Polymorphismic Changes in Blood Groups of the Naga Tribes of Nagaland

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Northern India is one of the oldest geophysical regions of human evolution and migration in the world. Genetic and anthropological studies have shown that the peopling the subcontinent is characterized by a complex history, contributed from different ancestral populations. Genetic level studies of polymorphisms always associated with health and diseases of population becoming the need of hours. The present study was aimed to explore the relationship of ABO blood groups association of populations and to assess the prevalence of blood groups in different categories of Northern India and to compare our results with other studies conducted in India. Blood samples from 155 unrelated individuals was collected from Naga tribal settlements Blood groups of individuals were evaluated with the presence of antigen using monoclonal antibody by a standard blood group O 67 (43.23%) was the commonest group prevalent in donors followed by group B 45 (29.03%), A 33(21.29%) and AB 10 (6.45%). The Rh negative was not observed in female donors, whereas in male it was found to be 1.3%. Data among tribal suggest their common origin as well as a drift from an original population due to the possible founder effect among tribal Naga Ten Tribal

Keywords: ABO blood groups, Rhesus factor, Diversity, Diphupar-B, , Nagaland.

The ABO blood group system was the first human blood group system to be discovered by Landsteiner in 1900¹. The ABO blood group system is the only system in which antibodies are consistently and predictably present in the serum of normal individuals whose red cells lack the antigens. Apart from differences amongst species, differences between the individuals of the same species have also been demonstrated. During the world wars, it was discovered for the first time that the frequency of ABO and Rhesus blood groups was different in persons native to different parts of the world²⁻³. ABO and Rh gene phenotypes vary widely across races and geographical boundaries despite the fact that the antigens involved are stable throughout life. The resultant polymorphism remains important in population genetic studies, estimating the availability of compatible blood, evaluating the probability of hemolytic disease in the new born, resolving disputes in paternity/

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maternity and for forensic purposes⁴. The frequency of ABO and Rh phenotypes in different populations hasve been extensively studied. Rh system emerged as second most important blood group system due to hemolytic disease of newborn and its importance in RhD negative individuals in subsequent transfusions once they develop Rh antibodies. The D antigen, after A and B, is the most important red cell antigen in transfusion practice⁵. Unlike the situation with A and B, persons whose red cell lacks the D antigen do not regularly have anti D in their serum. Blood bank usually has a problem of everchanging stock position and it being very difficult to predict the prevalence of a particular blood group at a particular time. The present study was done to assess the prevalence of blood groups in different categories of Northern India and to compare our results with other studies conducted in India and elsewhere in the world and its multipurpose future utilities for the health planners⁶⁻⁸.

MATERIALS AND METHODS

The study of ABO and Rhesus Blood Group was carried out on 155blood donors (male and female) during a period of two months from 1stseptember to 31st October 2018 in Diphupar-B area Dimapur, Nagaland. The blood donors were selected after taking a detailed history and a complete examination regarding their eligibility criteria for blood donation. Donor's name, age, sex, occupation, caste, complete postal address and contact number was recorded. Blood samples were obtained by standard procedures of venupuncture and subjected to determination of ABO and Rhesus blood group using "antisera" by combined slide and test tube method. Each sample of donors was tested for ABO and Rhesus status.

Laboratory analysis

ABO and Rh blood group tests are carried out by a standard protocol using AB D Antisera typing Kit.(Fig. 1.)

| Phenotype (blood group) | Genotype | Phenotype frequency | Genotypic frequency | Expected frequency |
|----------------------------|----------|------------------------|------------------------|--------------------|
| A | AA+AO | ηΑ | ηAA+ηAO | p2 + 2pr |
| В | BB+BO | ηΒ | $\eta BB + \eta BO$ | $q^2 + 2qr$ |
| 0 | AB | ηÅB | ηAB | 2pq |
| AB | OO | ηO | ηΟΟ | r^2 |

