diagnosing another family member with the syndrome. Previous studies have reported chronic hemolytic anemia with stomatocytosis in patients with Rh_{null} syndrome [3, 7]. In this study, we observed erythrocytes with somatotype morphology and chronic anemia in the patient. Anemia is mild and does not require any blood transfusion, as observed in the present case. There are only a few reported cases with Rh_{null} phenotype in Iran, and supply of blood supplements for patients with such a rare blood group phenotype is a significant challenge, and autologous blood transfusion is a crucial lifesaving choice for patients with such a rare phenotype. Establishing the ISBT, working party on rare donors, and identifying such individuals and their blood supply may be less challenging [6]. Alternatively, close-relative such as brothers and sisters may have a common blood group phenotype with patients with Rh_{null} phenotype and can be selected for blood transfusion, especially in emergencies, as described previously by other Iranian researchers (7). In the present study, we also used a close relative of the patient—her sister—as a selective donor to manage her complicated pregnancy that required two units packed red blood cells for blood transfusion.

Conclusion

Early identification of patients with rare blood group phenotypes is an important issue that can help in the timely management and appropriate treatment of patients in emergencies. Identifying and registering patients with rare blood phenotypes is straightforward for the early management of the individuals.

Conflict of Interest

The authors declare that they have no competing interests.

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