

Prevalence of Bombay Phenotype in O Blood Group Donors and Patients in Peshawar Khyber Pakhtunkhwa

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ABSTRACT

Background: The Bombay phenotype is an uncommon blood group identified using forward and reverse plasma categorizing. Individuals with the Bombay phenotype lack the antigens A, B, and H on their blood cells, but their serum has high levels of anti-A, anti-B, and anti-H antibodies. The Bombay phenotype needed donation of the same blood as the Bombay phenotype or homologous blood. As a result, it is critical to do blood grouping appropriately.

Objective: The objective of this is to determine the prevalence of Bombay phenotype among the O blood group donors and patients in Peshawar Khyber Pakhtunkhwa

Material and methods: This research was done at Khyber Medical University Peshawar's Institute of Paramedical Science. This trial lasted six months. The samples came from Peshawar's transfusion centres and hospitals. A total of 1050 O blood donors and patients were tested. The tube technique was used to categorise forward and backward blood. Forward blood grouping used anti-sera A, B, and D, whereas reverse blood grouping used known as red blood cells from A, B, and O blood types.

Results: No cases of Bombay phenotype were detected in the entire study.

Conclusion: According to the findings of this research, the Bombay phenotype is not prevalent in Peshawar.

Keywords: Bombay blood group, O blood group donors and patients, forward and Reverse blood grouping.

INTRODUCTION

Among the present blood group systems, the main blood group systems are ABO and Rhesus(1). Immunoglobulin's present in these blood type systems have the potential to cause haemolytic responses, as well as haemolytic illness in the foetus and the newborn. These two blood group systems are well-known for their importance in the practise of blood transfusion. Furthermore, they serve an essential part in the study of various disorders, population genetic research, and the resolution of various medico-legal issues(2). The ABO blood group system was discovered by an Austrian scientist called Karl Landsteiner in the early 1900s, and it continues to serve as the foundation for blood group examinations in forensics today. They do so because they are the most basic, widely seen, and immediately distinguishable of all the groups on the planet(3). He is the representative of a legislation known as Karl Land Steiner's law states that if an antigen is present on the RBCs of an individual, the respective antibody must be absent in plasma, and if an antigen is absent on the RBCs of an individual, the correlating antibody has to be present in plasma. According to Karl Land Steiner's law, if an antigen is present on the RBCs of an individual, the matched antibody must be present in plasma(4). The ABO blood group system is divided into four primary types: A, B, AB, and O(5). A blood group person has the A antigen on his or her red blood cells and the B antibody in his or her blood

plasma. The B blood group indicates that the B antigen is present in the blood and that the A antigen is detectable in the body. In a similar vein, the AB blood type is made up of both A and B antigens, but no antibodies are present. In people with the O blood type, no antigen is present, and both A and B antibodies are present(6).

In addition to the primary forms of ABO blood groups, there is an uncommon blood group called as Bombay blood group (H/H or Oh) that is present. These individuals can only get blood transfusions from persons who have the Bombay blood group(4). Doctor Bhendi discovered it in 1952 in Bombay. Because it was identified in Bombay (Mumbai), it is known as the Bombay blood group. The lack of A, B, and H antigens on red blood cells is indicative of this unusual blood type(7,13).

The Bombay phenotype is the outcome of a point mutation in the (FUT1) gene, which is situated on chromosomal number 19 and causes the phenotypic to appear. As a result of this mutation, the person becomes deficient in H antigen and spontaneously develops anti-H antibodies. As a result, the blood group demonstrates the lack of A, B, and H antigens on red blood cells, but the plasma includes anti-A, anti-B, and anti-H antigens (A, B, and H antigens)(6). The Bombay blood group is the most uncommon blood type, with a ratio of one in ten thousand in India and one in a million in Europe, which is very low when compared to the whole population of Europe(8).

H antigen is produced by an enzyme called Fucosyl transferase. This enzyme is encoded by two genes, FUT1 (H gene) and FUT2 (Se gene). When both parents have the tiny h gene (H recessive allele) an uncommon genotype results (hh). Because H antigen is the precursor of A, B, and AB antigens, A, B, and AB antigens cannot develop in hh genotype. The Bombay blood group lacks H, A, B, and AB antigens but has all three antibodies(9).

