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# DISTRIBUTION OF ABO AND RH BLOOD GROUP ALLELES IN MANDI BAHAUDDIN DISTRICT OF PUNJAB, PAKISTAN

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**Abstract:** A study was conducted on blood groups of human subjects at DHQ Hospital, Mandi Bahauddin, Province of the Punjab. The study was done from January 2005 to December 2005 0n 2542 subjects comprising 2097 (83.08%) males and 427 (16.92%) females of all ages. The objective of this study was to determine frequency of different blood groups in this District. These were categorized according to ABO/Rh system. Allele frequency was computed according to Hardy-Weinberg law. The distribution of phenotypes in the total sample was 0.4076, 0.3050, 0.2115 and 0.0756 for groups B, O, A, and AB, respectively, with 0.9140 Rh-positive (R) and 0.0860 Rh-negative(r). B group was dominant while AB was rare in both genders. The distribution of the alleles was 0.1391, 0.2639, 0.0448 and 0.5522 for IA, IB, IAB and i, respectively. The study also revealed predominance of group B, in the order of B>O>A>AB, as well as Rh-positive antigen for both male (0.8941) and female (0.9039) with 0.1059 Rh-negative for men and 0.0961 for women. It was concluded that phenotypically B group was dominant in the Mandi Bahauddin District, with high allelic frequency of O group.

***Keywords:*** Alleles, gene frequency, blood groups, Rh factor, transfusion, Mandi Bahauddin District of Pakistan

## Introduction

The discovery of almost universally present and naturally occurring antibodies in blood plasma led to the discovery of the ABO blood group system which remains, more than 100 years later, the most important and clinically significant of all blood groups [1]. The ABO system was the first genetic polymorphism defined in human beings. Since that time the blood groups have played a prominent role in the study of human polymorphisms. Because of its easy classification into different phenotypes and different frequencies in different populations, blood groups are useful genetic markers in population studies and in linkage analysis [2]. Blood is an individual's complete and unchangeable identity. Although almost 400 blood group antigens have been reported, the ABO and Rh have been recognized as the major clinically significant blood group antigens [3]. This system derives its importance from the fact that A and B are strongly antigenic and anti A and anti B occur naturally in the serum of persons lacking the corresponding antigen. These antibodies are capable of producing hemolysis *in vivo*. Rhesus blood group system was the fourth system to be discovered and yet it is the second most important blood group from the point of view of transfusion [4].

Blood group antibodies play an important role in transfusion medicine; both in relation to the practice of blood transfusion and in pregnancy, but not all are clinically significant [5]. Clinically significant antibodies are capable of causing adverse events following transfusion, ranging from mild to severe, and of causing hemolytic disease of the fetus and newborn

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through placental transfer from mother to the fetus. Assessing the clinical significance of antibodies relies heavily on mode of reactivity and historical data relating to specificity; functional assays are sometimes employed [5]. Blood group antigens are also marker of cancer. Blood group antigens are present on the surface of red blood cells and various epithelial cells. As the majority of human cancers are derived from epithelial cells, changes in blood group antigens are an important aspect of human tumor [6]. In some tumors, alteration of ABO/Lewis-related antigens is associated with malignant transformation [7]. A significant relationship between age and skin cancer and the old patients has 1.238 times higher risk for skin cancer [8].

The principle methodology for blood typing and antibody identification has changed little over the years, relying mainly on serological methods involving red cell agglutination. The recent advent of blood typing using DNA technology, although still in relative infancy, will surely eventually supersede serology [9].

The present study was planned to record the genotypic frequency of various alleles in the blood groups in a population of Mandi Bahauddin District of Punjab, Pakistan.

## Materials and Methods

*Subjects*

A total of 2524 subjects, comprising 427 female and 2097 males, were screened at District Headquarters (DHQ), Mandi Bahauddin, Punjab, for blood grouping during January 2005 to December 2005. The subjects belonged to both rural and urban areas. Males and females of all ages reporting at the hospital were advised blood grouping. Patients not belonging to Mandi Bahauddin and those with deep seated malignancies (e.g. carcinoma of colon) were excluded from the study. Each subject was interviewed before screening. His/her general particulars (address, age, sex, ethnic group) were recorded. Information regarding previous transfusion or blood donation was also obtained. Fingertip blood was routinely used for grouping.

