CASE REPORT

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A discrepancy of blood group A with missing antigen in anorectal malformation patient at Dr. Wahidin Sudirohusodo Hospital, Makassar, Indonesia

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ABSTRACT

Background: Blood group discrepancy is a discrepancy in the results of the cell grouping with serum grouping examination. The source of discrepancy is from technical errors or a patient's medical status such as age, disease, history of blood transfusion and medication, pregnancy. This case study aims to evaluate a discrepancy of blood group A with missing antigen in anorectal malformation patient at Dr. Wahidin Sudirohusodo Hospital, Makassar, Indonesia.

Case Presentation: A male baby, 6 months 19 days old, was unable to defecate since birth post-colostomy. Physical examination reveals anemia. A routine blood test showed a hemoglobin level of 8.8 g/dL. The diagnosis was high position anorectal malformation post colostomy suspect Hirschsprung Disease, with 110 cc Packed Red Cell (PRC) transfusion plan. A routine pre-transfusion procedure was performed. It was found a discrepancy of blood group A with missing antigen. Crossmatching with blood groups O and A both gave compatible results. Examination of peripheral blood smear after PRC blood group A transfusion found no signs of hemolytic.

Conclusion: Blood group discrepancy, in this case, was due to missing antigen, which was caused by the possibility of gastrointestinal bacterial infection.

Keywords: Discrepancy, Gastrointestinal Bacterial Infection, Missing Antigen.
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INTRODUCTION

The ABO and Rhesus system blood group examination (D typing) and a matched cross-test are minimal pre-transfusion tests that must be carried out according to the recommendations of the World Health Organization (WHO).[1] Blood group examination is a laboratory procedure performed to determine the type of blood group. Blood group examination was carried out both on the donor and the patient.[2] One of the problems in blood type examination that is often encountered is ABO blood group discrepancies. ABO blood group discrepancy is the occurrence of discrepancies or discrepancies in the results of the examination of cell grouping (forward grouping) with serum grouping (reverse grouping).[1-4]

Discrepancies need to be identified and resolved prior to cross-matching. The first step that must be taken to resolve the discrepancy problem is to identify the source of the discrepancy, whether it comes from a technical error or the patient's medical status (age, disease diagnosis, history of blood transfusion and medication, and pregnancy).[1-3]

Sources of discrepancy from technical errors are: (1) Incorrect identification and documentation that can come from mislabeling the sample and the type of tube used, incorrect recording, and incorrect interpretation of the results; (2) Errors in reagents and equipment, such as reagent quality control not appropriately done, contaminated reagents and hemolysis, incorrect centrifugation time or equipment not calibrated regularly; and (3) Errors in Standard Operating Procedure (SOP) such as procedures not in accordance with the instructions from the reagent manufacturer, adding inappropriate reagents or samples, the concentration of the red blood cell suspension is incorrect, and the cell deposits on the bottom are entirely suspended before the degree of agglutination is determined.[1,3,4]

Sources of discrepancy from problems in the sample include: (1) Problems with erythrocyte samples, namely (a) Extra antigens which can be caused by several factors, such as group A with acquired B antigen, B (A) phenotype, polyagglutination, rouleaux, hematopoietic progenitor cells, (b) Missing or weak antigen that can be caused by ABO subgroups, pathological causes, and transplant cases and (2)

Problems with serum or plasma samples, namely (a) Extra antibodies, for example in cases of A subgroups with anti-A1, cold alloantibodies, cold autoantibodies, intravenous immunoglobulin, (b) Missing or weak antibodies found in newborns, old age, pathological conditions, immunosuppressive therapy for transplantation.[1,3,4] Babies less than 4 months old have not been able to produce antibodies properly. Antibodies detected in the circulation are generally derived from maternal antibodies. At the age of 4-6 months, babies begin to be able to produce antibodies well, (c) Mixed-field reactions which can be caused by transfusion conditions in patients with blood type O with blood types A, B, AB, Hematopoietic progenitor stem cell transplants, and A3 phenotype.[1,3,4]

Based on those mentioned above, this case study aims to evaluate a discrepancy of blood group A with missing antigen in anorectal malformation patients at Dr. Wahidin Sudirohusodo Hospital, Makassar, Indonesia.

