Original Article

Subgroups of A in Thai Blood Donors

Pimol Chiewsilp, Chayaporn Pinyopornpanich, Sarika Makechay, Jintana Tubrod, Udom Tingtoy and Sineenart Oota

National Blood Centre, Thai Red Cross Society

Abstract:

The frequency of subgroups of A was found to be varied among different populations. The occurrence of anti-A1 in some cases may lead to the problem in blood transfusion, if they were clinically significant. ***Objective:*** *This study aimed to reveal the frequency of A subgroups in Thai blood donors. The occurrence and reactivity pattern of anti-A1 in A_2, A_2B and other A subgroup individuals were also studied.* ***Materials and Methods:*** *A total of 13,028 group A and 4,563 group AB were identified during July 10, 2017 to August 5, 2017. The techniques used in this study included standard tube technique for agglutination and indirect antiglobulin test.* ***Results:*** *Among group A, the frequencies of A_1, A_2 and A_3 were 13,000 (99.78%), 23 (0.18%) and 5 (0.04%), respectively. For group AB, the frequencies of A_1B, A_2B and A_3B were 4,483 (98.25%), 59 (1.29%) and 21 (0.46%), respectively. Approximately, 21.30%, 4.63%, 54.63% and 19.44% of total 108 A and AB subgroups were A_2, A_3, A_2B and A_3B, respectively. Anti-A1 detected at 4°C was higher in number than at room temperature for A_2 and A_2B. The most common reactivity pattern that showed discrepancy in ABO typing was for A_2B with weak anti-A1. All of anti-A1 found were of no clinical significance.* ***Conclusion:*** *The low frequency of A subgroup distribution in group A and group AB and the imbalance in A_2 and A_2B phenotype frequencies of ABO group in Thai blood donors were observed. In addition, anti-A1 in all subgroups of A in this study showed no clinical significance.*

Keywords : ● *Subgroups of A* ● *Anti-A1* ● *Non-A1* ● *Imbalance of A_2 and A_2B phenotypes*

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Correspondence should be addressed to Prof. Pimol Chiewsilp, MD, National Blood Centre, Thai Red Cross Society, Pathumwan, Bangkok 10330 e-mail: pimol_c@yahoo.com

นิพนธ์ต้นฉบับ

หมู่ย่อยของหมู่โลหิต A ในผู้บริจาคโลหิตไทย

พิมล เชี่ยวศิลป์ ชญาภรณ์ ภิญโญพรพาณิชย์ สาริกา เมฆฉาย จินตนา ทับรอด อุดม ติ่งต้อย และ สิณีนาฏ อุทา
ศูนย์บริการโลหิตแห่งชาติ สภากาชาดไทย

บทคัดย่อ

ความถี่ของหมู่เลือดย่อย A ของหมู่ A และ AB มีความหลากหลายในประชากรต่างๆ และที่สำคัญคือ การพบว่า ซีรัมของคนที่มีหมู่เลือดย่อยบางรายมี anti-A1 ซึ่งหากเป็นชนิดที่มีความสำคัญทางคลินิก อาจทำให้เกิดปัญหาในการรับโลหิตได้ ***วัตถุประสงค์*** *เพื่อศึกษาความถี่ของหมู่ย่อยของ A ในผู้บริจาคโลหิตคนไทย รวมทั้งศึกษาปฏิกิริยาของ anti-A1 ในคนหมู่ย่อย A_2, A_2B และหมู่ย่อยชนิดอื่นๆ* ***วัสดุและวิธีการ*** *ทำการศึกษาระหว่างวันที่ 10 กรกฎาคม ถึง 5 สิงหาคม พ.ศ. 2560 ได้ตรวจตัวอย่างโลหิตหมู่ A จำนวน 13,028 ตัวอย่าง และหมู่ AB จำนวน 4,563 ตัวอย่าง โดยทดสอบปฏิกิริยาการจับกลุ่ม และ indirect antiglobulin test ด้วยวิธีหลอดทดลอง* ***ผลการศึกษา*** *ในกลุ่มหมู่ A ความถี่ของ A_1, A_2 และ A_3 เท่ากับ 13,000 (99.78%), 23 (0.18%) และ 5 (0.04%) ตามลำดับ ส่วนหมู่ AB, มีความถี่ของ A_1B, A_2B และ A_3B เท่ากับ 4,483 (98.25%), 59 (1.29%) และ 21 (0.46%) ตามลำดับ anti-A1 ที่ตรวจได้ที่อุณหภูมิ 4°C มีจำนวนเพิ่มขึ้นจากที่ตรวจได้ที่อุณหภูมิห้อง สำหรับ A_2 และ A_2B ส่วนปฏิกิริยาที่พบบ่อยที่ทำให้มีความไม่สอดคล้องระหว่างเซลล์และซีรัมของหมู่โลหิต ABO คือ กลุ่ม A_2B ที่มี anti-A1 สำหรับ anti-A1 ที่พบทั้งหมดในการศึกษานี้ ทำปฏิกิริยาอย่างอ่อนและเป็นชนิดที่ทำปฏิกิริยาได้ดีในอุณหภูมิต่ำ ซึ่งนับได้ว่าไม่มีความสำคัญทางคลินิก* ***สรุป*** *ความถี่ของหมู่ย่อยของ A ในหมู่ A และ AB พบได้น้อยมาก และพบความไม่สมดุลของ A_2 และ A_2B ของหมู่เลือด ABO นอกจากนี้ยังพบว่า anti-A1 ที่ตรวจได้ในหมู่ย่อยทั้งหมดของ A ในการศึกษานี้ ไม่มีความสำคัญทางคลินิก*