*Collection of blood samples*

A 2.0 ml sample of blood, without anticoagulant, was drawn from the antecubital vein of each subject in a disposable syringe, and transferred immediately to a tube containing ethylene diamine tetra acetic acid (EDTA). In the case of doubtful or incomprehensible results, the tube method for blood grouping along with reverse grouping, using the patient's serum and known A, B and O cell was adopted.

*Determination of blood groups*

Blood grouping (ABO and Rh) was done by the antigen-antibody agglutination test. The antisera used were obtained from Plasmatic (Kent, UK). Plasmatic ABO monoclonal reagents are *in vitro* culture supernatants of hybridized immunoglobulin secreting mouse cell-line. For determination of Rh factor, plasmatic anti D (1.0 g) Lo-Du and LO-Du2 monoclonal reagents, prepared from different antibody producing human B-lymphocyte cell lines, were used.

*Computation of Allele Frequencies*

Allele frequency of the antigens was computed by application of the Hardy-Weinberg Law [5], on the basis of the number of subjects with different blood groups.

**Table 1**
Phenotypic frequencies of various blood groups (ABO and Rh) in the studied population.

| Blood Groups | Total Subjects | | | Male Subjects | | | Female Subjects | | |
|---|---|---|---|---|---|---|---|---|---|
| | Complete | Rh+ | Rh- | Complete | Rh+ | Rh- | Complete | Rh+ | Rh- |
| A | 0.2115 | 0.2084 | 0.2442 | 0.2083 | 0.2129 | 0.1689 | 0.2271 | 0.2098 | 0.3902 |
| B | 0.4076 | 0.4148 | 0.3317 | 0.3867 | 0.3982 | 0.2876 | 0.5105 | 0.5414 | 0.2195 |
| AB | 0.0756 | 0.0736 | 0.0967 | 0.0710 | 0.0713 | 0.0684 | 0.0983 | 0.0932 | 0.1463 |
| O | 0.3050 | 0.3029 | 0.3271 | 0.3338 | 0.3173 | 0.4748 | 0.1639 | 0.1554 | 0.2439 |
| Total | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 | 1.0000 |

Table 1 shows the phenotypic frequency of ABO blood groups in the studied population, with gender distribution. The distribution of phenotypes in the total sample was 0.4076, 0.3050, 0.2115 and 0.0756 for groups B, O, A, and AB, with 0.9140 Rh-positive (R) and 0.0860 Rh-negative (r), respectively. Blood group B was the commonest, while AB the rarest group in both genders. Table 2 shows the distribution of allele frequencies of ABO antigens in the studied population, in comparison with certain earlier studies. The distribution of the alleles in the total sample was 0.5522, 0.2639, 0.1391 and 0.0448 for i, IB, IA and IAB, respectively. The allelic frequency of O group was higher than that of the other blood groups in the Mandi Bahauddin population.

**Table 2**
Frequency of blood groups (ABO) studied in different populations.

| | Frequency of blood groups (%) | | | | |
|---|---|---|---|---|---|
| Population | A | B | AB | O | Reference |
| Kuwait | 0.2900 | 0.2300 | 0.1400 | 0.3500 | 11 |
| Britain | 0.4170 | 0.0860 | 0.0300 | 0.4670 | 12 |
| Kenya | 0.2620 | 0.2200 | 0.0440 | 0.4748 | 13 |
| Nigeria | 0.2443 | 0.2388 | 0.0275 | 0.4894 | 10 |
| Hungary | 0.2766 | 0.1218 | 0.0423 | 0.5553 | 14 |
| Ukraine | 0.2360 | 0.2250 | 0.0704 | 0.5760 | 15 |
| Germany | 0.2565 | 0.0810 | 0.0225 | 0.6400 | 16 |
| Turky | 0.1220 | 0.1213 | 0.0085 | 0.7398 | 17 |
| American Indians | 0.0390 | 0.0110 | 0.000 | 0.9500 | 18 |
| Bannu (Pakistan) | 0.3103 | 0.3623 | 0.0767 | 0.2507 | 19 |
| Rawalpindi | 0.2701 | 0.0375 | 0.0893 | 0.3031 | 20 |
| Peshawar (Pakistan) | 0.2800 | 0.3400 | 0.0700 | 0.3100 | 21 |
| Swabi (Pakistan) | 0.2760 | 0.3040 | 0.0880 | 0.3220 | 22 |
| India | 0.2470 | 0.3750 | 0.0530 | 0.3250 | 12 |
| Hazara (Pakistan) | 0.2400 | 0.3200 | 0.1100 | 0.3300 | 23 |
| Bahawalpur (Pakistan) | 0.2100 | 0.3600 | 0.0600 | 0.3700 | 24 |
| Wah Cant (Pakistan) | 0.1813 | 0.2450 | 0.0517 | 0.5400 | 25 |
| Gujrat (Pakistan) | 0.1740 | 0.2229 | 0.0435 | 0.5596 | 26 |
| **Mandi Bahauddin (Pakistan)** | **0.1583** | **0.2832** | **0.0448** | **0.5522** | **Present study** |

