Original article

doi:10.5633/amm.2026.0221

DETECTION OF D ANTIGEN VARIANTS AT THE BLOOD TRANSFUSION INSTITUTE IN NIS

Millica Randjelović, Zoran Stanojković, Danka Sokolović

Blood Transfusion institute of Niš, Niš, Serbia

Contact: Milica Ranđelović

48 dr Zorana Djindjića Blvd., 18000 Niš, Serbia

E-mail: milicarandjelovic990@gmail.com

The Rh system is very complex, polymorphic and the most important for clinical practice next to the ABO system. Antigen D is the most important antigen of the Rh system and the most immunogenic after the ABO antigen. Antigen D, which is made up of a mosaic of epitopes, is determined for all donors and patients. Monoclonal test sera have different affinities for antigen D epitopes. Determination of antigen D can cause operational difficulties.

This retrospective cross-sectional study was conducted at the Institute for Blood Transfusion in Nis. The study included voluntary blood donors from January 1 to December 31, 2024. A total of 45,970 voluntary blood donors were included in the study.

The aim of our research was to determine the serological prevalence of D weak in voluntary blood donors at our institution, as well as to analyze the Rh phenotype in such typed erythrocytes using serological methods, in order to enable the development of individualized transfusion strategies.

Of the total number of tested samples, 38,989 (84.82%) were D-positive, 6,847 (14.89%) D-negative, while the number of D weak was 134 (0.29%). The most common recorded D weak phenotype was CcDwee in 117 subjects (87.31%).

Serological methods depend on immunohematological techniques, and test reagents are not always able to unambiguously detect RhD variants. Therefore, RHD molecular typing is recommended to identify and confirm RHD variants.

Key words: D antigen, weak D, partial D, phenotype

Originalni rad

doi:10.5633/amm.2026.0221

DETEKCIJA VARIJANTI D ANTIGENA U ZAVODU ZA TRANSFUZIJU KRVI NIŠ

Millica Ranđelović, Zoran Stanojković, Danka Sokolović

Zavod za transfuziju krvi Niš, Niš, Srbija

Kontakt: Milica Ranđelović

Bulevar dr Zorana Đinđića 48, 18000 Niš, Srbija

E-mail: milicarandjelovic990@gmail.com

Sistem Rh je veoma kompleksan, polimorfan i najznačajniji za kliničku praksu pored sistema ABO. Antigen D najvažniji je antigen sistema Rh i najimunogeniji posle antigena ABO. Svim davaocima i bolesnicima određuje se antigen D, koji je sačinjen od mozaika epitopa. Monoklonski test serumi imaju različit afinitet za epitope antigena D. Određivanje antigena D može izazvati poteškoće u radu.

Ova retrospektivna studija preseka sprovedena je u Zavodu za transfuziju krvi u Nišu. Studijom su obuhvaćeni dobrovoljni davaoci krvi od 1. januara do 31. decembra 2024. godine. U studiju je uključeno ukupno 45.970 dobrovoljnih davalaca krvi.

Cilj našeg istraživanja bio je da se utvrdi serološka prevalencija D weak-a kod dobrovoljnih davalaca krvi u našoj ustanovi, kao i da se analizira Rh fenotip u tako tipiziranim eritrocitima korišćenjem seroloških metoda, kako bi se omogućio razvoj individualizovanih strategija transfuzije.

Od ukupnog broja testiranih uzoraka, 38.989 (84,82%) bilo je D-pozitivno, 6.847 (14,89%) D-negativno, dok je broj D weak iznosio 134 (0,29%). Najčešći zabeležen D weak fenotip bio je CcD^{w}ee i to kod 117 ispitanika (87,31%).

Serološke metode zavise od imunohematološke tehnike, a test reagensi nisu uvek u stanju da nedvosmisleno detektuju RhD varijante. Stoga se preporučuje RHD molekularna tipizacija za identifikaciju i potvrdu RHD varijanti.

