Original Article:

The incidence of ABO, Kell and Rh system blood groups in general population of Qazvin, Iran

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ABSTRACT

Introduction:ABO antigens and the glycoproteins constituting the blood groups such as Kell and Rh systems are the mostly focused blood groups in transfusion medicine. Their importance is tightly associated with the presence of natural isoantibodies, their protein structure, and immunogenicity. The aim of our study was to assess the distribution of major Rh and Kell antigens and the most probable genotype in a normal population of Qazvin city, Iran.**Materials and methods:**This study was done on 1000 healthy people who were candidates for getting driver's license. The blood samples were tested for Kell, ABO and major Rh antigens by standard tube agglutination method.**Results:**Out of 1000 samples studied, the prevalence of RhD was 86.6%. The incidence of other Rh antigens i.e. C, E, c and e was 73%, 29.8%, 72.1%, and 95.9% respectively. The most common phenotype in the samples was DCce and the least one was shown to be CcEe. Kell antigen frequency was 8.2%. On the other hand, 91.8% of people were indicated to be negative for the Kell antigen.**Conclusion:**Taken together, the amount of the individuals negative for the Kell and Rh system are adequate to provide new policies for identification of these antigens for both blood donors and recipients.

Keywords: ABO; Rh; Kell; Blood group

INTRODUCTION

Over 30 blood group systems and 600 blood group antigens are described since 1940 [1, 2]. ABO and glycoproteins are the key instructions in the major blood group systems such as Rh system and Kell which are the most important antigens in transfusion medicine. ABO blood group system has unique properties considered as the the most important blood group in blood banking procedures. Its importance is due to the incidence of natural isoantibodies in the sera of people who lack these antigens. These antibodies are usually IgM and can cause intravascular hemolysis in the cases of mismatch blood transfusion [3]. While the importance of Kell and Rh blood group systems is primarily due to their protein structure and immunogenicity. It should be noted that the presence of polymorphisms in the Rh antigens highlights its complexity [3-5]. The blood groups are inherited traits and usually useful for population genetics investigation and more importantly in blood banking departments and in the pathogenesis of hemolytic disease of newborns in perinatology cases [6]. In developing countries such as Iran during routine blood grouping, only ABO and RhD antigens are tested while other Rh antigens and Kell group are missed. This can increase the incidence of alloantibodies among general population especially among patients who are transfusion dependents such as major beta-thalassemia and aplastic anemia and patients under bone marrow suppressive chemotherapies [7-9]. It may complicate the clinical manifestations of mentioned patients and also burden much more medical costs to the health systems in developing countries. Therefore, the aim of this study was to determine ABO, Rh system, and Kell phenotypic frequency in general population of Qazvin city in order to provide beneficial data for the health system managers, and predicting the risk of alloimmunization.

MATERIALS AND METHODS

This cross-sectional descriptive study was performed over a period of two months from August to September 2018. As much as 1000 healthy people who referred to public hospital for health approval in order to get a driver's license were selected by signing the written consent. ABO, Rh & Kell antigens were determined according to the standard cell type agglutination tube test using commercially available antisera as recommended by the manufacturer. Briefly, one drop of cell suspension (2-5%) and two drops of corresponding antisera were mixed. Before centrifugation, the tubes were incubated for 5 minutes at room temperature. For Kell determination, the corresponding tubes were incubated in 37C˚ for 30 minutes. Then, the tubes were centrifuged at 1000 RPM for 1 minute and the reactions were checked for agglutinations. In all the procedures, negative and positive controls were included. Rh-subgroups negative or weak reactions were checked by microscopes and were followed by coomb's test in negative cases. The ABO grouping results were confirmed by back typing. Briefly, the plasma samples were incubated with determined ABO cell suspensions at room temperature for 5 minutes and then centrifuged at 1000 RPM for 1 minute and checked for agglutination. Statistical analysis of the resulted data was performed by Microsoft office excel software. This study received the ethic approval from Qazvin University of Medical Sciences, ethic Number: IR.QUMS.REC.1394.220.

RESULTS

In this study, the participants were 63% female and 37% male. The subjects' age ranged from 18 to 52 years old. The ABO phenotyping showed that O, A, B, and AB groups are the most and least frequent types, respectively. No significant difference between male and female groups was detected in this case. Table 1 and 2 show ABO, Rh system, and Kell antigen distribution in the subjected population. In the Rh system, the highest frequency belonged to "e" antigen followed by D, C, c, and E.

Table 1. ABO and Rh system antigens' frequency in general population of Qazvin city

Antigens	Frequency (%)
A	40.1
B	30
D	86.6
C	73
C	72.1
E	29.8
e	95.9
Kell	8.2

Table 2. ABO group with Rh (D) frequency in general population of Qazvin city

ABO with Rh	Frequency (%)
A+	28.4
A-	4.3
B+	19.8
B-	3.1
AB+	6.3
AB-	0.8
O+	32.6
O-	5

Since no genetic test was conducted on the Rh system in the mentioned population, according to the people race (white), the frequency of Rh phenotypes was presumed as described in Table 3.