CASE REPORT

The patient is a male, aged 6 months 19 days, with a history of being born at the South Konawe Health Center with aterm condition and assisted by a midwife. Babies are born crying immediately. The baby is unable to defecate since birth, has a bloated stomach, and vomits green. The colostomy was performed on the patient at the age of 3 days at the Kendari Hospital. History of pregnancy with the mother for routine antenatal care control, not taking certain drugs during pregnancy. The patient is moderately ill, composmentis, malnourished, body weight 7.5 kg, pulse 122 times per minute, breathing 29 times per minute, temperature 37.9°C. Physical examination revealed an anemic impression. Other examinations were within normal limits. The patient was referred from Kendari Hospital to Dr. Wahidin Sudirohusodo Hospital Makassar for further examination.

Routine blood laboratory examinations and hemostasis obtained results, as shown in Tables 1 and 2. Assessment of blood chemistry and electrolytes was carried out with the results in Table 3. The COVID-19 antibody test was carried out with the results in Table 4.

Table 1. Complete Blood Counts Examination.

Parameter	15/02/2021	24/02/2021	Units	Reference
WBC	8.8	10.9	10^3/µL	6.0-18.00
%Neutrophil	21.2	20.6	%	20-40
%Lymphocyte	67.3	66.7	%	48-78
%Monocyte	6.8	9.2	%	2-11
%Eosinophil	4.4	2.5	%	1-4
%Basophil	0.3	1.0	%	0-2
RBC	3.76	4.62	10^6/µL	3.60-5.20
HGB	8.8	11.3	g/dL	10.4-15.6
HCT	27	35	%	35-51
MCV	71	76	fL	78-102
MCH	23	24	pg	23-31
MCHC	33	32	g/dL	32-36
RDW-CV	13.5	14.2	%	11.5-14.5
PLT	337	446	10^3/µL	150-450
MPV	9.5	7.9	fL	6.50-11.0
PCT	0.00	0.35	%	0.15-0.50
PDW	9.3	12.8	%	10.0-18.0

WBC = White Blood Cell; RBC = Red Blood Cell; HGB = Hemoglobin; HCT = Hematocrit; MCV = Mean Corpuscular Volume; MCH = Mean Corpuscular Hemoglobin; MCHC = Mean Corpuscular Hemoglobin Concentration; RDW-CV = Red Cell Distribution Width Coefficient of Variation, PLT = Platelet, MPV = Mean Platelet Volume, PCT = Procalcitonin, PDW = Platelet Distribution Width; Interpretation: Microcytic Anemia; Blue: Low; Red: High.

Table 2. Hemostasis Laboratory Examinations.

Parameter	15/02/2021	Units	Reference
Hemostasis			
PT	11.4	Seconds	10-14
INR	1.1		1.1
APTT	28.4	Seconds	22.0-30.0

PT = Prothrombin Time; INR = International Normalized Ratio; APTT = Activated Partial Thromboplastin Time; Interpretation: Normal Range.

Table 3. Results of blood chemistry and electrolytes.

Parameter	15/02/2021	Units	Reference
Blood Chemistry			
Random Blood Glucose	86	mg/dL	70-140
Urea	8	mg/dL	8-23
AST/SGOT	46	U/L	9-80
ALT/SGPT	23	U/L	5-45
Creatinine	0.30	mg/dL	0.1-0.4
Electrolytes			
Natrium	138	mmol/L	136-145
Kalium	4.4	mmol/L	3.5-5.1
Chloride	107	mmol/L	97-111

AST/SGOT=Aspartate Aminotransferase/Serum Glutamic Oxaloacetic Transaminase; ALT/SGPT=Alanine Aminotransferase/ Serum Glutamic Pyruvic Transaminase; Interpretation: Normal Range.

The primary diagnosis was Anorectal Malformation (ARM) with high post colostomy with suspected Hirschsprung disease (HD). On admission to the hospital, therapy included frequent infusions of 100 cc/24 hours, paracetamol 70 mg/8 hours intravenously, and ceftriaxone 350 mg/12 hours intravenously. On February 24th 2021, the patient was scheduled to undergo Posterior Sagittal Anorectoplasty (PSARP) surgery as well as rectum and stoma biopsy. The patient's hemoglobin (Hb) level was low, so a transfusion was

Table 4. COVID-19 antibody test results.

Antibody Test COVID-19	15/02/2021	Reference
Antibody SARS-CoV-2 IgM	Non-Reactive	Non-Reactive
Antibody SARS-CoV-2 IgG	Non-Reactive	Non-Reactive

COVID-19 = Corona Virus Disease 2019; SARS-CoV-2 IgM = Severe Acute Respiratory Syndrome Coronavirus 2 Immunoglobulin M; SARS-CoV-2 IgG = Severe Acute Respiratory Syndrome Coronavirus 2 Immunoglobulin G; Interpretation: Normal.