The frequency of the Bombay blood type is at its highest in South India, and it is believed to be associated with the high incidence of consanguineous marriages in the region. When comparing rural and urban people, it is projected that consanguinity would be more in rural society(10).

Dr. Shreedevi and his colleagues conducted a descriptive research in a tertiary care hospital in Belgavi over a six-year period (2010-2016) and collected data. For the identification of the Bombay phenotype, a total of 2,74,361 blood samples were obtained and analysed. Only eleven examples of the Bombay phenotype were discovered out of a total of 2,74,361 cases (0.004 percent). Six of the eleven were patients, while the other five were donors(4).

Biplabendu and his colleagues performed a research in 2012 at the Medical College Hospital in Kolkata. Routine blood grouping tested 28934 volunteers. The Bombay phenotype had two men. Similarly, another research found 4 out of 27,531 Bombay phenotypic patients in the same year(2).

During the period 2007 to 2014, a retrospective research was conducted at the department of transfusion medicine at Sri Venkateswra Institute of Medical Sciences in Trupati, Andhra Pradesh, India. Blood grouping was performed on a total of 49,110 donor samples. There were 15 examples with Bombay phenotype (or 0.03 percent) discovered. Four of them were Rh negative, and eleven were Rh positive, for a total of fourteen(11).

From November 2010 to October 2011, a cross-sectional hospital based research at Government Medical College Jammu's transfusion medicine department. Over the course of a year, 13,281 blood units were grouped. No Bombay blood group was found(12).

Stakeholders were interviewed at the Usmanu Dan Fodiyo University Teaching Hospital in Sokoto. Blood group donors were obtained in four hundred fifty-three batches. 397 men and 53 girls The research found no Bombay phenotypic predominance(9).

In order to determine the frequency of this unusual blood type, it will be kept isolated from the O blood group in a refrigerator for the time being. It will do a cross-check before to the transfusion in order to preserve the recipient's life.

MATERIALS AND METHODS

Between March and August 2019, a prospective cross-sectional research was conducted at an Institute of Paramedical Sciences (IPMS) at Khyber Medical University (KMU) Peshawar. In this research, a convenience sampling strategy was adopted in which samples were taken from all subjects after verbal informed permission was obtained. This research included 1,050 people. The samples were gathered from a variety of Peshawar-based transfusion centres, including Lady Reading Hospital (LRH), Hayatabad Medical Complex (HMC), Khyber Teaching Hospital (KTH), Regional Blood Centre, Fatmi Foundation, and Northwest Hospital. Additionally, it is worth noting that the data collection occurred with ethics committee permission. The research excluded O blood type donors and patients who got blood from another person infected with HBS or HCV. Three millilitres of venous blood were obtained in an EDTA tube from O blood type donors and patients. The obtained samples were refrigerated between 2 and 8 degrees Celsius before further processing. The blood specimens were filtered and analysed utilising tube techniques for forward and reverse blood grouping. Anti-A and Anti-B sera were used to identify the antigen on red blood cells. Individual red blood cells are agglutinated by the proper anti-sera, suggests the existence of antigen on the red blood cells. While there is no agglutination with anti-sera indicating lack of antigen. To identify the antibody in an individual serum, reverse blood grouping was conducted using a 5% solution of A cells, B cells, and pooled O positive cells. Additionally, Rh typing was conducted using Anti-D to differentiate between the D positive and D negative groups using the tube technique. The Undergraduate Research Committee of an Institute of Paramedical Sciences of Khyber Medical University Peshawar authorised this work.

RESULTS

Of the total 1050 blood group O donors and patients including both male and female were investigated for the presence of anti-H antibody (Bombay phenotype). We observed a 0% prevalence of Bombay phenotype. Blood samples were categorized based on the O blood group donors and patients including male and female. Out the total 1050 blood samples were tested, 829(78.9%) were O blood group donors, 221(21.04%) were patients including male 133(12.6%) and female 88(8.30%). Out of 1050 blood samples 987 (94%) were Rh positive and 63 (6%) were Rh negative as shown in the table 3.1.