Table 3 compares the distribution of allele frequencies of Rh factor antigens in the Mandi Bahauddin population with earlier studies on different populations, suggesting the dominance of Rh positive group.

**Table 3**
Frequency of Rh antibodies in different populations.

| Population | Allele Frequency Rh+ | Rh- | Reference |
|---|---|---|---|
| China | 1.0000 | 0.0000 | 24 |
| Germany | 0.9500 | 0.0500 | 17 |
| Nigeria | 0.9430 | 0.0570 | 14 |
| Azad Jammu and Kashmir | 0.8480 | 0.1520 | 27 |
| Kenya | 0.8030 | 0.1970 | 13 |
| Gujrat (Pakistan) | 0.7958 | 0.2042 | 26 |
| Peshawar (Pakistan) | 0.7680 | 0.2320 | 21 |
| Wah Cantt (Pakistan) | 0.7390 | 0.2710 | 26 |
| Islamabad (Pakistan) | 0.7290 | 0.2710 | 20 |
| Lahore (Pakistan) | 0.7170 | 0.2830 | 28 |
| Bannu (Pakistan) | 0.6720 | 0.3280 | 19 |
| **Mandi Bahauddin (Pakistan)** | **0.9140** | **0.0860** | **Present study** |

## Discussion

Studies have been conducted by different researchers (Table 2) to estimate the gene frequency for ABO system in Nigeria (O=0.4894, A=0.2443 and B=0.2388), Hungary (O= 0.5593, A=0.2989 and B=0.1418), Ukraine O=0.5760, A=0.2360 and B=0.2250) and in South Western Germany (O=0.640, A=0.279 and B=0.081). In Pakistan, work on ABO blood group allelic distribution has been done in different areas (Table 2) including the gene frequency distribution in Gujrat (O=0.5596A=0. 1740 and B=0.2229), Wah cant (O=0.5400, A=0. 1813 and B=0.2450) and in Bahawalpur (O=0.3700, A=0.2100 and B=0.3600).

By comparing these data with the data of the present study (O=0.5522, A=0.1583 and B=0.2832) it is evident that ABO allelic distribution for Mandi Bahauddin region (Pakistan) is very close to the distribution for Hungary, Ukraine, Gujrat and Wah Cant. population [14,15,25,26] and is different compared with the population of Kuwait, Germany, Turkey, American Indians, Bannu, Rawalpindi, Peshawar, Swabi, India, Hazara, and Bahawalpur [11,12,16, 18,19,24]. However, comparison with the data from the British and African populations reveals that there is an equal dominance of group B and O in the Indo-Pak subcontinent, in contrast to dominance of only O group in the British and African populations [10,12,13]. The least reported group, in all the populations, has been AB. These studies suggest that the heterogeneity in the different populations is due to the genetic and environmental factors, which helps the expression of blood group B alleles. However, this requires further investigations on Pakistani populations.

In terms of presence of Rh antibodies, the data from several studies on Pakistani as well as certain African populations are compared in Table 3, along with the allele frequency of R and r. The findings of the present study are inconsistent with the results obtained in an earlier study carried out on the population of Nigeria (R=0.9430, r=0.0570) and Azad Jammu and Kashmir (R=0.8480, r=0.1520), where the allele frequency of Rh-positive (R) has been found to exceed the Rh-negative (r) cases. However the allele frequency of Rh positive was less than the population of China and Nigeria (Table3), but showing the highest rate (R=0.9140, r=0.0860) of Rh positive than in the other regions of the Pakistan studied so far. On the other hand, it has been reported that the Rh negative phenotype is more prevalent in patients with small-cell lung cancer than in the normal Caucasian population. [29-34].

These present results provide in depth information of the relative distribution of various alleles in the population, especially indicating the progressively increasing dominance of B positive group relative to the distribution of various alleles in the population in the Mandi Bahauddin District and promise help in planning for future health challenges, particularly regarding blood transfusion.

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