Table 3. Rh phenotypes and presumed genotype in the in general population of Qazvin city (white race)

Antigens					Phenotypes	Incident Genotypes		Frequency (%)
D	**C**	**c**	**E**	**e**		**DCE**	**Rh-hr**	
+	+	+	-	+	DCce	R^1r	DCe/dce	29
+	+	-	-	+	DCe	R^1R^1	DCe/DCe	25.8
+	+	+	+	+	DCcEe	R^1R^2	DCe/DcE	15.6
+	-	+	-	+	Dce	R_0r	Dce/dce	4.1
+	-	+	+	+	DcEe	R^2r	DcE/dce	8.8
+	-	+	+	-	DcE	R^2R^2	DcE/DcE	3.4
+	+	-	+	+	DCEe	R^1Rz	DCe/DCE	1.2
+	+	+	+	-	DCcE	R^2Rz	DcE/DCE	0.3
-	-	+	-	+	Dce	Rr	dce/dce	10.2
-	+	+	-	+	dCce	r'r	dCe/dce	1.4
-	-	+	+	+	dcEe	r˝r	dcE/dce	0.4
-	+	-	-	+	dCe	r''r'	dcE/dCe	0.6
-	+	+	+	+	dCcEe	r'r'	dCe/dCe	<0.01*

*Rare

DISCUSSION

Comparison of our results with other studies from Iran is in agreement with earlier studies from a different province [10, 11]. However, there are some variations between our findings and other reports from different countries [2, 12, 13]. This difference in Rh and Kell system distribution is clinically significant because Rh and Kell systems are the most clinically important blood groups after ABO system. More than 50 antigens have been identified in the Rh system; however, the D, C, E, e and c are among those which are mostly mentioned in clinical and population studies [4, 14]. For the following reasons similar studies are required in different parts of the world and each country:

Determining of the frequency and organizing the negative blood groups

Given to the prevalence of important clinical blood groups such as Kell and Rh systems, blood transfusion organization will be able to accurately organize their resources and provide a national and regional reserves of negative blood groups, particularly in the regions with high prevalence of specific antigens. This will provide the blood supply to high-risk people with negative groups and those who are blood transfusion dependent and already have alloantibodies due to previous blood transfusion such as thalassemia patients. For example, in the present study, the "e" antigen with the prevalence of 95.5% was shown to be the most frequent Rh antigen in Qazvin province. This suggests that 4.5% of the total population lack this blood group and need proper blood storage. This issue also applies to the Kell and other clinically important blood groups. In a multi-central study from Iran, Azarkeivan et al. screened thalassemia patients for RBC alloantibody formation. They reported a frequency of 12.1% for the alloantibodies against anti-Kell [15]. They also showed that the most double alloantibodies are against the combination of two Rh subgroups or combination of D and Kell antigens [16].

Anticipate and prevent the hemolytic disease of newborns

Hemolytic disease of newborns (HDN) is caused by blood group incompatibility between the mother and her fetus. In this case, the mother produces a specific antibody through immunization to the fetus blood group antigens which are not present in the mother's body. A better understanding of the distribution of clinically important blood groups among general population and couples could help the relevant institutions to predict HDN and to prevent its adverse consequences [17]. In a recent study in Iran, Shahverdi *et al.* showed that 4.5% of pregnant women had formed the alloantibodies. Anti-D was the most common alloantibody constituting 70.5% of all the antibodies. The prevalence of alloimmunization other than Rh group was 14.4% [18].

Revision and changing policy in national blood banking and pre-transfusion tests

A better insight into the distribution of Rh and Kell systems in a normal population can also provide proper predictive policies for pre-transfusion testing and blood banking. For

example, in our study, we demonstrated that co-expression of E and c antigens among the population were 96%. Thus, the incidence of E+/c- phenotype is rare in the population. Our findings can help to remove the unnecessary routine Rh antigen typing in the populations who are negative for especial antigens. This also offers a cost-effective blood transfusion system and precipitates the emergency blood transfusion protocols. Since the normal population and patients in our province in a large extent are genetically homogenous, it is notable to gather valuable data on the common Rh genotypes, since it can lead to a better management of blood products and their distribution among high-risk patients especially the transfusion-dependent patients. For instance, DCce is the most common Rh phenotype in Iranian population followed by DCe, thus it can be predicted that the most common alloantibodies among transfusion-dependent patients are anti-D and anti-E. Studies in Iran imply that most common alloantibodies in thalassemia patients are anti-Kell and anti- D, respectively [15, 19]. These findings are of a great importance due to the fact that the Kell antigen typing is not performed routinely on the blood products in Iran. Consistently we and others reported that anti-kell formation frequently occurs in blood recipients [15, 20]. On the other hand, as the availability of routine Rh typing, the anti-D prevalence is significantly low in Iranian population. This also highlights the importance of determining the weak or negative results in D typing. The significance of this confirmation is based on the fact that typing methods have different sensitivities and the results are influenced by many factors such as personal experience, test reagents etc. A negative result by a less sensitive method such as tube test may be weak or moderately positive by a more sensitive method like gel method. The clinical importance of having insights into this fact is that weak positive or Du individuals considered as Rh positive when they are selected as donors, and Rh negative when they are blood recipients. The above-mentioned fact implies that national policies and guidelines in Iran need to be revised according to the recent surveys.

CONCLUSION

This study concludes that determining the prevalence of negative Kell and Rh antigens are crucial for taking new policies regarding the blood donors and recipients. This is an integral part of transfusion service when we are dealing with transfusion-dependent patients such as thalassemia patients. We have included the ABO, Rh system, and Kell antigens in this study, while other antigens such as Duffy and Kidd are also clinically important, thus further studies are required to elucidate the phenotypic characteristics in different populations for these antigens.

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