Table 3.1: Prevalence of Bombay Phenotype in Male, Female, Rh Positive and Negative in O Blood group Donors and Patients

	Total	Male	Female	Rh Positive	Rh Negative	Bombay Phenotype
Blood Donors	829 (78.9%)	829 (78.9%)	0 (0%)	773 (78.31%)	56 (88.8%)	0%
Patients	221 (21.04)	133 (12.6%)	88 (8.3%)	214 (21.68%)	7 (11.12%)	0%
Total	1050	962	88	987	63	0

The donors and patients were also categorized based on their age groups. Table 3.2 shows significant number of

O blood donors, 312 (37.63%) were in the 28-37 years age group, 256 (30.88%) were in the 17-27 years age group,

172 (20.74%) were in the 38-47 years age group and 89 (10.73%) were in the 48-57 years age group. Similarly, patients were also categorized based on their age groups. Table 3.2 shows most significant number of patients, 71 (20.81%) were in the 46-50 years age group, 15 (6.78%) were in the 6-15 years age group, 41 (18.55%) were in the 16-25 years age group, 27 (13.12%) were in the 26-35 years age group, 46 (20.81%) were in the 36-45 years age group and 21 (9.50%) were in the 56-65 years age group.

Table 3.2: Age wise distribution of O Blood group Donors and Patients

O Blood Group Donors	Age group	No of donors and patients
	17-27y	256 (30.88%)
	28-37y	312 (37.63%)
	38-47y	172 (20.74%)
	48-57y	89 (10.73%)
	Total	829 (100%)
O Blood Group Patients	6-15y	15 (6.78%)
	16-25y	41 (18.55%)
	26-35y	27 (13.12%)
	36-45y	46 (20.18%)
	46-55y	71 (32.12%)
	56-65y	21 (9.50%)
	Total	221 (100%)

DISCUSSION

We examined 1050 O blood donors and patients for the Bombay phenotype. We found 0% prevalence among O blood donors and patients. As in prior studies, the Bombay phenotype is uncommon, with little detection among hundreds or millions. Some studies yielded null findings for the Bombay phenotype, as we got 0%. However, prior investigations in India and the US have documented several instances (3). In Pakistan there is no cases registered till now.

It was performed in the southern region of West Bengal at several blood donation camps arranged by the medical college and the Hospital of the University of Kolkata. Only two cases of Bombay phenotypes were detected out of 28,934 donors. In the same years same institute detect 4 cases of Bombay phenotypes out of 27,531 patients. The overall prevalence of Bombay phenotype in donors and among the patient population was 0.011% (6 out of 56,465) (2). The variable in this study were patients and donors which is same to our study. But in our study, we did not find out any case of Bombay phenotype which is may be due to small sample size.

The study conducted in Andhra Pradesh state south India which showed that 26,638 blood samples was processed. Out of these, 13 (0.048%) cases were reported (10). The blood samples contained all types of ABO and RH groups. The method used in the study was forward and reverse blood typing. It was determined that the red cells were typed by utilising commercial antisera, and the serum grouping was determined by using known cells obtained from pooled blood units. The commercial Anti H lectin from ulex Europe's was used to analyse the blood samples that contained O groups. It was just O blood group donors and patients that were looked into in the present investigation. The forward reverse tube methods were used to process the blood samples. Manually produced O cells

from a blood unit were used in the laboratory for the identification of the Bombay phenotype.

Another study was done in a tertiary care hospital at Peninsula Malaysia for routine investigation of antibodies screening. Three cases of Bombay phenotype (anti H) were detected. The method used in study was commercial three panel sets (11). In the current study, we used O positive cells manually for the detection of anti H. The differences could be due to the difference in method and procedure. They employed a commercially available three-cell panel set, while the latest research used tube-based forward and reverse blood typing.