Table 5. Cross-matching test dated February 23rd 2021.

Variable	Major	Minor	*Auto Control*
PRC O-Blood Type	-	-	-
PRC A-Blood Type	-	-	-

PRC = Packed Red Cells; Interpretation: Compatible

Table 6. Examination of post-transfusion blood group on February 25th 2021.

Forward/cell grouping		Rhesus	Reverse/serum grouping	
Anti-A	Anti-B	D	Cell A1	Cell B
+2	-	+4	-	+3

Interpretation: Blood type-A rhesus D positive.

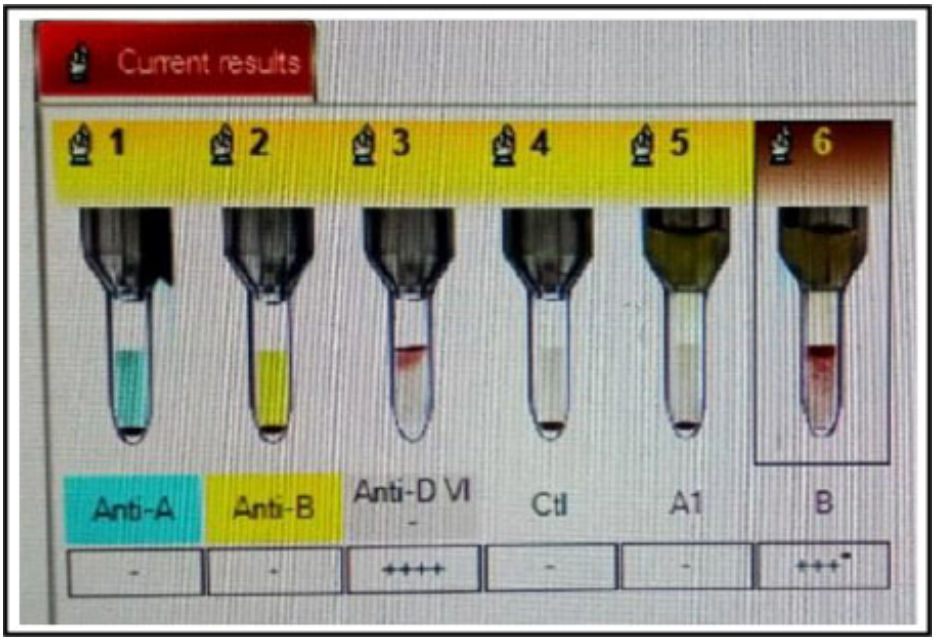

Figure 1. Blood group examination of patients using the gel method on February 23rd 2021. Impression: Discrepancy of blood group A with missing antigen. Suggestion: Compatible cross-test with blood type A and blood type O (Source: personal documentation).

performed before the procedure. On February 23th 2021, a pre-transfusion examination was carried out in the form of a blood type examination and a matched cross-test at the UTD Dr. Wahidin Sudirohusodo Hospital. Examination of the patient's blood group showed blood group A discrepancies with missing antigen-suggestions for cross-matching with blood type A and blood type O as shown in Figure 1. The cross-match test with blood groups O and A gave compatible results, as shown in Table 5.

The patient was then transfused with PRC blood group A rhesus D positive as much as 110 cc. The routine post-transfusion blood examinations on February 24th 2021, showed an increase in Hb levels from 8.8 g/dL to 11.3 g/dL. On February 25th 2021, a post-transfusion blood group examination was carried out, the results of blood type A rhesus D were positive, as shown in Table 6.

Peripheral blood smear (ADT) post-transfusion showed no hemolytic signs. The impression of hypochromic microcytic anemia with leukocytes with signs of infection is shown in Figure 2.

DISCUSSION

Anemia, in this case, can occur due to infection even though there is no bleeding. In acute infection, normochromic normocytic anemia occurs, which can then progress chronically to hypochromic microcytic anemia. In infectious/inflammatory conditions, the inflammatory mediator interleukin-6 (IL-6) will stimulate increased hepcidin expression, which inhibits iron absorption, inhibits erythropoietin production, closes ferroportin, and suppresses erythropoiesis, which can cause iron-deficiency anemia.[5] Poor nutritional status can also cause iron-deficiency anemia.[6]

This patient's blood transfusion was performed during the preoperative period, namely before the Posterior Sagittal Anorectoplasty (PSARP) operation and rectal and stoma biopsy, with a Hb of 8.8 g/dL. The routine pre-transfusion procedure in this patient was an ABO and Rhesus blood group examination as well as a matched cross test. Blood type discrepancy occurs where the impression of blood group O is obtained according to forward grouping and the impression of blood group A is according to reverse grouping.

