



Identification of Genetic Lineage of Peshawar and Nowshera Tribes through Dental Morphology

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ABSTRACT

Analyzing the diversity of non metric phenotypic traits of the mature teeth, in three ethnic groups viz. Daudzai, Khalil and Mohmand of Peshawar districts, of the Khyber Pakhtunkhwa Province (KP) of Pakistan. Dental casts obtained from both mandible and maxilla of 375 volunteers in which 75 male and 50 females of all the tribes. The dental casts for each individual were analyzed for a set of 35 tooth-trait accordance with the Arizona State University Dental Morphology System. The data was compared with the secondary information of 3842 individuals representing 36 other ethnic groups of different parts of KP, peninsular India prehistoric samples of Indus valley Central Asia and South Asia. Inter sample affinities based upon pair wise MMD values were examined with neighbor-joining cluster analysis and multidimensional scaling. The results remained highly consistent throughout different data reduction techniques. The consistency in phenetic affinities was even evident when they were considered in the backdrop of living ethnic groups of peninsular Indians and prehistoric inhabitants of the Indus Valley and South-Central Asia. When consideration was limited to the people of Northern Pakistan, the uniqueness of Khalil and Daudzai, was emphasized. A comparison of the people from Peshawar emphasizes the uniqueness with south-central Asia, and shared no affinities to South Asians either living or prehistoric. These results show the influence of the past expansions and migrations have had upon the Pakistani population.

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Authors' Contribution

HA and BEH conceived the idea and designed the research project. MZ and MS conducted sampling and generated data. MZ and MT performed the data analysis. MZ and MS drafted the manuscript. BEH and HA reviewed the manuscript.

Key words

Daudzai, Dental anthropology, Khalil, Mohmand, Peshawar, Phenetic affinities

INTRODUCTION

Review of the available secondary information shows that the modern human beings spread over the entire world stating their journey from Africa about 100 thousand years ago (kya), a group of these people migrated from Africa to other parts of the world. No one knows for sure what initiated this migration. It has been speculated that after evolving in Africa, population sizes of modern humans grew rapidly, and they need more natural resources. They started their journey in search of new territories to explore additional food. There may also have been other causes for the dispersal, such as behavioral changes (Mellars, 2006). Archaeological and genetic data in particular shows that the descendants of those who migrated from Africa ceased their number to form most present-day human populations (Oestigaard *et al.*, 2004)

Furthermore, these migrants initially colonized South Asia comprising India, Pakistan, Nepal, Bhutan, Bangladesh, Sri Lanka and parts of South-East Asia, particularly Myanmar. The exact route taken by these migrants is not known. Some researchers suggest that the first traveled via a southern coastal route, earliest settlers are not known, although it has been suggested that they traveled via a southern coastal route. Based on different geographic distances between various populations and measures of population differentiation derived from quantitative cranial datasets, multiple dispersals took place between approximately 37 and 135 kya (1000 years ago). As the modern human expansion continue outside Africa, they settled down in different geographical locations in a small group. The process of settling may have taken place serially or in a stepwise fashion. That is, as a group of humans moved out of Africa and settled down in a new geographical area, they continuously inhabit new area for several hundred or several thousand years, until the population expansion and associated pressure on natural resources forced a group of humans from the new area to

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settle yet area. This process continues for thousands of years until they occupy the entire world (Ramachandran *et al.*, 2005). Because of the creations of new ethnic populations, and the evolutionary accumulation of resulting in new genetic variation, it is expected that the genetic distance between the African ethnic population and its descendant groups will increase with increasing geographic distance between them. Indeed, such a relationship has been observed. A possible situation of the process of peopling is that the group of modern humans who moved out of Africa settled in a new place and then experienced population expansion in the new place and then their subgroup further moved on to another place. In other words, a long serial expansion, separation and movement perhaps resulted in the major geographical spread of modern humans. Support for this kind of migration of people has been obtained from genetic data (Ramachandran *et al.*, 2005). The process of local population expansion followed by dispersal to newer areas, coupled with evolution of cultural practices and geographical distance acting as a barrier to human contact, also resulted in the formation of intra-marrying groups; groups within which there was considerable gene exchange among individuals, but between which there was little genetic exchange. The development and evolution of language and culture exaggerated the formation of such groups. Genetic diversity within a geographical area is a reasonable indicator of the age of the populations' residence in that area. However, it is also dependent on the effective sizes of populations, implying thereby that the assessment of antiquity may not be straightforward. It is now well-established that Africa harbors the highest genetic diversity among continental populations. Notably, the genetic diversity in India, the heartland of south Asia, is second only to that of Africa. Thus, it is likely that India became peopled in one of the early waves of migration that originated in Africa. What remain controversial is whether there were multiple waves of migration from Africa, and what the timing of these migration events was. Lahr and Foley (1994) for instance, postulated that there were two waves: one via a northern exit route through the Nile valley associated with Upper Paleolithic blade-dominated technology (as found in excavation sites in Israel and Lebanon, both dated between 45 and 50 kya) and another, associated with the simpler Middle Paleolithic technology, via a southern exit route from the Horn of Africa across the mouth of the Red Sea along the coastline of India to southeastern Asia and Australia. The dispersal through the southern exit route was postulated to have taken place substantially earlier (about 70–80 kya) than that through the northern route (about 45 kya). Archaeological evidence to resolve this controversy has been scanty, primarily