Table 1. Hardy-Weinberg model for ABO blood group

 Table 2. Overall allele frequencies for the ABO and Rh antigens in the Ten studied populations

| Group | <i>p</i> [A] | Gene frequency q[B] | <i>r</i> [0] | Hardy- Weinberg loglikelihood | Genotypic frequency | χ2 | Pvalue | Rh+(D) | Rh-(d) |
|------------|--------------|---------------------------|--------------|-------------------------------------|------------------------|------|--------|--------|--------|
| Total | 0.16 | 0.2 | 0.64 | -179.3 | O>B>A | 0.11 | 0.73 | 0.99 | 0.01 |
| Chekhesang | 0.13 | 0.30 | 0.57 | -57.48 | O>B>A | 0.95 | 0.32 | 1.00 | 0.00 |
| Sumi | 0.27 | 0.03 | 0.70 | -15.20 | O>A>B | 0.39 | 0.52 | 1.00 | 0.00 |
| Angami | 0.10 | 0.10 | 0.8 | -15.09 | O > A = B | 0.39 | 0.53 | 1.00 | 0.00 |
| Ao | 0.20 | 0.27 | 0.51 | -40.03 | O>B>A | 1.45 | 0.22 | 0.97 | 0.03 |
| Mao | 0.2 | 0.16 | 0.64 | -16.40 | O>A>B | 1.34 | 0.25 | 1.00 | 0.00 |
| Lotha | 0.13 | 0.07 | 0.8 | -07.30 | O>A>B | 0.17 | 0.68 | 1.00 | 0.00 |
| Konyak | 0.23 | 0.44 | 0.32 | -09.17 | B>O>A | 0.47 | 0.50 | 1.00 | 0.00 |
| Zeliang | 0.15 | 0.15 | 0.70 | -08.07 | O > A = B | 1.59 | 0.20 | 1.00 | 0.00 |
| Sangtam | 0.28 | 0.28 | 0.44 | -05.75 | O > A = B | 0.42 | 0.51 | 0.63 | 0.33 |
| Phom | 0.42 | 0.18 | 0.40 | -07.62 | A>O>B | 0.02 | 0.90 | 1.00 | 0.00 |

The study populations used in blood groupings occurred in the order O > B > A > AB. The allele frequency of blood group O was the highest ABO allele p (A), q (B), r (O) as 0.16, 0.2, 0.64, respectively. This occurred in the order O > B > A. The allele frequency of blood group O is the highest $\chi 2$. The goodness of fit test was resulted in value was = 0.11 and p value was 0.73. The genotype frequencies are reached (D) = 99.0% and (d) = 1%, genotype frequencies are (D) = 0.99 and (d) = 0.01. There is the high proportion of Rh (D) +ve individuals than the Rh - vein the study populations

Statistical analysis

The gene and allele frequencies of blood group, are calculated by Hardy-Weinberg model using S2 ABO estimator software⁷. Allele frequencies are calculated under the assumption of Hardy–Weinberg equilibrium and expressed as percentages. The chi - square test is used to compare observed allelic and genotypic frequency distributions of the blood group and Rh antigens to that of under the Hardy–Weinberg⁹. ABO blood was aligned according to their respective frequencies in populations using PAST software : PCA and Phylogenetic trees were constructed, using Neighbor-Joining (NJ) method total of 155 (male and female)donor population was compared. Amongst Rh positive female donors blood group O was found to be most prevalentin group 34(45.95%), followed by group B 23(31.08%), A 14(18.92%) and AB 3(4.05%). Amongst Rh positive male donors blood group O was the most prevalent group 33(41.77%), followed by group B 21(26.58%), group A 18(22.78%) and group AB 7(8.86%). The Rh negative does not exist in any female donors. The Rh negative appears only in male donors in blood group A-ve(1) and B-ve(1)with 1.3%..

DISCUSSION

RESULTS

The ABO and Rhesus blood Group in Diphupar-B showsed various typical results. The frequency of ABO and Rh blood groups in a

The Neighbour Joining Analyses

Population relationships can be easily represented by Neighbor Joining (NJ) trees. To study the genomic affinities of the tribal populations of Nagalnd, NJ trees was constructed

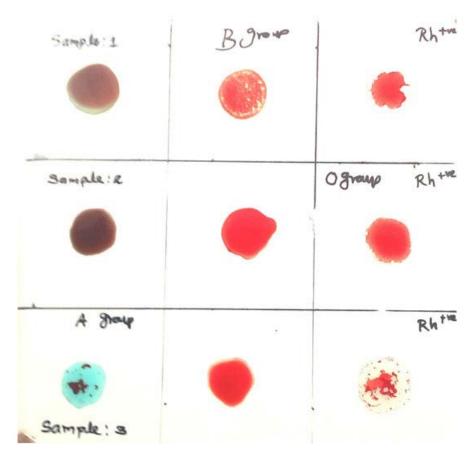


Fig.1. The Plate Showing the the result of A B O blood groups

by using various reported investigations in the tribes of India and globally by means of same set of loci/alleles. In the first NJ tree analysis, given in Fig.2. ABO blood polymorphic loci alleles are used to construct the tree to find the genomic affinities among the tribal populations of Nagaland under study Interestingly, this tree showed that *Mao* are clustered with Sumi who are geographically distant from *Mao* instead of *Zeliang*, who are thought as same to *Sumi, Angami* and *Lotha*. Another surprising genetic relation was established between Ao and Chekesang, though both are geographically