A retrospective analysis was carried out in a tertiary care teaching hospital transfusion centre in the southern Indian state of Tamil Nadu. A total of 49,110 blood donors were evaluated for blood type, with just 15 instances (0.03 percent) being discovered in the process. According to this research, the incidence is quite high when compared to previous studies conducted in Tamil Nadu (0.004 percent), Karnataka (0.005 percent), and Bengaluru (0.004 percent) (0.016 percent) (8). This research included donors of all blood types. Forward blood grouping was done using anti A, anti B, anti AB, and anti D tube agglutination. Anti-H lectin testing ruled out Bombay phenotype. Reverse blood grouping was done utilising transfusion centre A, B, and O cells. This was a prospective study. Both forward and reverse reactions were tube reactions.

The Bombay phenotypes were also detected in South East Asians countries, the cases have been reported in Japan, Thailand, Malaysia, and Srilanka. The incidence of the Bombay phenotype and the Para Bombay phenotype in Japan has been determined to be 1 to 2 in per 3000 individuals (12). The current study does not show any case of Bombay phenotype. And up till now no case observed in Pakistan.

CONCLUSION

Our study shows 0% prevalence of Bombay phenotype among the patients and O blood group donors in district Peshawar Khyber Pakhtunkhwa (KPK). O group phenotype does not respond to Anti-A and Anti-B antibodies and behaves in the same way as a normal O phenotype when exposed to these antibodies. As a result, it is necessary to do reverse blood typing in order to discover the Bombay phenotype.

REFERENCES

1. Worledge S, Ogiemudia S, Thomas C, Ikoku B, Luzzatto L. Blood group antigens and antibodies in Nigeria. *Annals of Tropical Medicine & Parasitology*. 1974;68(3):249-64.
2. Gadwalkar S, Kumar NS. Distribution of blood groups in and around Bellary, Karnataka. *Ind Jour Clin Prac*. 2013;24(3):247-50.
3. Neiders M, Standish S. Blood group determinations in forensic dentistry. *Dental clinics of North America*. 1977;21(1):99-111.
4. Talukder B, Datta SS, Mukherjee S, Mukherjee K. Prevalence of Bombay group blood in southern Bengal population. Springer; 2014.
5. Daniels G. ABO, Hh and Lewis systems. *Human Blood Groups: Blackwell Science*. 1995:7-67.
6. Nikam V, Kashid V, Khapare J, Gaikwad S. Bombay blood group: An overview. *Inventi Rapid: Pharmacy Practice*. 2017;3:1-2.

7. Race C, Watkins WM. The enzymic products of the human A and B blood group genes in the serum of "Bombay" Oh donors. *Febs Letters*. 1972;27(1):125-30.
8. Ghatak S, Bhattacharya P, Mohammed S, Gulati S. Incidental detection of Bombay blood group phenotype in a patient undergoing Whipple's pancreatoduodenectomy for chronic calcific pancreatitis with pancreatic cancer.
9. Oriol R, Danilovs J, Hawkins B. A new genetic model proposing that the Se gene is a structural gene closely linked to the H gene. *American journal of human genetics*. 1981;33(3):421.
10. Verma I, Prema A, Puri R. Health effects of consanguinity in Pondicherry. *Indian Pediatr*. 1992;29(6):685-92.
11. Kotwal U, Raina TR, Sidhu M, Dogra M. Distribution of ABO & Rh (D) blood groups among blood donors of Jammu region with respect to various ethnic groups. *Journal of Medical Thesis*. 2014;2(1):31-4.
12. Panchabhai TS, Noronha SF, Davis S, Shinde VM, Kshirsagar NA, Gogtay NJ. Evaluation of the activity of CYP2C19 in Gujrati and Marwadi subjects living in Mumbai (Bombay). *BMC clinical pharmacology*. 2006;6(1):1-5.
13. G. L. Daniels, Fletcher A, Garratty G, Henry S, Jørgensen J, Judd WJ, et al. International Society of Blood Transfusion Working Party on Terminology for Red Cell Surface Antigens. *Vox Sang*. 2004;87:304–3016.