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Purwanto AB, Budi Mulyono, Jusak Nugraha, Rahajuningsih Dharma

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RESEARCH

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## **ANALYSIS OF RHESUS AND KELL GENOTYPE IN PATIENTS WITH TRANSFUSION REACTION**

*(Analisis Genotipe Rhesus dan Kell Pasien dengan Reaksi Transfusi)*

**Sukmawaty, Rachmawati Muhiddin, Mansyur Arif**

### **ABSTRAK**

Transfusi darah dapat memberikan reaksi imunologis. Yang paling sering adalah reaksi transfusi terkait pecahnya sel darah (hemolitik) akibat tidak terdapatnya antibodi. Anti serum (Ab) yang disarankan saat ini untuk mendeteksi keberadaan antigen sel darah merah yaitu ABO, Rhesus dan Kell. Penelitian ini bertujuan untuk mengetahui genotipe pasien Rhesus dan Kell dengan reaksi transfusi, menggunakan metode potong lintang di 35 subjek yang mengalami reaksi transfusi pada masa waktu antara bulan Juni–Agustus 2015 di RS. Dr. Wahidin Sudirohusodo Makassar. Semua sampel diperiksa antigen Rhesus dan Kell menggunakan sampel darah pasien yang ada di Bank Darah RS. Dr. Wahidin Sudirohusodo. Subjek penelitian rerata berumur 49,91 tahun dan sebagian besar berjenis kelamin perempuan (71,43%). Golongan darah yang paling banyak mengalami reaksi transfusi adalah golongan darah O (54,29%). Semua sampel adalah Rhesus positif (100%). Antigen D (100%), C (62,86%), c (40%), E (57,14%) dan e (91,43%). Antigen Kell diperoleh K (8,57%) dan k (91,43%). Dari semua sampel sebagian besar antigen yang didapatkan adalah antigen D dan e, serta k.

**Kata kunci:** Reaksi transfusi, golongan darah ABO, rhesus, Kell

### **ABSTRACT**

Blood transfusion may cause an immunologic reaction. The most frequent one is hemolytic transfusion reaction due to the antibody incompatibilities. Nowadays, antigens that are recommended for detecting the presence of erythrocytes antibodies are: ABO, Rhesus and Kell. This study is aimed to know the determination of the Rhesus and Kell genotype in patients with transfusion reaction. The cross-sectional study was conducted on 35 subjects who developed transfusion reaction within the period between June to August 2015 at Dr. Wahidin Sudirohusodo Hospital Makassar. All samples were examined for Rhesus and Kell antigen using patient's blood samples which were available at Dr. Wahidin Sudirohusodo Hospital Blood Bank. The average age of the subjects is 49.91 years old and most of the subjects are female (71.43%). The O group is the blood group which developed transfusion at the most reaction (54.29%). All samples have the positive Rhesus (100%), Antigen D (100%), C (62.86%), c (40%), E(57.14%), and e (91.43%). The researchers found Kell antigen K (8.57%) and k (91.43%), also found antigen D, antigen e and antigen k for the most part of all the samples.

**Key words:** Transfusion reaction, ABO blood group, rhesus, Kell

### **INTRODUCTION**

Blood transfusion is the donation of blood or their components from a healthy donor to a recipient. The blood consists of cells and plasma. The blood cell consists of erythrocytes, leukocytes and platelets. The transfusion of blood and their components requires ABO blood type and Rhesus (D) type examinations

as well as the cross matching examination between the donor and the recipient for avoiding agglutination or haemolysis reaction that can harm the recipient to death.<sup>1</sup>

Consequently, the blood transfusion is only conducted on the basis of indication and urgency, since if it is performed inappropriately and irrationally, it can generate fatal reaction. Human erythrocytes can

be classified into four groups, namely A, B, AB and O. After Landsteiner found the first blood group, ABO, there are also other blood types, such as MN, Levi, Duffy, Rhesus, Kidd, Lutheman and others. The other blood types have fewer clinical interests. The natural antibodies of P, Lewis and MN systems, for instance, usually only react at low temperatures (4°C) and resulting not in any clinical problems.<sup>1-3</sup>

The transfusion reaction, moreover, is an unfavorable situation that occurs during or after the transfusion. The transfusion reaction can occur as soon as the transfusion begins or several days after the transfusion. Consequently, precautions for transfusion reaction are required by performing certain examinations before the transfusion. For example, the blood type must be examined repeatedly by conducting cross matching to ensure that the donor's blood type matches the recipient's blood type.<sup>4</sup>

Hemolytic reaction, furthermore, is usually detected during an evaluation of antibody responses (Rhesus, Kell, Duffy, Kidd and other non-ABO antibodies) after exposure to antigens of the donor's erythrocytes. However, antibodies cannot directly be detected during cross matching process which is conducted before the transfusion since the antibody-antigen interaction is a secondary immune response detected after three (3) to seven (7) days.<sup>4</sup>