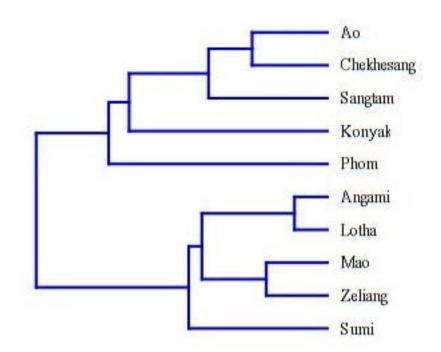


Fig. 2. Dendrogram of Ten Naga Tribal populations on diversity for ABO polymorphic loci

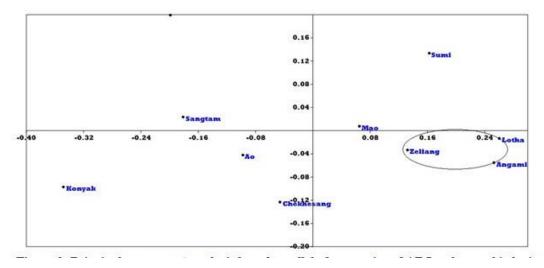


Fig.3. Principal component analysis based on allele frequencies of ABO polymorphic loci

| $ \begin{array}{cccccccccccccccccccccccccccccccccccc$ | Sl No. | Name | Age | Sex | Blood Group | Tribe |
|--|--------|--------------|-----|-----|-------------|------------|
| 3 Asenuo 22 F O+ Angami 4 Superro 19 F B+ Lotha 5 Nyemang 21 F O+ Konyak 6 kilingna 20 F O+ Zeliang 7 Amy 19 F A+ Sumi 8 Woben 21 F O+ Lotha 9 Savinu 21 F O+ Angami 10 Wesly 21 F O+ Angami 12 Pelevituo-u 24 F O+ Angami 13 Mhasilenuo 45 F O+ Angami 14 pazini 22 F B+ Mao 15 A.moses 22 M B+ Mao 16 Amenla 23 F A+ Mao 19 Leshini 30 F A+ Mao 20 Kokhrolu 24 M O+ Mao 23 <td< td=""><td></td><td>Medou</td><td></td><td></td><td>O+</td><td>Chakhesang</td></td<> | | Medou | | | O+ | Chakhesang |
| $ \begin{array}{cccccccccccccccccccccccccccccccccccc$ | | Awistoli | | F | O+ | Sumi |
| 5 Nyemang 21 F O+ Konyak 6 kilingna 20 F O+ Zeliang 7 Amy 19 F A+ Sumi 8 Woben 21 F O+ Lotha 9 Savinu 21 F O+ Lotha 9 Savinu 21 F O+ Chakhesang 10 Wesly 21 F O+ Sumi 12 Pelevituo-u 24 F O+ Angami 13 Mhasilenuo 45 F O+ Angami 14 pazini 22 F B+ Mao 15 A.moses 22 M B+ Mao 18 Sentiro 22 F A+ Mao 20 Kokhrolu 24 M O+ Mao 21 Eliveyi 24 M O+ Sumi | | Asenuo | 22 | F | O+ | Angami |
| 6 kilingna 20 F O+ Zeliang 7 Amy 19 F A+ Sumi 8 Woben 21 F O+ Lotha 9 Savinu 21 F A+ Angami 10 Wesly 21 F O+ Chakhesang 11 Kenlingunuuo 19 F O+ Angami 13 Mbasilenuo 45 F O+ Angami 14 pazini 22 F B+ Mao 16 Amones 22 M B+ Mao 16 Amones 23 F A+ Mao 19 Leshini 30 F A+ Mao 20 Kokrolu 24 M O+ Mao 21 Eliveyi 24 M O+ Mao 22 Rokositi 23 M A+ Angami <tr< td=""><td></td><td>Supenro</td><td>19</td><td>F</td><td>B+</td><td>Lotha</td></tr<> | | Supenro | 19 | F | B+ | Lotha |