because the coastlines of that period have become deeply submerged because of the rapid rise of sea levels (Stringer, 2000). However, some recent archaeological finds from India have been interpreted to reveal that the major route of dispersal was through the southern route. This route was reinforced by tooth record of the fossil human. Teeth demonstrate the smallest amount for cellular output of the body structure, and that they are unit promptly reachable for analysis. To comprehend the organic process relationship among completely different populations is increasing with the passage of time (Reich *et al.*, 2009). To overcome the issue completely different models are unit used. Tooth morphology provides necessary biological structures that are human specific and stick with it a lot of like alternative genetic personas e.g. Blood group, genetic factor, finger prints configurations, color and height etc. The exploration of dental morphology drives back to 19th century, once dental anatomists exposed several dental personas, such as associate further edge on mesiolingual exterior of jaw molars (Carabelli, 1842). Teeth are sturdy structures that preserve terribly sound in archaeological and remnant record and might simply be cycled for launching associations with breathing inhabitants. They are wonderful material in alive populations for genetic, odontology, anthropologic inquiries, it's typically believed that various morphologic features of teeth are unit genetically indomitable. The selection of knowledge concerning human progression (Hillson, 2002), immigration configurations and for finding rhetorical circumstances (Brown, 1992). Also it's helpful to commence educations in dental social science through tooth morphology could also be associate gauge of genetic detachments among populations, it ought to be observed with thoughtfulness. The learning of the quantity of edges do not seem to be solely necessary in social science it's additionally necessary within the learning of Dental obstruction, Dental medicine, Restorative odontology and Prosthetic odontology. Learning of tooth pinnacle morphology are vital in hominid biology and phylogenies. Above the last 30 years, a combined struggle was created to systematize the cataloging of dental persons and to research however their manifestation differs among human populations. In explicit, dental morphological traits are acknowledged for his or her significance as a phenotypical manifestation of genetic variations among teams among teams. By the ways of dental morphology associate investigations is feasible evaluation to genetic populace dynamics. The learning of non metric dental attributes contains genetically modulated feature terminologies which will be cycled for judgments among populations. The cusps on inferior on first and 2nd bicuspid generally; mesiolingual,

mesiobuccal, distobuccal, distolingual and centrobuccal that area unit on an individual basis situated matched to cusps on alternative teeth (Kraus and Furr, 1953). At the distal fringe of the crown of inferior premolars there could be an extra cusp to lingual of the distobuccal, called cusp 6. An extra cusp referred to as seven may lie on lingual fringe of the crown among the distolingual and mesiolingual cusps at alike teeth (Hillson, 1996). The total population of 30523371 is dependably assumed part of a door for trespassers in old occasions (Hilali, 2016). Pakhtuns are the largest racial gathering is represented by 73.9% while the Hindko speakers are only 20.4% of the total population. Peshawar is the capital of province associated with inborn offices in south, north, and west and with Nowshera in the east (Tahir, 1998). Peshawar District is situated in the north-west of Pakistan; the capital city of Khyber Pakhtunkhwa Province (KP) is Peshawar, around 160 km west of the national capital Islamabad. Peshawar is important both geo-strategically and an enhanced history. The history of Peshawar goes back to no less than 539 B.C.E., making it one of the most ancient cities in Pakistan and the oldest one in South Asia (Caroe, 1957). The city has seen the ascent and fall of numerous civic establishments like Bactrian's, Gandara, Ionians, Kushans, Hepathalites. Nowshera is located 34.02° North latitude and 97° East greatit's border by urban center district to West, Mardan to North, Swabi District to the North-East, Charsadda to North-West, Kohat to south, Attock to east and Orakzai agency to southeast 1,748km² total spaces (District at a Glance Nowshera, 2016). Traditionally inhabitants use to talk Hindko that is spoken in areas of Akorakhattack, Nowshera kalan, Shaidu and a number of other communities located on the grand highroad. The main tribe within the dirstrict comprises of Khesghi, Khattak, Kaka khels, Mohammadzai, Salarzari and Ahmadzai (Hart, 2001). Ethnicity could be a cluster of people known on the basis of region, race, language and color of a particular area. The surroundings culture groups don't seem to be outlined solely support natural situation however additionally the existences events of alternative ethnic teams and every ethnic cluster will have used a region of total surroundings and remain and leave the massive elements of it exposes for alternative group to be cycled. For the comprehension of the source still as progress of any species largely kinsmen (*Homo sapiens*) partaking progression in their genetics that provides a heap of up keep in proofs for biological, social science and biological process data for quality (Cann *et al.*, 1987). Many researchers have unconcealed that the populace is not varied in reports of treatment reactions and illness risk (Bamshad *et al.*, 2001).