The source of discrepancy can come from technical errors or can also come from the patient's medical status (age, disease diagnosis, history of blood transfusion and medication, and pregnancy). Sources of discrepancy from technical errors in blood type-examination, in this case, have been removed, namely identification is appropriate, samples have been checked, reagents have been validated, and equipment has been calibrated. Blood group examination has been carried out according to SOPs. The

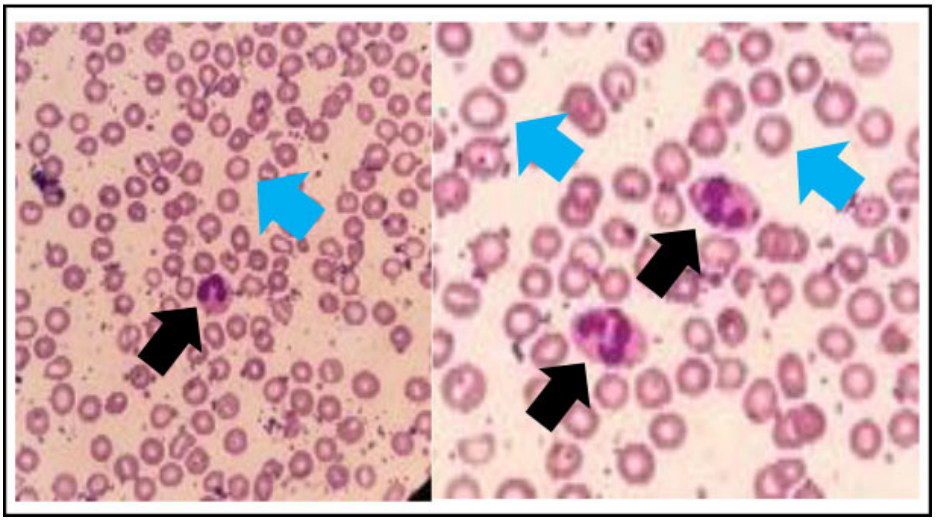

Figure 2. Overview of post-transfusion patient ADT on February 25th, 2021. Impression: Hypochromic microcytic anemia with leukocytes with signs of infection (Source: personal documentation).

source of discrepancy from problems in plasma samples in the form of missing or weak antibodies has also been ruled out because the patient is more than 6 months old, has been able to produce antibodies properly so that antibodies have been detected in plasma and the diagnosis of Anorectal Malformations (ARM) disease is high post-colostomy with suspected HD in this patient. Unrelated to plasma abnormalities.

The source of discrepancy from the problem in the erythrocyte sample in the form of missing antigen cannot be ruled out because patients with high ARM disease post-colostomy suspected of Hirschsprung Disease (HD) have the possibility of gastrointestinal bacterial infection that can cause missing antigen.[7] ARM are congenital abnormalities due to failure of normal growth of the rectum or anus.[8] At the same time, HD is a disease of the digestive system, especially the large intestine (colon), enlargement of the large intestine (megacolon) and congenital intestinal motility disorders due to the absence of ganglion cells in the distal intestine.[9] They were acquired in neonates and even children, leading to distal bowel obstruction.[8,9] The state of ARM disease accompanied by HD in this patient resulted in faecal stasis, which led to bacterial growth resulting in gastrointestinal infection.

Research conducted by Tofft L et al., showed that the bacterial culture test on surgical wound complications of ARM patients found growth of bacteria species *Enterococcus, E. coli,* and *Staphylococcus*.[10] Bacterial culture test was not carried out because the patient had been discharged from the hospital treatment room, so it can only be concluded that the possibility of surgical wound complications in this ARM patient was due to bacterial infection. Certain gram-negative bacteria such as *Escherichia* and *Proteus* that infect the gastrointestinal tract produce substances resembling A and B antigens that bind to antisera so that the patient's antigen is undetectable.[11] It is possible that these bacterial enzymes convert antigens through the deacetylation pathway resulting in missing antigens.[12] There are also differences in the expression of A and B antigens in neonates compared to children and adults. A and B antigens are weakly expressed in neonates.[13]