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| 28Khrilazo22MO+Sumi29Savito22MO+Sumi30Nyewe-u21FO+Chakhesang31Julie19FB+Chakhesang32Krecheni22FO+Mao33Aniphro22FA+Mao34Marini21FO+Mao35Tsukhum22MB+Mao36Fuchumlo22FO+Lotha37Lutsu22FO+Pochury38Asen22FO+Yimchunger40Hukai24MO+Sumi41Asang49FB+Ao42Kehosedel54MO+Angami43Roko22MB+Angami44Nieketouzo22MO+Angami45Kenli23FO+Lotha46Alokeu24FO+Lotha47Lanu25FO+Sumi48Mhalevino22FA+Sumi49Rencham022FO+Ao50Vilasilie22MB+Angami | | | | | | |
| 29Savito22MO+Sumi30Nyewe-u21FO+Chakhesang31Julie19FB+Chakhesang32Krecheni22FO+Mao33Aniphro22FA+Mao34Marini21FO+Mao35Tsukhum22MB+Mao36Fuchumlo22FO+Lotha37Lutsu22FO+Pochury38Asen22FA+Ao39Atsu21FO+Yimchunger40Hukai24MO+Sumi41Asang49FB+Ao42Kehosedel54MO+Angami43Roko22MB+Angami44Nicketouzo22MO+Angami45Kenli23FO+Angami46Alokeu24FO+Lotha47Lanu25FO+Sumi48Mhalevino22FA+Sumi49Rencham022FO+Ao50Vilasilie22MB+Angami | | | | | | - |
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| 42Kehosedel54MO+Angami43Roko22MB+Angami44Nieketouzo22MO+Angami45Kenli23FO+Angami46Alokeu24FO+Lotha47Lanu25FO+Sumi48Mhalevino22FA+Sumi49Rencham022FO+Ao50Vilasilie22MB+Angami | | | | | | |
| 43Roko22MB+Angami44Nieketouzo22MO+Angami45Kenli23FO+Angami46Alokeu24FO+Lotha47Lanu25FO+Sumi48Mhalevino22FA+Sumi49Rencham022FO+Ao50Vilasilie22MB+Angami | | | | | | |
| 44Nieketouzo22MO+Angami45Kenli23FO+Angami46Alokeu24FO+Lotha47Lanu25FO+Sumi48Mhalevino22FA+Sumi49Rencham022FO+Ao50Vilasilie22MB+Angami | | | | | | 6 |
| 45Kenli23FO+Angami46Alokeu24FO+Lotha47Lanu25FO+Sumi48Mhalevino22FA+Sumi49Rencham022FO+Ao50Vilasilie22MB+Angami | | | | | | |
| $\begin{array}{cccccccccccccccccccccccccccccccccccc$ | | | | | | |
| 47Lanu25FO+Sumi48Mhalevino22FA+Sumi49Rencham022FO+Ao50Vilasilie22MB+Angami | | | | | | |
| 48Mhalevino22FA+Sumi49Rencham022FO+Ao50Vilasilie22MB+Angami | | | | | | |
| 49Rencham022FO+Ao50Vilasilie22MB+Angami | | | | | | |
| 50 Vilasilie 22 M B+ Angami | | | | | | |
| | | | | | | |
| | | | | | | |
| 51 Vileto 24 M A+ Angami 52 Nchumbeni 21 F O+ Angami | 51 | Vileto | 24 | М | A+ | Angami |