Ključne reči: D antigen, weak D, partial D, fenotip

Introduction

The Rh system is the most complex of all erythrocyte blood group systems and contains 56 antigens. Antigens of the Rh system are encoded by two homologous, closely linked genes, located on the short arm of chromosome 1: RHD, which leads to the formation of antigen D and RHCE, which leads to the formation of antigens CC and Ee. The genes RHD and RHCE code for the formation of RhD (CD240D) and RhCcEe (CD240CE), two highly hydrophobic, non-glycosylated proteins that cross the erythrocyte membrane 12 times(1-3).

The first described antigen of the Rh system is D which is composed of several epitopes. This means that there are several individual parts of the antigen, and against each of them an antibody can be formed (2,3).

Before the introduction of RhD immunoprophylaxis, anti-D antibody was a common cause of severe forms of hemolytic disease of the fetus and newborn (HBFN). Although most people are either RhD-positive or RhD-negative, there are also variants of antigen D that are manifested in the form of its weak (weak D) or partial (D partial) expression. The D- (neg.) phenotype occurs as a consequence of the absence of the Rh protein from the erythrocyte membrane. In Caucasians, the D- (neg.) phenotype is most often the result of homozygosity for the deletion of the RHD gene, while in D- (neg.) Africans, an inactive RHD gene is a common finding (2, 3).

D weak antigen is often associated with single missense mutations in the RHD gene that encode amino acid substitutions in the cytoplasmic or membrane part of the D protein, while partial D antigen is usually caused by changes in extracellular loops. Partial antigen D is characterized by gene conversion, point mutations such as missense mutations in the extracellular part of the protein, as well as multiple missense mutations spread from one end of the RhD protein to the other (4-5).

The terms "weak" and "partial" D are still in use to help bring some order to the large number of aberrant D antigens, but there are many valid arguments for replacing these terms with one—the "variant" D antigen (4-6).

Many different mutations cause the emergence of a large number of phenotypes that are today classified from type 1 to type 66, although there are also working versions that have not yet received

their number. The most common is type 1, which together with types 2 and 3 represents about 90% of D weak phenotypes proven in persons of European origin (7-9).

Erythrocytes with partial antigen D were given this name in the past due to the fact that these were people whose erythrocytes showed a strong agglutination reaction with anti-D test serum, and after immunization with erythrocytes with normal D, they created anti-D antibody. In individuals with partial D antigen, the altered or missing part of the RHD gene is replaced by the corresponding parts of the RHCE gene. The newly formed protein is so different from the normal D antigen that anti-D antibodies can be formed (10, 5).

The presence or absence of anti-D antibodies cannot differentiate between weak and partial D antigen when immunohematological techniques are applied (11). The most common forms of partial antigen D in Europe are DNB (12), DVI (13) and DVII (14). In the United States, partial forms of the D antigen are most commonly found in people of African descent.

The RhD antigen is determined at each blood donation. The last edition of the Recommendation for the preparation, use and quality assurance of the components of the Council of Europe implies the determination of RhD antigen in donors in duplicate, using monoclonal reagents of different clones or human sera of different series. It is advised to perform an indirect antiglobulin test in order to prove the weakly expressed antigen D. In addition, the reagent used in the subject must recognize the most important variants of the antigen D as RhD-positive (15).

For the first administration, the Standards recommend the mandatory use of two techniques, one of which must be automated. In the absence of a positive finding in the initial testing, the Standards provide that the examination of antigen D variants should be continued with the indirect antiglobulin test technique (16).

The International Forum for Demonstration of Less Expressed Antigen D recommended the routine use of an indirect antiglobulin test in all D-negative donors, especially those with antigen C or E in their phenotype. The frequency of blood donors whose antigen D has been proven only in the indirect antiglobulin test, according to the data of this forum, ranges from 0.01% in Spain to 4.1% in Denmark (17-20).

Aim

The aim of our research was to determine the serological prevalence of D weak in voluntary blood donors at our institution, as well as to analyze the Rh phenotype in such typed erythrocytes using serological methods, in order to enable the development of individualized transfusion strategies.

Materials and methods

This retrospective cross-sectional study was conducted at the Institute for Blood Transfusion in Nis, Serbia, after obtaining approval from the Ethics Committee of the Institute for Blood Transfusion in Nis (No. 471, dated August 29, 2025). The study included voluntary blood donors from January 1 to December 31, 2024. A total of 45,970 voluntary blood donors were included in the study. Donors of both sexes, aged between 18 and 65 years, were included.