MATERIALS AND METHODS

The study was conducted on the dental morphology of three major tribes i.e. Mohmand, Khalil, Daudzai of Peshawar districts of KP. From each tribe, 70 samples male and 50 samples females were collected for phenotypic analysis of teeth. The samples were collected from school going volunteers in different villages in both the districts (Table I).

Sampling criteria

Four factors *viz.* age, affinity, dental status and ethnic group identity were considered during sample collection. The samples were taken from individuals with age between 12 and 22 years with full eruption of all permanent teeth except third molars. A questionnaire containing required information about ethnic group, geographical origin of their grandparents and first language were filled by the all participants of this study.

Biosafety measures

Disinfected dental trays were used to take dental impressions in alginate. A hard plaster was used to fill the template after taking impression. Small amount of water was added to make semi fluid mixture and poured immediately into tray of exact size of individual. The tray was seated into mouth which has the exact size of individual jaw. After few minutes, the tray was taken out from the mouth of individual and kept in an open air.

Pouring

The casting material was cast down to the impressions and gently taped to remove the air bubble. After 30 minutes, the impression material and impression tray was gently separated from cast. The fully prepared cast was used for further analysis while the tray was treated with some antiseptic to reuse.

Data collection

After dental cast collection, the nonmetric dental traits were characterized and documented (Turner *et al.*, 1991; Scott and Turner, 1997). The dental characterization and their charting was done. The different characters found in the upper jaw (Maxilla) and lower jaw (Mandible) are following.

Maxilla

Winging, curvature, shoveling, double shoveling, interruption grooves, tuberculum dentale, canine distal accessory ridges, premolar mesial and distal cusps and ridges, premolar accessory cusp, premolar accessory ridges, metacone, UTO-aztecan premolar, hypocone, metaconule, protoconule, parastyle, carabelli's trait, pegging.

Table I. Summary of the sampling.

S. No	Ethnic group	Sampling site	Gender	No. of caste
1	Mohmand	Peshawar (Ashi Korona, Dab Kor)	Male	75
2	Mohmand	Peshawar (Ashi Korona, Kado Kor)	Female	75
3	Daudzai	Peshawar (Nelavi, Bela, Baramand Khel)	Male	75
4	Daudzai	Peshawar (Nelavi, Bela, Shagi)	Female	75
5	Khalil	Peshawar (Abdara, Pawakai)	Male	75
6	Khalil	Peshawar (Pawakai, Spena Warai)	Female	75
7	Kheshgi	Nowshera (Lali khel, Meta Khel, Naqdezi)	Male	75
8	Kheshgi	Nowshera (Miangan, Kher Khel, Umerzai)	Female	75
9	Mohmandzai	Nowshera (Nowshera Kalan, Nowshera Khurd)	Male	75
10	Mohmandzai	Nowshera (Nowshera Kalan, Nowshera Khurd)	Female	75
11	Khattak	Nowshera (Akora Khattak, Sar Derai)	Male	75
12	Khattak	Nowshera (Hakim Abad, Manki Sharif)	Female	75

Mandible

Shoveling anterior, distal accessory ridges, premolar accessory cusps, premolar groove, anterior fovea, Y groove pattern, major cusps number, deflecting, wrinkles, protostylid, cusp 5 (Hypoconulid), cusp 6 (Entoconulid), metaconulid, lingual cusp fusion.

Statistical analysis

The data were analyzed by using Hierarchical Cluster Analysis with Ward's Linkage.

Table II. Particulars of the maximum number of samples possible to score from prehistoric samples from Central Asia, Indus Valley and Peninsular India.

S. No	Sample	Abb.	N (max) 1
1	Chl. Mehrgar	ChlMRG	25
2	Djarkutan	DJR	39
3	Harappa	HAR	33
4	Inamgaon	INM	41
5	Kuzali	KUZ	24
6	Molali	MOL	41
7	Neo. Mehrgarh	NeoMRG	49
8	Sapalli Tepe	SAP	43
9	Sara i Khola	SKH	15
10	Timargarha	TMG	25
Total			335

RESULTS*Dental analyses*

District Peshawar and Nowshera individuals were used for the collection of dental samples and were compared with ancient regional populations (Table II). The data were analyzed and compared hierarchical cluster analysis with Ward's method, and compatibility model

possessing mean measure of divergence (MMD) (Table III). The results provide the following description of living and ancient people of the region (Fig. 1).

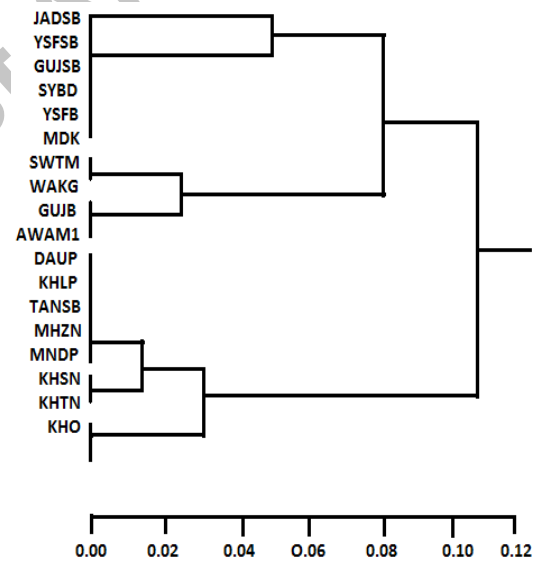


Fig. 1. All living samples of Northern Pakistan through cluster tree presenting split between Khattak (KHTn), Kheshgi (KHSn), Mohammadai (MHZn), Mohmand (MNDp), Khalil (KHLp) and Daudzai (DAUp).