Table 3. Detailed list of all donors and their corresponding bio-data

| 53 | Adino | 25 | F | O+ | Muslim |
|----------|-------------|----------|--------|----------|--------------|
| 54 | Jabeda | 25 | F | A+ | Muslim |
| 55 | Yantsuntung | 26 | М | A+ | Lotha |
| 56 | Sai | 23 | М | O+ | Zeliang |
| 57 | Merlin | 2 | F | B+ | Pochury |
| 58 | Sentilemla | 24 | F | A+ | Ao |
| 59 | Akato | 25 | Μ | O+ | Suni |
| 60 | Kewe | 26 | М | O+ | Sumi |
| 61 | Mercy | 4 | F | O+ | Sumi |
| 62 | Manen | 22 | F | O+ | Sumi |
| 63 | Nally | 23 | М | A+ | Phom |
| 64 | Seyiesu | 24 | F | B+ | Zeliang |
| 65 | Waltrina | 25 | М | AB+ | Angami |
| 66 | Alona | 26 | F | A+ | Garo |
| 67 | Kivisu | 21 | М | O+ | Angami |
| 68 | Binlozu | 30 | F | B+ | Rengma |
| 69 | Keyiening | 32 | F | O+ | Zeliang |
| 70 | Vikeithozo | 23 | М | O+ | Angami |
| 71 | Vinoka | 22 | M | B+ | Sumi |
| 72 | Temjen | 22 | M | A+ | Ao |
| 73 | Wapang | 22 | M | AB+ | Ao |
| 74 | Yapani | 23 | F | B+ | Loyha |
| 75 | Nungsen | 23 | M | O+ | Ao |
| 76 | Sianglu | 23 | F | A+ | Zeliang |
| 77 | Esther | 23 | F | B+ | Konyak |
| 78 | Levilu | 23 | F | AB+ | Chakhesang |
| 78 79 | Lipok | 22 | M | AB+ | Ao |
| 80 | Lijingba | 30 | M | A- | Sangtam |
| 80 81 | Bishoka | 30 24 | F | A- O+ | Dimasa |
| 82 | Alemla | 24 | F | 0+ 0+ | Ao |
| 82 83 | Aloto | 23 24 | г М | A+ | Sumi |
| 83 84 | Vihuka | 24 | M | A+ A+ | Sumi |
| | | 23 21 | | | |
| 85 | Anoka | | M | A+ | Sumi |
| 86 | Bokato | 25 | M | A+ | Sumi |
| 87 | Mercy | 23 | F | B+ | Konyak |
| 88 | Akok | 21 | M | A+ | Ao |
| 89 | Anengla | 22 | F | B+ | Ao |
| 90 | Sunglu | 22 | F | B+ | Ao |
| 91 | Mabeni | 24 | F | O+ | Lotha |
| 92 | Menibeni | 21 | F | A+ | Lotha |
| 93 | Alisong | 23 | М | O+ | Ao |
| 94 | Aka | 22 | M | B- | Ao |
| 95 | Easter | 24 | F | O+ | Sangtam |
| 96 | Kese | 23 | М | O+ | Angami |
| 97 | Even | 23 | M | B+ | Konyak |
| 98 | Juniya | 24 | F | B+ | Dimasa |
| 99 | Lily | 24 | F | O+ | Konyak |
| 100 | Hemsu | 23 | F | B+ | Khiamniungun |
| 101 | Asen | 22 | F | AB+ | Ao |
| 102 | Vili | 21 | М | A+ | Sumi |
| 103 | Velu | 21 | F | O+ | Chakhesang |
| 104 | Imti | 21 | М | A+ | Ao |
| 105 | Wati | 23 | М | B+ | Ao |
| 106 | Lanu | 23 | М | AB+ | Ao |
| 107 | Amsong | 23 | М | O+ | Ao |
| | | | | | |