คำสำคัญ : ● *Subgroups of A* ● *Anti-A1* ● *Non-A1* ● *Imbalance of A_2 and A_2B phenotypes*
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Introduction

The distribution of ABO blood group was observed to be varied among ethnic groups.[1-3] Besides the variation in the distribution of A subgroup was also recognized.[4,5] A_1 is differentiated from A_2 on the basis of agglutination of A_1 cells but not A_2 cells with lectin anti-A1 or monoclonal anti-A1. Other subgroups of A can be defined by observing their weak reactivity with anti-A which are A_3, A_{end} and A_X, while A_m, A_y and A_{el} are not.[6] It can be further serologically differentiated by additional techniques,[7] such as agglutination strength of cells with anti-H, detection of anti-A1 in reverse grouping, adsorption elution technique with polyclonal anti-A from group B and group O individuals, and detection of H and/or A substances in saliva.[7]

The individuals with subgroup of A, as A_2, A_2B and others so called non-A_1 may possess anti-A1 in their sera. Although it usually demonstrates a low temperature amplitude and is of no clinical significance.[8] However, anti-A1 had been reported as the cause of acute hemolytic transfusion reaction[9] as well as severe delayed hemolytic transfusion reaction.[10] Then it is important to study the nature of this antibody in addition to unexpected antibody whenever subgroup A patient with anti-A1 needs blood transfusion.

Objective

The aim of this study was to reveal the frequency of subgroups of A in Thai blood donors since it has not been established. In addition, the occurrence as well as the reactivity pattern of anti-A1 in A_2, A_2B and other A subgroup individuals will also be included in the study.

Materials and Methods

During July 10 to August 5, 2017, among 61,618 blood donations at National Blood Centre, Thai Red Cross Society, Bangkok, Thailand, a total of 17,591 group A blood samples from routine blood processing laboratory were studied. This study was approved by the Human Ethics Committee of National Blood Centre, certificate number NBC 10/2017.

Blood grouping reagents (National Blood Centre, Thai Red Cross Society) used in this study were as follows:

Anti-A, Lot No. 60022; Anti-B, Lot No. 60013; Anti-AB, Lot No. 59025; Anti-A1, Lot No. 59012; Anti-H, Lot No. 60020; Anti-humanglobulin (AHG), Lot No. 60011; and A cells, B cells, A_2 cells and Pooled O cells, Lot No. 60070

Exclusion criteria: Samples with transfusion transmitted infectious reactive including HBsAg, HCV, HIV and syphilis were excluded. The study was intended to conduct at a period of one month in order to exclude the repeat donation.

Methods

A_2 subgroup was identified by agglutination technique using monoclonal anti-A1. All A_1 cells but not A_2 cells will agglutinate with anti-A1.

All negative samples which were A_2 and weakly reactive samples with anti-A1 were further retested for ABO typing by cell and serum grouping. These cells were also tested with anti-H. Since A_2 cells express more H antigen as compare to A_1 cells, so this test is useful for the differentiation of some A_2 cells from A_1 cells, in case when weak reaction with anti-A1 was observed. In addition, anti-A1 and unexpected antibody screening were performed by testing all serum samples in this set with A_1, A_2 and O cells at 4°C, room temperature (RT) at (22°-24°C), 37°C and antihuman globulin phase (AHG) phase.[11]

A_3 was identified by observing mixed-field agglutination of cells with anti-A and anti-A,B under microscope.