Venous blood samples with ethylenediaminetetraacetic acid (EDTA) were collected from blood donors in the Republic of Serbia. To determine the blood group in the ABO system and RhD typing, a fully automated system for immunohematological tests of blood groups using the microtiter plate method (Galileo; Immucor Inc., Norcross, GA, USA) was used in accordance with the manufacturer's instructions. We used anti-D test reagents (Immuclone Anti-D monoclonal, human IgM clone: ROM-1 and Immuclone Anti-D blend monoclonal human IgM + IgG clones: MS-26, TH-28). The presence of agglutination tells us about the presence of D-antigen on the tested erythrocytes. The absence of agglutination requires the performance of an indirect antiglobulin test to determine the presence of the weak (D weak) variant of the D-antigen (formerly called Du). Some monoclonal anti-D reagents can give a weakly positive reaction with D weak positive erythrocytes without performing an IAT. We can declare Rh D weak positive only if the direct anti human globulin test (DAT) with the erythrocytes tested is negative. This means that DAT must be performed for samples that are D weak positive. In donors whose D weak antigen has been confirmed, the Rh phenotype must also be determined using monoclonal test serums anti-C (seraclone® Clone Ms24), anti-c (seraclone® Clone Ms33), anti-E (seraclone® Clone Ms258/906) and anti-e (seraclone® Ms16/Ms21/Ms63).

Of the 134 confirmed D weak antigens, two samples were sent for genetic analysis to determine the D weak antigen type.

Results

A total of 45,970 samples of voluntary blood donors were tested in the period from January 1 to December 31, 2024. Of the total number of tested samples, 38,989 (84.82%) were D-positive, 6,847 (14.89%) D-negative, while the number of D weak was 134 (0.29%). (Table 1)

Table 1. Prevalence of D weak antigen in the studied population

RhD status	RhD-positive	RhD-negative	RhDweak
	38.989 (84,82%)	6.847 (14,89%)	134 (0,29%)
Total	45.970 (100%)		

The distribution of D weak phenotypes showed that the most common recorded phenotype was CcDwee in 117 subjects (87.31%). CCDwee phenotype was found in 12 subjects (8.96%), while ccDwee phenotype was detected in 3 (2.24%) and ccDwEe phenotype in only 2 samples (1.49%).

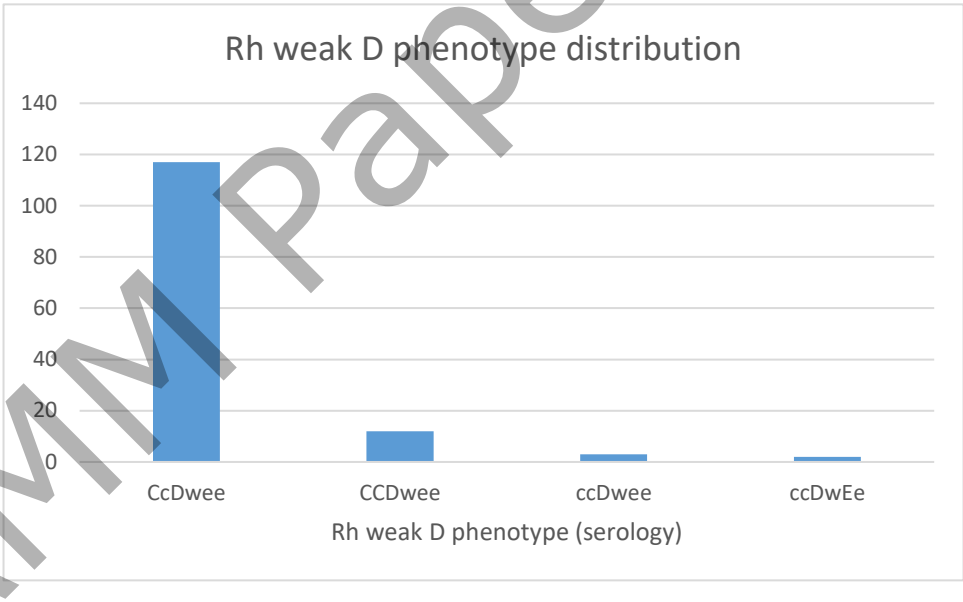

Figure 1. Distribution of D weak phenotypes

The sent samples were genotyped with the RHD microchip (RHD BeadChip, Immucor BioArray Solutions, Warren, NJ). Both D weak samples were typed as Weak D type 11.