| 108 | Tatong | 25 | М | \mathbf{B}^+ | Ao |
|-----|-----------|-----------|---|----------------|--------------|
| 109 | Temjen | 22 | Μ | O+ | Ao |
| 110 | Akaba | 19 | М | O+ | Ao |
| 111 | Atubu | 22 | М | \mathbf{B}^+ | Ao |
| 112 | Sama | 21 | М | AB+ | Ao |
| 113 | Tia | 18 | М | O+ | Ao |
| 114 | Moa | 21 | М | \mathbf{B}^+ | Ao |
| 115 | Sunep | 22 | М | B+ | Ao |
| 116 | Tako | 23 | М | A+ | Ao |
| 117 | Sashi | 27 | М | O+ | Ao |
| 118 | Toshi | 45 | М | O+ | Ao |
| 119 | Yinu | 43 | М | B+ | Ao |
| 120 | Melekho | 22 | М | O+ | Chakhesang |
| 121 | Vekho | 22 | М | B+ | Chakhesang |
| 122 | Veto | 22 | М | O+ | Chakhesang |
| 123 | Vethi | 22 | М | A+ | Chakhesang |
| 124 | Achi | 22 | М | A+ | Chakhesang |
| 125 | Akho | 21 | М | A+ | Chakhesang |
| 126 | Ato | 21 | М | B+ | Chakhesang |
| 127 | Khoto | 24 | М | \mathbf{B}^+ | Chakhesang |
| 128 | Khrupu | 25 | М | O+ | Chakhesang |
| 129 | Khruhu | 25 | М | B+ | Chakhesang |
| 130 | Shehu | 23 | М | B+ | Chakhesang |
| 131 | Michivo | 24 | M | A+ | Chakhesang |
| 132 | Vechipo | 21 | M | B+ | Chakhesang |
| 133 | Kuvezo | 24 | M | AB+ | Chakhesang |
| 134 | Vebuzo | 42 | M | AB+ | Chakhesang |
| 135 | Vemucho | 33 | M | B+ | Chakhesang |
| 136 | Khoshe | 31 | M | O+ | Chakhesang |
| 130 | Zhosheku | 31 | M | O+ | Chakhesang |
| 138 | Vepopu | 31 | M | O+ | Chakhesang |
| 139 | Роро | 34 | M | O+ | Chakhesang |
| 140 | Alepu | 22 | M | B+ | Chakhesang |
| 140 | Aku | 22 | M | O+ | Chakhesang |
| 142 | Khrope | 22 | M | B+ | Chakhesang |
| 143 | Atalu | 34 | F | B+ | Chakhesang |
| 144 | Asulu | 32 | F | O+ | Chakhesang |
| 145 | Avelu | 23 | F | B+ | Chakhesang |
| 146 | Shelu | 23 | F | B+ | Chakhesang |
| 140 | Khrulu | 21 | F | O+ | Chakhesang |
| 148 | Khotalu | 22 | F | B+ | Chakhesang |
| 149 | Abi | 22 | F | O+ | Chakhesang |
| 150 | Atho | 32 | F | B+ | Chakhesang |
| 150 | Vebunielu | 34 | F | AB+ | Chakhesang |
| 151 | Shetolu | 24 | F | AB+ A+ | Chakhesang |
| 152 | Shekhrulu | 43 | F | A+ A+ | Chakhesang |
| 155 | Tsinolu | 23 | F | B+ | Chakhesang |
| 154 | Vetholu | 23 | F | B+ | Chakhesang |
| 100 | , culoiu | <u>~1</u> | 1 | ' U | Chakinesailg |
| | | | | | |

very far-off. Overall, this tree showed that the Naga tribes under consideration are interrelated and equally distant from each other in terms of ABO blood allele frequencies.

Principal Component Analysis

Principal component analyses (PCA) was performed to identify possible clustering among the populations. In the PCA (Figure.3.)

using ABO blood, the populations *Zeliang*, *Lotha* and *Angami* are observed to be falling under a single cluster among the populations.

Human shares the same blood group systems, differing only in the frequencies of specific types. The incidence of ABO, Rh and MN groups varies in different parts of the world and in different races¹⁰. Geographical and ethnic groups and socioeconomic groups,¹¹⁻¹². Many studies have revealed possible associations of various diseases with the ABO blood group, but the reasons for such associations are remain controversial¹³⁻¹⁴. With Blood group O has a greater incidence of association with hypertension¹⁵ and melanoma¹². As far as abortion is concerned, it is found to be higher in between A type husband and O type wife and high in couples with combination of A type husband and B type wife. The frequencies of Rh positive and Rh-negative individuals are 96.5% and 3.5% respectively¹⁶. ABO blood groups are important in determining migration of races and in hereditary diseases¹⁷. Some diseases are more common to develop in certain blood groups; hence the relationship of different blood groups with diseases is important¹⁸.

The genomic affinities among eleven study populations are represented in Fig2& 3, using allele frequency data of four loci by a standard NJ tree and Principal component analysis. It has been that all student groups are genetically related to each other, to asses genomic affinities of the ten study populations, the data from the results of presents study groups are assured together and represented in an others NJ tree andPrincipal component analysis (Fig.2& 3).

CONCLUSION

In conclusion, based on ABO blood group poylmpohsms the Naga tribes have geneticroots with others Indian population. Studieds association of ABO blood group polymorphismand disease prevalence in populations of India need to be established using molecular markers, such as nDNA, mtDNA etc. Whichright clear the picture in the human biography of the Indian subcontinent

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