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Letter to Editor

Transfusion Medicine Section

An Unusual Case of ABO Discrepancy: Lessons Learnt

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Dear Editor,

During the pre-transfusion testing, ABO blood group typing is mainly done by agglutination. Sometimes, weak agglutination reactions may be seen with the reagent antibodies, as a result of weak expression of A and B antigens on red cell surface. These antigens may lead to significant discrepancies in ABO typing during reverse and forward grouping.

A young male came for a regular voluntary blood donation to Blood Bank, Northern Railway Hospital, New Delhi, India. The blood group known to the donor was O+. The blood group was re-performed and on forward grouping, it was O+ while on reverse grouping, reaction with only B cells noted. This suggested presence of a weaker subgroup of "A". He was referred to Blood Bank, Lady Harding Medical College for further work up. Similar results were obtained at three temperatures (4°C, 25°C and 37°C). His Direct Coombs Test (DCT) and Indirect Coombs Test (ICT) profile were negative. Anti H was strongly positive while that with anti A1 serum was negative [Table/Fig-1].

Incubation time and temperature	Reactions of donor red cells with known antisera				Reactions of donor serum against reagent red blood cells		
	A	B	AB	H	A1 cells	B cells	O cells
At 22°C	0	0	0	4+	0	3+	0
At 37°C	0	0	0	4+	0	3+	0
4°C (15 minutes)	0	0	0	4+	0	3+	0
4°C (30 minutes)	0	0	0	4+	0	3+	0

[Table/Fig-1]: Extended blood grouping.
Key:0 = No red blood cell agglutination; 1+ to 4+ = Grades of red blood cell agglutination.

Red cells were subjected to adsorption-elution studies [1]. Polyclonal O and B group antisera were adsorbed to washed test red cells. Centrifugation at 3000 g for 5 minutes was done, supernatant was discarded. Adsorbed cells were washed six times with normal saline. The last supernatant was retained separately. Elution was done. This eluate and the last supernantant were kept in parallel and the reaction was seen with A, B, O cells separately. Agglutination with A cells and no reaction with B cells suggested presence of A subgroup. Final wash solution did not show agglutination with A, B and O cells thereby validating the test.

Weaker subgroups of A include A_3, A_x, A_y, A_{end}, A_m and A_{el} [2]. These are weaker than A2 and occur very infrequently (<1%) but they can be the cause of ABO discrepancy [3]. This patient showed no reaction with Anti A, B and AB; strong positivity with Anti H and no reaction with Anti A1. This suggested the possibilities of subgroups A_y, A_m and A_{el} [Table/Fig-2].

Identification of such weaker subgroups is necessary to avoid haemolytic transfusion reactions. The subgroup typing should include a series of tests as done in the present case. Identification of a weak reaction can be further facilitated by increasing the incubation time, lowering the reaction temperature and by treatment of red cells with enzymes. Confirmatory tests for diagnosing these weak subgroups are still beyond the scope of most of the Blood banks as the antisera for these are not routinely available. However, family studies, molecular studies and in certain cases, special tests like serum glycosyl transferase estimation can be helpful tools for confirmation of these subtypes [4].

Phenotypes	Testing of red cells				Naturally occurring antibodies in serum		
	Anti A	Anti B	Anti AB	Anti H	Anti A	Anti-B	Anti-A1
A_3	mf 2+	0	mf 2+	3+	No	Common	Sometimes
A_x	Wk/0	0	2+	4+	No	Common	Always
A_{end}	Wk mf	0	wk mf	4+	No	Common	Sometimes
A_m	0/wk	0	0/+	4+	No	Common	No
A_y	0	0	0	4+	No	Common	No
A_{el}	0	0	0	4+	Sometimes	Common	Yes

[Table/Fig-2]: Serological reactions observed in subgroups A_y, A_m and A_{el}.
Key: 0 = No red blood cell agglutination; wk= Weak red blood cell agglutination; 1+ to 4+ = Red blood cell agglutination of increasing strength; mf = mixed field pattern of agglutination

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