Discussion

In this study, the prevalence of serological D weak in the region of Southeast Serbia was 0.29%, which corresponds to the results described in the literature (21). The distribution of the Rh phenotype is the same as in the European population (4).

The prevalence of RhD negative phenotype among participants in this study was 14.89%; which is similar to the findings of other studies in European and North American populations, with prevalences of the RhD negative phenotype ranging from 15 to 17.3% (22, 23).

This prevalence is much higher than that found in people of Asian descent, for whom a figure of 1% or less has been reported, (13) and higher than the findings in Nigeria and many African countries that have reported prevalence rates ranging from 2.0 to 5.1% (4, 24-28).

In India, the prevalence of D weak is estimated to range from approximately 0.0075 to 0.2% of the total donor population from different geographical areas (29). Differences in RhD typing have been attributed to multiple factors, such as different methods (tube, gel, microplate) used for typing, different stages of testing (saline or Coombs phase), different avidity and specificity of monoclonal antisera, and diversified RhD alleles with different phenotypic expression (30).

It is important to highlight the fact that the distribution and diversity of RhD variants differ depending on the population, race and geographical location. Most of what is known about RhD variants comes from studies focusing either on populations of European descent or on patients with sickle cell anemia (31-33). It has recently been suggested that D weak types 1, 2, and 3 represent less than 30 percent of the serologic D weak phenotypes identified among Brazilians, who are intensely racially admixed (34). Relatively high frequencies of D weak type 38 and D weak type 11 have also been shown in this population (34, 35), which is an interesting finding, as these variants are rare among whites and have not been reported among people of African descent.

Weak antigen D proven by serological techniques or serologically weak phenotype D, is defined as the form that does not give a reaction with the anti-D reagent in direct agglutination or gives a weakly

positive reaction, ≤2+, while the reaction with the antihuman globulin reagent is moderate or strong (36-39). Many forms of serologically weak antigen D arise as a result of one or more amino acid substitutions in the part of the RhD protein that is inside or under the erythrocyte membrane, which leads to reduced expression of the antigen on the erythrocyte surface (39). The prevalence of serologically proven weak D phenotype varies among races and nations (4). It is estimated that 0.2% to 1.0% of Caucasians inherit RHD genes that code for the emergence of a serologically weak D phenotype. In most cases, these are weak D types 1, 2, and 3 (4, 40, 41, 38). These forms of the weak D antigen are the most frequently demonstrated D variants in Europe and the United States. Reports on the prevalence of the serologically weak D phenotype vary depending on the testing method used (manual tube method vs. automated analyzer), the anti-D reagent used (polyspecific serum vs. monoclonal blend), and the use of a reaction enhancer (bromelain) (38). The first results of molecular testing of blood donors show that weak D type 3 is predominant in the Republic of Srpska, while weak D type 1 is the most common in Serbia (42).

Most data on RHD alleles and risk of alloimmunization in serologically weak D phenotypes derive from observational studies conducted in central Europe (4, 42-44, 32). These studies indicate that blood recipients who have weak D type 1, 2, or 3, in homozygous or heterozygous form, are not at risk of developing anti-D antibody after administration of RhD-positive erythrocytes that have normal D expression (44, 45). About 95% of Caucasians in Central Europe who are serologically found to have weak D antigen have weak D type 1, 2 or 3. They are treated as RhD-positive and can receive a transfusion of RhD-positive blood. The absence of anti-D antibodies in persons with the mentioned phenotypes seems to be due to the fact that the different RHD allele encodes the generation of all RhD antigen epitopes in these persons compared to persons with normal D antigen expression, although the antigen density on the surface of erythrocytes with weak D type 1, 2 or 3 is less than in those with normal D (4). D antigen alloimmunization and anti-D antibody have been demonstrated in some other types of weak D antigen, such as weak D type 4.2 (44, 45, 33), DAR (4), type 11 (44, 32), type 15 (44, 32), type 21 (46), and type 57 (14).