Phenetic affinities

Compatibility model possessing mean measure of divergence (MMD) (Table III) describing phenetic affinities possessed by all the selected tribes of Peshawar and Nowshera and their standardized pair wise distances are presented in the table. The remaining analyses were identified by the values which were obtained from the compatibility model (Table III). Values nearer to zero

or below zero shows close relationship and connection between the discussed samples.

Table III. Compatibility model describing comparison and analysis of the living population as well as extinct group of Pakistan, Peninsular India and Central Asia through average measure of divergence.

AWAm1	DAUp	GUJb	GUJsb	JADsb	KHLp	KHTn	KHSn	KHO	MDK	MHZn	MNDp	SWTm	SYDb	SYDsb	TANsb
0.0000															
0.0000	0.0000														
0.0048	0.0000	0.0000													
0.0411	0.0368	0.0363	0.0410												
0.0000	0.0000	0.0000	0.0000	0.0408											
0.0052	0.0000	0.0000	0.0083	0.0745	0.0000										
0.0110	0.0000	0.0000	0.0200	0.0508	0.0000	0.0000									
0.0275	0.0000	0.0065	0.0210	0.0713	0.0077	0.0000	0.0000								
0.0043	0.0129	0.0143	0.0153	0.0401	0.0131	0.0367	0.0421	0.0513							
0.0114	0.0000	0.0088	0.0268	0.0419	0.0000	0.0000	0.0000	0.0040	0.0485						
0.0002	0.0000	0.0000	0.0091	0.0539	0.0000	0.0000	0.0000	0.0000	0.0245	0.0000					
0.0028	0.0095	0.0114	0.0115	0.0113	0.0108	0.0411	0.0382	0.0557	0.0000	0.0375	0.0235				
0.0000	0.0000	0.0000	0.0000	0.0092	0.0000	0.0033	0.0000	0.0170	0.0143	0.0000	0.0014	0.0029			
0.0063	0.0000	0.0000	0.0000	0.0120	0.0000	0.0078	0.0000	0.0028	0.0045	0.0116	0.0000	0.0000	0.0000		
0.0000	0.0000	0.0018	0.0076	0.0105	0.0000	0.0164	0.0024	0.0254	0.0233	0.0000	0.0099	0.0068	0.0000	0.0036	
0.0000	0.0025	0.0128	0.0159	0.0283	0.0022	0.0376	0.0245	0.0442	0.0048	0.0369	0.0202	0.0027	0.0058	0.0040	0.0000
0.0085	0.0129	0.0253	0.0381	0.0517	0.0147	0.0476	0.0292	0.0585	0.0163	0.0461	0.0297	0.0196	0.0194	0.0232	0.0088
0.0094	0.0000	0.0000	0.0000	0.0000	0.0000	0.0059	0.0165	0.0134	0.0221	0.0167	0.0013	0.0111	0.0000	0.0000	0.0094
0.0199	0.0123	0.0000	0.0000	0.0264	0.0143	0.0303	0.0334	0.0361	0.0124	0.0449	0.0170	0.0040	0.0000	0.0000	0.0254
0.0522	0.0364	0.0352	0.0473	0.0513	0.0554	0.0533	0.0471	0.0355	0.0756	0.0370	0.0417	0.0486	0.0486	0.0198	0.0418
0.0508	0.0477	0.0455	0.0623	0.0486	0.0603	0.0596	0.0603	0.0469	0.0610	0.0439	0.0441	0.0415	0.0610	0.0298	0.0512
0.0376	0.0397	0.0560	0.0679	0.1249	0.0554	0.0495	0.0631	0.0425	0.0649	0.0570	0.0401	0.0655	0.0854	0.0544	0.0620
0.0169	0.0268	0.0353	0.0503	0.0828	0.0298	0.0534	0.0484	0.0209	0.0230	0.0621	0.0304	0.0254	0.0536	0.0219	0.0420
0.0207	0.0249	0.0425	0.0581	0.1015	0.0307	0.0438	0.0493	0.0321	0.0409	0.0500	0.0307	0.0447	0.0591	0.0437	0.0448
0.0335	0.0356	0.0370	0.0566	0.0388	0.0410	0.0635	0.0591	0.0349	0.0409	0.0520	0.0368	0.0188	0.0440	0.0204	0.0415
0.0886	0.0698	0.0460	0.0291	0.0984	0.0733	0.0967	0.1145	0.0778	0.0929	0.1105	0.0950	0.0743	0.0564	0.0318	0.0854
0.1296	0.0904	0.1002	0.0858	0.1453	0.1109	0.0783	0.0912	0.1112	0.2078	0.0554	0.1116	0.1715	0.0844	0.1310	0.0851
0.0219	0.0105	0.0346	0.0383	0.0424	0.0063	0.0734	0.0563	0.0265	0.0514	0.0578	0.0485	0.0249	0.0256	0.0292	0.0012
0.0272	0.0389	0.0557	0.0610	0.1276	0.0593	0.0530	0.0702	0.0415	0.0657	0.0683	0.0516	0.0736	0.0697	0.0884	0.0521
0.1247	0.0685	0.0952	0.0900	0.1181	0.0879	0.0802	0.0802	0.1231	0.1896	0.0481	0.1073	0.1648	0.0583	0.1129	0.0669
0.0756	0.0384	0.0385	0.0264	0.0651	0.0467	0.0553	0.0479	0.0547	0.1205	0.0368	0.0700	0.0900	0.0201	0.0437	0.0289
0.0905	0.1023	0.1117	0.0959	0.2076	0.1234	0.1251	0.1720	0.1094	0.1203	0.1576	0.1260	0.1229	0.1409	0.1432	0.1291
0.1148	0.0775	0.0733	0.0682	0.0996	0.0815	0.0759	0.0602	0.0870	0.1773	0.0409	0.0984	0.1405	0.0485	0.0829	0.0530
0.0821	0.0556	0.0827	0.1246	0.1500	0.0827	0.0679	0.0484	0.0799	0.1580	0.0374	0.0770	0.1499	0.0840	0.1224	0.0599
0.0000	0.0000	0.0000	0.0160	0.0945	0.0000	0.0000	0.0000	0.0009	0.0394	0.0095	0.0000	0.0555	0.0140	0.0126	0.0126