Blood transfusion also can provide an immunological reaction, a hemolytic transfusion reaction due to antibody incompatibility. Anti-serums currently recommended for detecting the presence of erythrocyte antigens are derived from ABO, Rhesus (Rh) and Kell blood types. Most of antibodies in ABO, are Rhesus and Kell blood types, which often trigger the hemolytic transfusion reaction and Hemolytic Disease of Newborn (HDN).<sup>5,6</sup>

Rhesus blood type is considered to be important because it is immunogenic. Anti-D, also known as anti-Rhesus, which has already existed naturally. But, if someone does not have D antigen in his body, the anti-D still can be produced if they are exposed to antigen D in blood transfusion, resulting in hemolytic transfusion reaction. The Rhesus blood type is governed by the structural genes, RhD and RhCE, encoding a membrane protein which carry D, Cc and E antigens. Phenotypically, RhD gene is known as Rh D + or Rh D.<sup>6</sup>

The Kell antigens, on the other hand, are the stronger antigens after those in ABO blood types. The Kell antigens also can lead to HDN and hemolytic

transfusion reaction. Kell antibody (anti-K) was discovered in 1946 by Coombs, antibody (anti-k) was discovered by Levine in 1949. The Kell blood type system has two forms, namely K (+) and K (-).<sup>6,7</sup>

In addition, genotype is a genetic structure in the organisms, related to certain traits. The genotype refers to the entire set of genes in a cell. In a single gene, there will be a particular character or trait that may exist in two allele forms, one of which is dominant and another is recessive. The gene that regulates Rhesus antigen is located in the short arm of chromosome 1. Fisher Race suggests that there are three loci on a chromosome, each of which is occupied by genes and alleles, namely C and c, D and d, as well as E and e. These three pairs of genes will form eight combinations of genetic variations of Rhesus, namely: cde, Cde, cDE, cDe, Cde, cdE, CDE and CdE. On the other hand, the gene that regulates Kell antigen is located on chromosome 7, which has two alleles, namely K and k.<sup>8</sup>

In anemia patients, according to a research conducted by Akre<sup>8</sup> most of transfusion reactions are affected by sickle cell (62.8%), 44.44% by Rhesus and 27.78% by Kell. The most common antibodies found are anti-E, anti-C and anti-KEL1. Another previous research performed by Icote<sup>9</sup> in South Africa reveals that antigen c and antigen e (99.85%) are mostly found in donors with Rhesus positive, followed with antigen C (21.97%) and antigen E (13.82%). Meanwhile, in donors with Rhesus negative there was none obtained.<sup>9</sup>

## METHODS

This research was a retrospective research with cross-sectional approach. This research was conducted by taking blood samples from patients undergoing transfusion reactions at the Blood Bank of Dr. Wahidin Sudirohusodo Hospital which have been taken between June to August 2015. The total number of blood samples was 35 (thirty-five) samples taken from patients with suffer transfusion reactions between June to August 2015 at the Dr. Wahidin Sudirohusodo Hospital in Makassar. Further on, they were examined for the Rhesus and Kell antigens using Rh-Subgroups + K BIO-RAD. Then the results obtained were analyzed descriptively. The data analyzed were classified based on the purpose and type of the data. The data then were processed and presented in the form of discussion and tables.

## RESULTS AND DISCUSSION

This research was conducted by taking blood samples of patients undergoing transfusion reactions from June to August 2015 at the Blood Bank of Dr. Wahidin Sudirohusodo Hospital in Makassar. During this period, there were thirty-five blood samples obtained. Those blood samples were taken from thirty-five patients undergoing the transfusion reactions, namely twenty-five females (71.43%) and ten males (28.57%). The age range of those patients was 27–63 years with the mean age of 49.91 years. The diagnosis of those patients then was classified into several groups, namely 14 patients (40%) for neoplasm group, 13 patients (37.14%) for metabolic endocrine, 4 patients (11.43%) for trauma bleeding, and 4 patients (11.43%) for other cases. The blood types of those patients was also divided into four

blood type groups, namely 3 patients (8.57%) for A, 8 patients (22.86%) for B, 19 patients (54.29%) for O and 5 patients (14.29%) for AB. All of those patients' rhesus is positive (100%). The further details of data are shown in Table 1.

The frequency of Rhesus and Kell antigens can be seen in Table 2. All the samples were Rhesus positive (D +). The second largest antigen obtained was antigen e about 32 samples (91.43%). Meanwhile, the number of samples for antigen C was 22 (62.86%), 14 samples (40%) for antigen c and 20 samples (57.14%) for antigen E. Antigen K was obtained in 3 samples (8.57%), while k antigen in 32 samples (91.43%). On the other hand, at Table 3 shows that there were seven genotypes obtained from the samples, most of which was R1R1 or R1r '(37.14%). Meanwhile, the most Kell genotype found was k + (91.43%).