The American Association of Blood Banks and the College of American Pathologists formed a Interorganizational Work Group on RHD Genotyping that reviewed published and unpublished reports to determine whether weak D types 4.0 and 4.1 can receive RhD-positive blood without the

consequences of alloimmunization. There are no published papers demonstrating allo- or autoanti-D antibodies in individuals with weak D type 4.1 in large studies conducted in Europe (44, 45) despite their frequency (32, 47) and frequent transfusion treatment with RhD-positive erythrocytes (47). The working group analyzed published data on people who had weak type D 4.0 and developed anti-D antibodies. Of which three were described in Germany (44), nine in France (45), one in Tunisia (48) and three in the United States of America (48, 49). Of the 16 cases described in Europe, only one patient developed alloanti-D antibody (45), while the remaining 15 developed autoanti-D. Because of all of the above, the Working Group limited the recommendations for the use of RhD-positive blood exclusively to people with weak D types 1, 2 and 3, until more data are collected. However, the latest results of an international study by Flegel et al., published in 2019, bring new data, supplement and extend the recommendations, after studying the Tunisian population, which turned out to have the highest prevalence of the weak D type 4.0 allele in the world (50). The evidence for the existence of allo- and autoanti-D in patients with certain forms of weak D was reviewed in detail in 2015, and since then no new evidence of their detection has been reported either in people with weak D type 4.0 or in people with weak D type 4.1. in accordance with the absence of additional data on unwanted clinical effects, the conclusion was published in the last study that patients with serologically weak weak D phenotype should be tested by molecular methods, in order to prove weak D types 4.0 and 4.1. The strategy of using RhD+ blood should be based only on molecular testing. Therefore, it is recommended that patients and pregnant women with weak D phenotypes 1, 2, 3, 4.0 and 4.1 be treated as RhD+ and not be administered RhDIg, because according to all available data, it has not been proven that they could have any clinical benefits from RhD immunoprophylaxis (22).

Conclusion

In conclusion, the prevalence of D weak variants in Southeast Serbia is similar compared to the frequencies previously described for other European populations.

Given the genetic diversity of D weak variants among ethnic groups, comprehensive sequencing strategies are essential for safe transfusion practices. Clinicians should integrate molecular testing into routine RhD assessment to minimize the risks of alloimmunization and ensure accurate patient

care decisions. Understanding the molecular basis of D weak phenotypes is necessary to optimize transfusion strategies and obstetric management.

Serological tests rely on immunohematology, which can fail to clearly identify RhD variants like partial D, DEL, or weak D type 11. Consequently, RHD molecular typing is recommended to accurately detect and confirm these variants.

References

1. Knight R. Fundamentals of Biomedical Science. Transfusion and Transplantation Science. Transfusion Science Consultant. Oxford, University Press, 2013.

2. Daniels G. Human Blood Groups, 3th ed. Wiley Blackwell, 2013.

3. Harmeing DM. Modern Bloode Banking & Transfusion practices, 5th ed. United States of America, 2012.

4. Daniels G. Rh blood group system. In: Daniels G. Human Blood Groups. 2nd ed. Oxford: Blackwell Science, 200: 195-274.

5. Westhoff CM. Review: The Rh blood group antigen. Dominant, diverse and difficult. Immunohematology 2005; 21: 155-63.

6. Klein HG, Anstee DJ. The Rh blood group system (and LW). In: Klein HG, Anstee DJ. Mollison's Blood Transfusion in Clinical Medicine. 11th ed. Blackwell Publishing; 2007: 163-208.

7. Flegel WA, Wagner ff. Molecular Biology of partial D and weak D: Implications for Blood Bank Practice. Clin Lab 2002; 48: 53-9.

8. Wagner FF, Flegel WA. The Rhesus site. Available from: http://www.uniulm.de/~fwgner/RH/RB/weakD.htm.

9. Mota M, Fonseca N, Rodriguez A, Kutner JM, Castillo L. Anti-D alloimmunisation by weak D type 1 red blood cells with a very low antigen density. VoxSang 2005; 88: 130-5.