Hierarchical cluster analysis of living Northern Pakistanis only method wards

Hierarchical cluster analysis with Ward's method yields two primary aggregates of samples, each of which has two sub-aggregates. The most apparent finding is that the various Pathan groups do not aggregate together to the exclusion of non-Pathan groups. The first aggregate contains highland dwellers (MDK, WAKg, WAKs) and their affiliates (SWTm) as well as an array of Pathan groups, mostly from Swabi (JADsb, YSFsb, GUJsb) but also a pair of Pathan samples from Buner (SYDb, YSFb) (Table IV). When examined by sub-aggregate, one of these encompasses all of the highland and highland-affiliated groups to the exclusion of all other samples. Further, the two Wakhi samples (WAKg, WAKs) are identified as showing nearest similarities to one another, as expected if

these represent two geographically discrete samples drawn from the same population (*i.e.*, ethnic group) (Table IV).

The second sub-aggregate identifies all of the Pathan samples as sharing close affiliated to one another, except for the Jadoons (JADsb), who are identified as a distant outlier. As expected, the two samples of both Sayeds (SYDb, SYDsb) and Yusafzais (YSFb, YSFsb) are identified as sharing close affinities to one another, respectively.

Table IV. Particulars of the samples taken under consideration from living Northern Pakistan.

S. No	Sample	Abb.	N (max)1
1	Awans	AWA2	93
2	Daudzai	DAUp	150
3	Gujar(Buner)	GUJb	66
4	Gujar(Swabi)	GUJsb	60
5	Jadoon(Swabi)	JADsb	80
6	Khalil	KHLp	148
7	Khattak	KHTn	160
8	Khesghi	KHSn	165
9	Khowars	KHO	144
10	Madaklasht	MDK	185
11	Mohammadzai	MHZn	140
12	Mohamand	MNDp	145
13	Swatis	SWT	178
14	Syed(Buner)	SYDb	70
15	Syed(Swabi)	SYDsb	86
16	Tanolis(Swabi)	TANsb	75
17	Wakhis(Gulmit)	WAKg	162.0
18	Wakhis(Sost)	WAKs	146.0
19	Yousafzais(Buner)	YSFb	66
20	Yousafzais(Swabi)	YSFsb	72
Total			2391

The second aggregate includes the Pathan samples from Peshawar and Nowshera Districts, but these samples are divided across the two sub-aggregates. Further, these Pathan samples are recognized as expressing nearer similarities to non-Pathan samples than to some other Pathan groups. Beginning with the first sub-aggregate, two of the Pathan samples from Peshawar (DAUp, KHLp) are recognized as expressing equally close similarities to Awans belonging to Mansehra area (AWAm1) and Buner District (GUJb), while Mohammadzais (MHZn) from Nowshera are identified as possessing close affinities to Tanolis from Swabi (TANsb). With regard to the fourth sub-aggregate, Mohmands from Peshawar (MNDp) are recognized as expressing close similarities to Khesghis (KHSn) and Khattaks (KHTn) from Nowshera, but also to Khows (KHO) from Chitral District (Fig. 1).

Prehistoric and living, central Asians, Pakistanis and peninsular Indians through hierarchical cluster method

Hierarchical cluster technique with Ward's procedure yields four aggregates in which one aggregate, composed of prehistoric samples from Central Asia, stand out as an outlier to all other samples included in the analysis. The remaining three aggregates may be described as:

1) Prehistoric and living samples from West Central Peninsular India (Table V), along with the Neolithic sample from Mehrgarh (Table II).