The blood type must be determined before further cross-matching is carried out. This patient's blood group discrepancy was concluded not to be a blood type O discrepancy with missing antibodies because the patient was more than 6 months old, was able to produce antibodies so that antibodies had been detected in plasma, and the diagnosis of ARM disease with high post colostomy suspected HD in this patient had no relationship. with plasma abnormalities. It is concluded that the discrepancy of blood group A with missing antigen is because antibody B is almost completely formed (+3), the possibility of gastrointestinal bacterial infection that produces substances resembling antigens A and B that bind antisera so that the patient's antigen is not detected, and the possibility of bacterial enzymes converting antigens resulting in missing antigens. After the patient's blood group has been determined, a cross-match test is then performed. Although the cross-match test with blood types O and A gave compatible results, blood type A transfusion was chosen because a more specific blood group was selected according to the patient's blood group, which had been determined before the match cross-test was conducted. The positive rhesus D blood type PRC transfusion, in this case, was concluded to be in accordance with the patient's blood type. Seen from the impression of post-transfusion ADT, no hemolytic signs were found because if it is not appropriate, intravascular hemolysis will occur. There was an increase in post-transfusion Hb from 8.8 g/dL Hb to 11.3 g/dL.

CONCLUSION

In this case, the blood group discrepancy is due to the missing antigen caused by the possibility of gastrointestinal bacterial infection.

CONFLICT OF INTEREST

There are no conflicts of interest regarding this case study.

ETHICS CONSIDERATION

This case study has followed COPE and ICMJE guidelines based on the publication ethics protocols as well as received informed consent from the family prior to the study being conducted.

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AUTHOR CONTRIBUTIONS

Yohanes Kusumo Adi Arji Atmanto wrote the manuscript. Raehana Samad, Rachmawati Muhiddin, and Mansyur Arif were as Supervisor.

REFERENCES

1. Mitra R, Mishra N, Rath GP. Blood groups systems. Indian J Anaesth. 2014;58(5):524-528.
2. Mulyantari NK, Prabawa IPY. Analysis of Soluble Cluster of Differentiation 40 Ligand (SD40L) levels between thrombocyte apheresis and thrombocyte whole blood products in Sanglah General Hospital blood bank. Bali Medical Journal. 2018;8(1):173-178.
3. Khan MN, Khan TA, Ahmed Z. Discrepancy in ABO blood grouping. J Coll Physicians Surg Pak. 2013;23(8):590-592.
4. Makroo RN, Kakkar B, Agrawal S, Chowdhry M, Prakash B, Karna P. Retrospective analysis of forward and reverse ABO typing discrepancies among patients and blood donors in a tertiary care hospital. Transfus Med. 2019;29(2):103-109.
5. Cherian S, Forbes DA, Cook AG, Sanfilippo FM, Kemna EH, Swinkels DW, et al. An insight into the relationships between hepcidin, anemia, infections and inflammatory cytokines in pediatric refugees: a cross-sectional study. PLoS One. 2008;3(12):e4030.
6. Kabir Y, Shahjalal HM, Saleh F, Obaid W. Dietary pattern, nutritional status, anaemia and anaemia-related knowledge in urban adolescent college girls of Bangladesh. J Pak Med Assoc. 2010;60(8):633-638.
7. Frykman PK, Short SS. Hirschsprung-associated enterocolitis: prevention and therapy. Semin Pediatr Surg. 2012;21(4):328-335.
8. Wood RJ, Levitt MA. Anorectal Malformations. Clin Colon Rectal Surg. 2018;31(2):61-70.
9. Kenny SE, Tam PK, Garcia-Barcelo M. Hirschsprung's disease. Semin Pediatr Surg. 2010;19(3):194-200.
10. Tofft L, Salö M, Arnbjörnsson E, Stenström P. Wound Dehiscence after Posterior Sagittal Anorectoplasty in Children with Anorectal Malformations. Biomed Res Int. 2018;2018:2930783.
11. Berman HJ, Smarto J, Issitt CH, Issitt PD, Marsh WL, Jensen L. Tn-activation with acquired A-like antigen. Transfusion. 1972;12(1):35-45.
12. Rahfeld P, Withers SG. Toward universal donor blood: Enzymatic conversion of A and B to O type. J Biol Chem. 2020;295(2):325-334.
13. Jacob EA. Hematological Differences in Newborn and Aging: A Review Study. Hematology & Transfusion International Journal. 2016;3(3):178-190.