10. Westhoff CM. The Rh system. In: Roback JD. Combs ME, Grossman BJ, Hillyer CD. Technical manual, 16th ed, AABB, Bethesda, Maryland, 2008.

11. Le Maréchal C, Guerry C, Benech C et al. Identification of 12 novel RHD alleles in western France by denaturing highperformance liquid chromatography analysis. Transfusion 2007; 47: 858-63.

12. Yazer MH, Triulzi DJ. Detection of anti-D in D+ recipients transfused with D' red blood cells. Transfusion 2007; 47: 2197-201.

13. Wagner FF, Eicher NI, Jorgensen JR et al. DNB: a partial D with anti-D frequent in Central Europe. Blood 2002; 100: 2253-6.

14. Wagner FF, Kasulke D, Kerowgan M et al. Frequencies of the blood groups ABO, Rhesus, D category VI, Kell, and of clinically relevant high-frequency antigens in South-Western Germany. Infusionsther Transfus Med 1995; 22: 285-90.

15. Council of Europe. Guide to the Preparation, use and quality assurance of blood components. Recommendation no.r (95)15. 14th ed. European Directorate for the Quality of Medicines and HealthCare of the Council of Europe, 2009.

16. The Government of the Republic of Serbia. Ministry of Health. National standards. Safe blood for all in the third millennium. Belgrade, May 2006.

17. Gassner C, Doescher A, Drnovsek TD, Rozman P, Eicher NI, Legler TJ, Lukin S, Garritsen H, Kleinrath T, Eggaer B, Ehling R, Kormoczi GF, Kilga-Nogier S, Schoenitzer D, Petershofen EK. Pressence of RHD in serologicaly D-, C/E+ individuals: a European multicentric study. Transfusion 2005; 445: 527-38.

18. Wagner FF, Frohmajer A and Flegel WA. RHD positive haplotypes in D negative Europeans. BMC Genetics (2001) 2:10. Available from: http://www.biomedcentral.com/1471-2156/2/10.

19. Flegel WA. Blood group genotyping in Germany. Transfusion 2007; 47 (1 Suppl):47S-53S.

20. Avent D, Martiney A, Flegel W, Olsson M, Nogues N, Pisacka M, Scott M, Daniels G, de Haas M. The Bloodgen project: Toward mass scale genotyping for human blood group antigens. Vox Sang 2006; 91: (suppl 3).

21. Sandler, S.G.; Queenan, J.T. A guide to terminology for Rh immunoprophylaxis. Obstet Gynecol 2017; 130: 633–5.

22. Garratty G, Glynn SA, McEntire R. ABO and Rh(D) phenotype frequencies of different racial/ethnic groups in the United States. Transfusion 2004; 44(5):703–6.

23. Lehmann H. The distribution of the Human Blood Groups and other Polymorphisms. J Biosoc Sci. 1977;9:371–3.

24. Anifowoshe AT, Owolodun OA, Akinseye KM, Iyiola OA, Oyeyemi BF. Gene frequencies of ABO and Rh blood groups in Nigeria: a review. Egypti J Med Hum Genet. 2017; 18(3): 205–10.

25. Bergström S, Pereira C, Hagstrom U, Safwenberg J. Obstetric implications of rhesus antigen distribution in Mozambican and Swedish women. Gynecol Obs Invest. 1994; 38(2): 82–6.

26. Apecu RO, Mulogo EM, Bagenda F, Byamungu A. ABO and Rhesus (D) blood group distribution among blood donors in rural south western Uganda: a retrospective study. BMC Res Notes. 2016; 9(1): 1–4.

27. Mukhtar IG, Abdulkadir AY. Frequencies of ABO and Rhesus (D) blood group phenotypes among pregnant women attending antenatal clinic at Murtala Muhammad Specialist Hospital, Kano, Nigeria. J Med Trop 2019; 21(1): 31–6.

28. Mukhtar IG, Muhammad SM, Yakasai BW, Salisu IA. ABO and Rh blood group phenotypes of Nigerian blood donors at Murtala Muhammad specialist hospital, Kano—a one-year retrospective study. Bayero J Med Lab Sci 2018; 3(1): 48–53.