2) A mixed set of living samples from Nowshera and Swabi District, prehistoric samples from northern Pakistan and the Punjab, as well as the highland sample of Khowsand Wakhis.

3) A mixed set of samples from southern peninsular India, most of the Pathan and non-Pathan samples from Buner and Swabi Districts, along with the highland sample from Madak Lasht and the highland-affiliated sample of Swatis from District Mansehra. It is clear that the samples of prehistoric Central Asians from SapaliTepe and Djarkutan in Uzbekistan share close similarities with each other are dramatically identified from all South Asian specimens, both living and prehistoric, with one dramatic exception, Sarai Khola (SKH), the latest of the Indus Valley samples. Such results not only reveal the genetic separation of prehistoric Central Asians from prehistoric and living South Asians, but also suggest that if there was *any* introgression of Central Asian genes into South Asia, such introgression must have occurred long after the 2nd millennium BC as there is no affinity with the remains recovered from Timargarha (last 2nd of early 1st millennium BC) and the remains from SaraiKhola are believed to date to the last quarter of the 1st millennium BC. The remaining three aggregates are arranged in such a way that the specimen from West Central Peninsula of India (along with the earliest prehistoric Indus Valley sample from Neolithic Mehrgarh (NeoMRG) and the prehistoric sample from Inamgaon(INM) located in Maharashtra) stand as an outlier to the other two aggregates. Within this outlier, the three living samples belonging to Maharashtra (MDA, MRT, MHR) (Table V) share closer similarities to each other than to the two ancient samples. The finding of a phenetic link between Neolithic Mehrgarh to the Late Jorwe inhabitants of Inamgaon has been found in previous analyses and may reflect an early continent-wide indigenous population. The fact the living inhabitants of this region still show closet affinities to the prehistoric inhabitants of this region (INM) suggests longstanding population continuity in this region of peninsular India. Membership if the third aggregate is enigmatic and can be further divided into three sub aggregates. The 1st sub aggregate includes the two Wahki Samples (WAKg,

WAKs) and the late Chalcolithic era sample from Harappa located in the Punjab. This association makes no sense. Membership in the second sub-aggregate appears to include a random assortment of non-Pathan (TSANsb, AWAm1, GUJb) and Pathan samples (MHZn, KHLp, DAUp) (Table IV) as well as the belatedly Bronze Early Ironage specimens belonging to Timargarha (TMG), located in Lower Swat. Again, this association of samples doesn't make any sense. The remaining sub-aggregate is composed of three of the Pathan samples from Nowshera and Peshawar Districts (KHSn, KHTn, MNDp). This sub-aggregate makes some sense, as the association of these Pathan samples with the Khows of Chitral District was found when the analysis was limited to living northern Pakistani Samples. Membership of the fourth aggregate is intriguing and is better understood when considered by sub aggregate. The peripheral members of the fourth aggregate consist of the three Dravidian speaking ethnic groups from Andhra Pradesh in southeastern Peninsular India as has been found in previous studies, the middle-status Hindu caste Pakanati Reddis (PNT) stand apart from their low-status caste counterparts the Gompadhompoti Madigas (GPD) and non-Hindu tribal Chenchus (CHU). The next sub-aggregate is composed of Pathan (YSFb, SYDb, YSFsb) and non-Pathan samples (GUJsb) from Buner and Swabi Districts, along with two of the highland (MD) or highland affiliated groups (SWTm). The two outliers to this sub aggregate include the Jadoons from Swabi (JADsb) and the early Chalcolithic inhabitants of Mehrgarh distally (ChlMRG). Apart from the two living highland and highland-affiliated samples, as well as the prehistoric sample from Chalcolithic Mehrgarh, the remaining members of this aggregate are the same (except GUJb) as those identified consistently in the neighbor-joining and Multidimensional Scaling analyses of the living northern Pakistani data set.

Table V. Details of the maximum number of individuals possible to score from living Peninsular Indians.

S. No	Sample	Abb.	N (max)1
1	Chenchus	CHU	194
2	Gompad. Mad.	GPD	178
3	Madia Gonds	MDA	169
4	Mahars	MHR	195
5	Marathas	MRT	198
6	Pakanati Red	PNT	182
Total			1116

When considered overall, hierarchical cluster analysis

presents a generally confusing picture. Four aspects of the dendrogram make sense. The separation of prehistoric Central Asians from both living and prehistoric South Asians makes sense and has been identified in previous analyses. The grouping of the three living samples from West Central Peninsular India with prehistoric samples from this same region (INM) and from the earliest Indus Valley sample (NeoMRG) have also been identified in previous studies (Table III). The shared close affinities among the living ethnic groups from Southeast Peninsular India have likewise been previously identified. The general separation of Pathan and non-Pathan samples into separate aggregates dominated by samples from Buner and Swabi Districts versus Nowshera and Peshawar Districts also occurs when consideration is limited to living northern Pakistani ethnic groups.