**Table 1.** The characteristics of the research subjects

Variable	n (%)
Number of subjects	(n = 35)
Age: 27–63 years	35 (100)
Sex: Males	10 (28.57)
Females	25 (71.43)
Diagnosis: Neoplasm	14 (40)
Metabolic endocrine	13 (37.14)
Trauma bleeding	4(11.43)
Others	4 (11.43)
Blood types: A	3 (8.57)
B	8 (22.86)
O	19 (54.29)
AB	5 (14.29)
Rhesus: Positive	35 (100)
Negative	0 (0.0)

Source: Primary Data

**Table 2.** The frequency of Rh and Kell antigens on the patients' blood sample

Antigen	n	%
D+	35	100
d+	0	0
C+	22	62.86
c+	14	40
E+	20	57.14
e+	32	91.43
K+	3	8.57
k+	32	91.43

**Table 3.** The frequency of Rh and Kell genotypes

Antigen	Fisher-race Wiener	Genotypes	Total	%
D+C-E-c+e+	Dce/dce	R0r	3	8.57
	Dce/Dce	R0R0		
D+C+E-c-e+	DCe/dCe	R1 r'	13	37.14
	DCe/DCE	R1R1		
D+C-E+c+e+	DcE/dce	R2r	9	25.71
	DcE/Dce	R2R0		
D+C+E+c-e+	DCe/dCe	R1r''	6	17.14
	DCe/dCE	R1ry		
	DCe/DCE	R1Rz		
	DCE/dCe	Rzr'		
D+C+E+c+e-	DCE/dCE	Rzrz	2	5.71
	DcE/DCE	R2Rz		
	DcE/dcE	R2r''		
	DCE/dcE	Rzr''		
D+C+E+c-e-	DCE/DCE	RzRz	1	2.86
	DCE/dCE	Rzry		
D+C-E+c+e-	DcE/DcE	R2R2	1	2.86
	DcE/dcE	R2r''		
Kell				
K-k+			32	91.43
K+k+			3	8.57

Moreover, the samples used in this research were 35 (thirty-five) blood samples taken from patients undergoing transfusion reactions in the Blood Bank of Dr. Wahidin Sudirohusodo Hospital. Those samples were then examined using Rh-Subgroups + K BIO-RAD. Most of the samples were females, about 25 patients (71.43%) with a mean age of 49.91 year. All Rhesus acquired was Rhesus positive (100%). Similarly, a research conducted by Zoysa NS<sup>10</sup> shows that the Rhesus positive is more prevalent in Asian populations than in Europe.<sup>11</sup>

Besides, a research conducted by Akre<sup>8</sup> reveals that in sickle cell anemia patients, 62.8% of those patients receiving blood transfusions were affected by Rhesus and Kell antigens. Alloantibodies widely found in those patients were anti-E, anti-C and anti-Kell.<sup>9</sup> Unlike the

previous research, in this research, antigen e was the most antigen found in the patients undergoing transfusion reactions.<sup>9</sup>

In addition to antigen e (91.43%), Rhesus antigen which mostly found was antigen C (62.86%). Similarly, a research conducted by Bogui Siransi<sup>9</sup> also finds that the frequency of antigen e is more common than antigen C in Asian and European people.<sup>9</sup> Like the previous research, a research conducted by Thakral *et al.*<sup>11</sup> in North India also illustrates that in patients with Rhesus positive, antigen e is mostly found, followed by antigen C, antigen c and antigen E.<sup>11</sup>

Furthermore, there were seven genotypes found in this research. The most genotype found was genotype R1r '(37.14%). Unlike this research, a research conducted by Bogui Siransi<sup>9</sup> in Africa indicates that

the most common genotype was R0r, whereas in the white skin people the most genotype was R1r for the Rhesus positive blood group.<sup>9</sup>

In addition, antigen K in the Kell blood type system has a strong immunogenity after the Rhesus system. In this research, the results show that all of the samples examined had antigen k. Similarly, a research conducted by Bogui Siransi<sup>9</sup> reveals that the most antigen found in donors' blood is antigen k, while antigen K is more common in Europeans (whites).<sup>9</sup>

Rhesus genotype is usually used for paternity testing in patients with Hemolytic Disease of New Born (HDN) and for predicting HDN through Rhesus genotype examination on a father if the mother has anti D. The most common father's genotypes provide a 0%, 50%, or 100% chance for the child to have a D Rhesus positive antigen.<sup>11</sup> However, this research did not aim to discuss the correlation between the diagnosis and the genotypes of the patients undergoing transfusion reactions.

## CONCLUSION AND SUGGESTION

In conclusion, the most commonly found antigen in patients with Rhesus positive experiencing transfusion reactions is antigen e. Of the seven genotypes obtained, genotype R1r/R1R1 was mostly found in those patients. This study has some limitations, therefore should be followed by further research using larger samples and in addition to know the parents's genotype by screening in its association with the transfusion reactions.

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