Results

A total of 13,028 group A and 4,563 group AB were identified in this study. Among group A, the frequencies of A_1, A_2 and A_3 were 13,000 (99.78%), 23 (0.18%) and 5 (0.04%), respectively. For group AB, the frequencies of A_1B, A_2B and A_3B were 4,483 (98.25%), 59 (1.29%)

and 21 (0.46%), respectively (Table 1). Moreover, among 108 A and AB subgroups, 21.30%, 4.63%, 54.63% and 19.44% of total 108 A and AB subgroups were A_2, A_3, A_2B and A_3B, respectively (Table 2)

Anti-A1 detected at RT for A_2, A_3, A_2B and A_3B was 1 in 23 (4.35%), 1 in 5 (20%), 4 in 59 (6.78%) and 1 in 21 (4.76%), respectively. At RT and 4°C, the occurrence of anti-A1 was 4 in 23 (17.39%) ,1 in 5 (20%), 11 in 59 (18.64%) and 1 in 21 (4.76%) for A_2, A_3, A_2B and A_3B, respectively (Table 2).

There were 2 samples of A_2B gave positive unexpected antibody screening, moderate (1+ to 3+) with A_1, A_2 and O cells at 4°C only.

Four different patterns of ABO discrepancy in 7 blood samples observed in this study were shown in Table 3. The most common pattern was for A_2B with weak anti-A1. Otherwise, there was only one of each that observed for A_2, A_3 and A_3B with anti-A1. One A_3B subgroup clearly showed mixed-field agglutination.

Table 1 Frequency of subgroups in group A and AB blood donors of National blood Centre, Thai Red Cross Society (July 10, 2017 to August 5, 2017)

ABO blood group	N (%)	Subgroup	N (%)
A	13,028 (74.06)	A_1	13,000 (99.78)
		A_2	23 (0.18)
		A_3	5 (0.04)
		Total	**13,028 (100.00)**
AB	4,563 (25.94)	A_1B	4,483 (98.25)
		A_2B	59 (1.29)
		A_3B	21 (0.46)
		Total	**4,563 (100.00)**
Total	**17,591 (100.00)**		

Table 2 Distribution of A and AB subgroups and anti-A1 detection at RT and RT -4°C

A, AB subgroups	N (%)	RT N (%)	RT- 4°C N (%)
A_2	23 (21.30)	1 (4.35)	4 (17.39)
A_3	5 (4.63)	1 (20.00)	1 (20.00)
A_2B	59 (54.63)	4 (6.78)	11 (18.64)
A_3B	21 (19.44)	1 (4.76)	1 (4.76)
Total	**108 (100.00)**	**7**	**17**

Table 3 Patterns of ABO discrepancies observed in the confirmatory test of 7 A subgroups with anti-A1

A, AB subgroup	N	Cell grouping					Serum grouping			
		Anti-A	Anti-B	Anti-AB	Anti-A1	Anti-H	A_1 cells	A_2 cells	B cells	O cells
A_2	1	4+	0	4+	0	4+	1+	0	4+	0
A_3	1	1+ ^{mf}	0	1+ ^{mf}	0	4+	1+	0	4+	0
A_2B	4	4+	4+	4+	0	2+	1+^{w}	0	0	0
A_3B	1	3+ ^{mf}	4+	3+ ^{mf}	0	2+	1+^{w}	0	0	0

mf = mixed-field agglutination

Discussion

Our results showed that A_1 and A_1B were the most common subgroups among A and AB blood groups, respectively. The frequency of A subgroups in group A and AB individuals in Thai blood donors was 0.21% and 1.75%, respectively which was low as compared to other populations.[4,5] It is worth mention that in this study, the frequency of A_2B subgroup in group AB was approximately 7 times (1.29% : 0.18%) higher than that of A_2 subgroup in group A. The similar finding was also observed in South Indian, South Sudan (African) and Japanese populations, but not in Caucasian populations.[12-15]

Subgroups of A and AB individuals may have anti-A1 in their serum. The presence of anti-A1 in A_2B was higher than in A_2 individuals in most populations including this study.[4,5] All anti-A1 detected in this study group were cold reacting and rather weak antibodies. This finding was also similar to the other reports.[4,5] Only one of A_3 and A_3B individuals possessed anti-A1. The two samples with cold reacting unexpected antibodies observed in the non-A_1 group may most likely be anti-I or anti-HI. However, no further attempt for antibody identification had been performed.

Conclusion

The significant observations in this study were the low frequency of A subgroups in Thai blood donors as compared to other populations and the imbalance in A_2 and A_2B phenotype frequencies of ABO group in Thai blood donors was also observed. In addition, anti-A1 in all A subgroups found in this study showed no clinical significance.

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