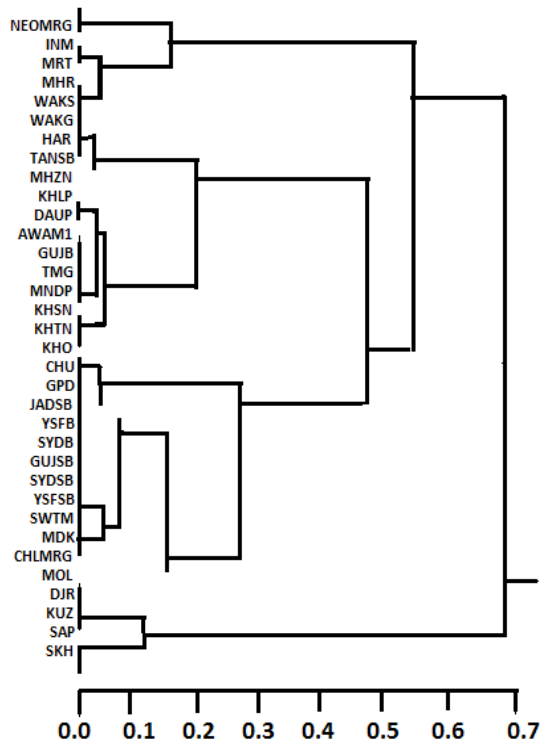


Fig. 2. All samples prehistoric and living central Asians, Pakistanis and peninsular Indians, through hierarchical cluster method.

Nevertheless, two other aspects do not make sense. The first is the association of Swatis from Mansehra District and the inhabitants of Madaklasht of Chitral District with an array of mainly Pathan ethnic groups from Buner and Swabi. This association was not identified when consideration was limited to living Pakistanis. Second, is

the association of the prehistoric sample from Harappa with the two Wakhi samples there is no apparent reason why this association should occur (Fig. 2).

DISCUSSION

Pakistan is known for its highest level of human genetic diversity. Pakistan is settled by several foreign populations and there have been some historic migrations into Pakistan. Pakistan is an ethnically diverse country with a similar variation in its natural resources and wildlife (Bellew, 1880; Rose, 1911). The region of South Asia consists of some sort of social miscellanies, ethnic variations, and prominent way of living patrimony (Bhasin, 2006). The history, race and cultural features of this area are till now to be discovered. Studies in relation to the early dwellers of this region are still in development in different aspects. It is intricate to fix the origin and settlement of these people in the pre-historic time; whether they were the defeater or the citizens of this locale. If these people were migrates, which path they used. However, the merger of new races and cultures through the coming of migrants with the passage of time has made the Indian inhabitants more compound and heterogeneous (Lukacs and Hemphill, 1991). A touch given to the history of this region can be seen by Sankalia (1962, 1979) and Allchin (1983). The areas possessing Pakistan have encountered with different inhabitants, cultures and have seen strange faces from the 4th century onwards for several centuries. The Greeks Parthian and Khushans, were the first to enter the area after the establishment of Aryan civilization (Thapar, 1969; Maloney, 1974). The Huns also stayed in a large number at the end of Gupta dynasty. Different people and cultures from the external of the region mixed with the hindus. The Jews and Parsis also settled on the west coast. The Arab people as well as Turks, and Afghanistanies made their direction towards south Asian landscape especially to the sub-continent. Although the Muslim colonization in the subcontinent though before the assaults of Arab in the parts of Sindh in earlier time of 7th century and blended the culture and art of the area. The Arab settling, establishment and integration with the localities ended with the establishment of Mughal Empire in the 16th century. It was this empire which brought changes to the structure and ethnicity of foreign population. Religion acceptance started and numerous numbers of Hindus converted to Islam. According to Mourant (1983) that people were involed in from the very ancient time and the pre human era. Pak-Indo seems to have adopted much part of their civilization from the following sources:

1st = The Balochistan and the Makran Coastline

2nd = The Assam and the Arakan Seaside (Bhasin,

2006; Fairservis, 1971).

The study conducted in Asian nation includes 2 districts of city Division viz. Peshawar and Nowshera, where the inhabitants have totally different teams, ethnicity, castes and tribes UN agency reside face by side for hundred or perhaps thousands of years in an exceedingly little geographical region, have typically preserved their distinct entities by active union very little attention has been paid to the biological proof of skeletal records and genetic distances, as existing within the ester sequences and mutations in genes occurred from time to time. As there's no proof concerning the movements and shifts of population within the inter-regional background, that the temporal depth and patterning of biological interactions stay principally explored. Genetic information of the Pakistani populations shows very little variation comparison to the opposite world populations. Earlier studies have claimed that Pakistan's population may be a single unit that is wrong, as a result of it consists of quite twelve totally different ethnic teams (Wilson-Wilde *et al.*, 1997; Hansen, 2000; Zhivotovsky *et al.*, 2001). The few studies that are on the market with relevancy Pakistani ethnic teams have exposed clear variations among them (Mansoor *et al.*, 1998; Mehdi *et al.*, 1999). The populations living at this time within the northern and southern elements of the country are often distinguished on the premise of those studies. The attention to the appearance and allotment of latest human beings was the vital reason of the researchers for several decades (Ingman *et al.*, 2000; Wolpoff *et al.*, 1997; Vigilant *et al.*, 1991).