29. Gupta A, Mirza S, Khurana S, Singh R, Chaturvedi S, Singh B. Enigmatic weak D antigen: an experience in a Tertiary Care Hospital of East Delhi. J Clin Diagn Res 2016; 10(6): EC12–5.

30. Roback JD, Grossman BJ, Harris T, Hillyer CD, editors. Technical manual – American Association of Blood Banks. 17th ed. Bethesda: AABB Press; 2011: 407.

31. Reid ME, Halter Hipsky C, Hue-Roye K, Hoppe C. Genomic analyses of RH alleles to improve transfusion therapy in patients with sickle cell disease. Blood Cells Mol Dis 2014; 52: 195–202.

32. Wagner FF, Frohmajer A, Ladewig B, et al. Weak D alleles express distinct phenotypes. Blood 2000; 95: 2699–708.

33. Noizat-Pirenne F, Tournamille C. Relevance of RH variants in transfusion of sickle cell patients. Transfus Clin Biol 2011; 18: 527–35.

34. Dezan MR, Oliveira VB, Gomes CN, et al. High frequency of variant RHD genotypes among donors and patients of mixed origin with serologic weak-D phenotype. J Clin Lab Anal 2018; 32: e22596.

35. Costa S, Martin F, Chiba A, Langhi D Jr, Chiattone C, Bordin J. RHD alleles and D antigen density among serologically D– C+ Brazilian blood donors. Transfus Med 2014; 24: 60–1.

36. Agre PC, Davies DM, Issitt PD et al. A proposal to standardize terminology for weak D antigen. Transfusion 1992; 32: 86-7.

37. Daniels G. Variants of RhD-current testing and clinical concequences. Br J Haematol 2013; 161: 461-7.

38. Jenkins CM, Johnson ST, Bellissimo DB et al. Incidence of weak D in blood donors typed as D positive by the Olympus PK 7200. Immunohematology 2005; 21: 152-4.

39. Fung MK, Grossman BJ, Hillyer CD et al. Technical manual 18th ed. Bethesda: AABB Press, 2014.

40. Müller TH, Wagner FF, Trockenbacher A et al. PCR screening for common weak D types shows different distributions in three Cental European populations. Transfusion 20001; 41: 45-52.

41. Garratty G. Do we need to be more concerned about weak D antigens? Transfusion 2005; 45: 1547-51.

42. Guzijan G, Jovanovic Srzentic S, Pavlovic Jankovic N, Djilas I, Lilić M. Implementation of Molecular *RHD* Typing at Two Blood Transfusion Institutes from Southeastern Europe. Transfus Med Hemother 2019; 46(2): 114-20.

43. Hemker MB, Ligthart PC, Berger L et al. DAR, a new RhD variant involving exons 4,5 and 7, often in linkage with ceAR, a new Rhce variant frequently found in African blacks. Blood 1999; 94: 4337-42.

44. Flegel WA. How I manage donors and patiens with a weak D phenotyte. Curr Opin Hematol 2006; 13: 476-83.

45. Pham BN, Roussel M, Peyrard T et al. Anti-D investigations in individuals expressing weak D Type 1 or weak D Type 2: allo- or autoantibodies: Transfusion 2011; 51: 2679-85.

46. McGann, Wenk RE. Alloimmunization to the D antigen by a patient with weak D type 21. Immunohematology 2010; 26: 27-9.

47. Yu X, Wagner FF, Witter B et al. Outliers in RhD membrane integration are explained by variant RH haplotypes. Transfusion 2006; 46: 1343-51.

48. Ouchari M, Chakroun T, Abdelkefi S et al. Anti-D-auto-immunization in a patient with weak D type 4.0. Transfus Clin Biol 2014; 12: 43-6.

49. Flegel WA, Denomme GA. Allo- and autoanti-D in weak D types and in partial D. Transfusion 2012; 52: 2067-9.

50. Chou ST, Jackson T, Vege S et al. High prevalence of red blood cell alloimmunization in sickle cell disease despite transfusion from Rh-matched minority donors. Blood 2013; 122: 1062-71.