In Pak-into ground the populace account of assorted grouping was extended beneath the critical eye (Kivisild *et al.*, 2003; Basu *et al.*, 2003; Chaubey *et al.*, 2007; Barnabas *et al.*, 2006). Present sight might even be whole into 3 totally different replica. The main might even be referred to as the Long-Standing permanence Model (Hemphill, 2013), that illustrate the basis of the fashionable populace of South Asia to the emergent dispersion of man some 62-75000 years past. The advocates of the replica declare that South-Asian inhabitants, once documented, are little littered with aspect consequence inhabitants adjacent to the sub continent or by any important population movements within the ground (Sahoo *et al.*, 2006; Krithika *et al.*, 2009; Kennedy *et al.*, 1984; Epperson, 1993; Manel *et al.*, 2003). Easy isolation by distance is management to be the aim as given among the design of biological affinities among ethnic groups that populations most connected in past and in geographic nearness are most alike geologically. Second replica usually noted as Aryan Invasion Model (Hemphill, 2013), flow on a chain of modernism that happended in the middle of the Bronze Age populace of Asian's central parts, similar to domestication

of the horse and so the improvement of the conflict chariot (Jarrige, 1984; Renfrew, 1987, 1996; Bryant, 2001). Indo-Aryan languages renowned by complete northern a pair of 3rd of ground, and metaphors of horse increased invaders winning the fortress of the snout less Dasusin the Rg sacred writing as evidence of an Asian assault into the ground all through the center of ordinal millennium B.C.

3rd replica is additionally named as a result of the Out of Asian nation Model. The sponsor of the model of presume that several hints of the primary materialization of sophisticated and agricultural towns and cities, every Indus geological formation andso North Asian nation's Doab (McAlpin, 1981), intend that the southren populace of Asia unfold external into joined regions of southwestern and Central Asia. The temporal arrangement of the partition couldn't be sustain by its supporters. In one of the reports, from that populace speaking the mentioed languages scattered all through the primary third millennium B.C. Among the 2nd account, the dispersion of languages is taken under consideration to be means later, all through the primary Iron time of the 1st millennium B.C.

Overall impressions

A consideration of the results obtained from the data reduction techniques yields six (6) important findings.

1. Regionally consistent aggregates are formed. These include: 1) highland and highland-affiliated groups (WAKg, WAKs, SWTm, MDK, AWAm1) (Table II). In all cases, the two geographically distinct samples of Wakhis were identified as possessing closest affinities to one another.
2. The Khows of Chitral District were found to not aggregate with other highland or highland-affiliated samples. Instead, they were consistently identified as sharing rather peripheral affinities to Pathan samples from Nowshera District, specifically Khattaks (KHTn), and Mohmands from Peshawar District (MHDp). Indeed, neighbor-joining cluster analysis identifies all three Pathan samples from Nowshera as being biologically more similar to Khows than are the three Pathan samples from Peshawar. Such results indicate that, despite close geographic proximity, the Khows do not share close biological affinities to the inhabitants of MadakLasht, but with Pathan groups from Nowshera.
3. Affinities between Pathan groups from Nowshera and Peshawar Districts are more diffuse than those between Pathan groups of Buner and Swabi Districts, but are marked by greater consistency with regard to geography. This is least apparent from the dendrogram yielded by hierarchical cluster analysis and is most evident from the dendrogram obtained with neighbor-joining cluster analysis. The former shows no geographic structure

- whatsoever, while the latter is marked by only one minor exception (Khalils of Peshawar District [KHLp]).
4. The sample of Jadoons from Swabi (JADsb) are consistently identified by all data reduction techniques as being phenetically isolated from all of the other samples included in this analysis, exhibiting only slightly closer affinities to Pathan and non-Pathan samples from Buner and Swabi Districts than to samples from other regions of northern Pakistan. Additional sampling is required to determine whether the Jadoons of Swabi District are true biological outliers due to genetic drift, founder effect, and/or population bottlenecking or whether their affinities are to be found with ethnic groups that have not, as yet, been sampled.
 5. The phenetic position of the Tanolis from Swabi District (TANsb) remains unresolved. Hierarchical cluster analysis identifies these Tanolis as possessing closest affinities to Mohammadzais from Nowshera District with secondary affinities to an array of Pathan and non-Pathan samples from Buner, Peshawar, and Mansehra Districts. In contrast, the dendrogram generated by neighbor-joining cluster analysis shows Tanolis from Swabi District as possessing closest affinities to Khalils from Peshawar (KHLp). Still further, multidimensional scaling, regardless of algorithm employed, identifies Tanolis of Swabi District as sharing closest affinities to Sayed from Buner District. Resolution of the phenetic position of these Tanolis requires additional samples of Tanolis from other districts to determine if this phenetic volatility is genuine or the consequence of sampling effect.
- Statement of conflict of interest*
The authors have declared no conflict of interest.
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