# Mitochondrial Haplogroup Analyses of the Madia Gond Tribe: Tracking the Past Events of Prehistory of India

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ABSTRACT The origin and spread of agriculture in India is poorly understood. To address this, we analyzed mtDNA sequences of some ethnic groups from Eurasia to check for the support for the two most prevalent models of agricultural diffusion that is, "Demic expansion" Vs "Cultural diffusion". Presence of haplogroup T1 in the Madia Gond and Baluch, and its exact identity with population inhabiting in and around Mediterranean region points towards Neolithic linkages between these populations. Further, occurrence of rare haplogroup N1a in one of the Baluch individual and its identity with 7500 old Neolithic farmer sequences from Europe further reinforce dispersal of human from the Fertile Crescent region. Our phylogenetic analysis indicates clustering of Madia Gond and Baluch sequences in the distinct T1 and N1a Neolthic clusters. Application of molecular clock to this cluster yields a time depth of ~10,000 which further reconfirms its Neolithic origin. Though upper Paleolithic origin of majority of Indian population is well established, but presence of T1 and N1a haplogroup in Madia Gond and Baluch respectively supports Neolithic Demic expansion model.

## **INTRODUCTION**

Even after their early exit from Africa (Quintana-Murci et al. 1999) ~50,000 years before present (ybp) human remained in hunting and food gathering stage till ~10,000 ybp. The civilization begins when humans started farming and became settled agriculturalist independently in the Middle East and in East Asia (Cavalli et al. 1994). Though farming in the Middle East and East Asia started ~10,000 ybp but the former expanded more widely to other parts of the world. Soon after humans adopted farming they resorted to cattle domestication in the Near East (Troy et al. 2001). Though the origin of agriculture in India is unclear (Vishnu-Mittre 1977) but Mehrgarh in Balochistan, Pa-

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kistan shows earliest sign of farming and animal domestication in the Indian subcontinent (Possehl 1996). There are genetic evidences of cattle domestication during the early Neolithic in the Indian subcontinent (Baig et al. 2005, Chen et al. 2010). Pottery, metallurgy were the other innovations that took place after the agriculture and animal domestication during the prehistory. These innovations spread to other parts of the world from their respective centers by the process called as diffusion. The method by which these innovations diffused across the world has always been a question of debate amongst archeologist. Some support "Demic expansion" model in which people from the land of innovation move en masse to other places along with their innovations (Ammerman and Cavalli-Sforza 1973). The other model "Cultural diffusion" favors spread of innovations mainly through trade and culture without involving en masse movement of people (Alinei 1991). Are farming and animal domestication brought to India by Demic expansion or by Cultural diffusion? As archeological data related to this is

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obscure, we analyzed mitochondrial DNA (mtDNA) sequences of Madia Gond tribe and one migrant Baluchi group from Eastern region of Maharashtra. The sequences from India were analyzed with those available in the GenBank using homology BLAST searching on (www. ncbi.nlm.nih.gov) site.

#### MATERIAL AND METHODS

#### Material/Sequence

Fourteen (14) Madia Gond and seven Baluch (07) samples previously sequenced for the mtDNA's D-loop and published (Baig et al. 2004) by us were retrieved from Gen Bank (Table 1). Properties like strict maternal inheritance (Giles et al. 1980), lack of recombination (Meriwether et al. 1991) and fast mutational rate (Brown et al. 1979) made mtDNA a molecule of choice in evolutionary genetics studies worldwide. Over the years mtDNA has become an efficient marker to analyze origin, genetic relationship and divergence of intra- or inter-mammalian populations (Brown et al. 1982; Giuffra et al. 2000). The ethnological information regarding group included in the study is as follows:

*Madia Gond:* The Gonds are mainly confined to central India and Eastern Maharashtra. They belong to Dravidian linguistic group and speak Gondi dialect. While most members of this tribe are now settled agriculturalists some groups are still found in hunting and food gathering stage and hence grouped under primitive tribe. The tribe is being more extensively studied by the social anthropologist. The biological information in general and molecular genetic information in particular about this tribe is scarce.

**Baluch:** The Baluch is a migrant group found in few cities of Maharashtra. They are more frequently called as Baluchi on the basis of known history of their migration from the Baluchistan region of Iran. They speak dialect belonging to Irani linguistic group which in turn belongs to Indo- European linguistic group. The Baluch belongs to Islamic religious group and are engaged in small businesses.

*Maragatos:* The Maragatos inhabit the western mountainous region of north Spain. Due to immigration their population in the region is now restricted to ~3000. The Maragatos follow strict endogamic custom and developed agriculture and established merchant activity throughout the Iberian Peninsula since the Middle Ages (Luengo A 1980). The Maragatos were included because of presence of second most common Neolithic T1 haplogroup in them and its homology with one of the Madia Gond sequence.

#### **Data Analysis**

The sequences were retrieved using BLAST homology searches on www.ncbi.nlm.nih.gov site. The sequences were aligned using Clustal X 1.81 (Thompson et al. 1997) with default parameters. The sequences (HVS-I positions 16024-16383) were aligned with revised Cambridge Reference Sequence (Andrew et al.1999). The relatedness amongst sequences was studied by constructing phylogenetic tree using Neighbor-joining method (Saitou and Nei 1987). The haplogroup age was estimated using  $\rho$  (rho) estimator. The  $\rho$  is the average transitional distance from the founder haplotype (Forster et al. 1996) with standard deviation described by Saillard et al. (2000).

## **RESULTS AND DISCUSSION**

As a global practice in mitochondrial DNA study, by combining HVS-I and restriction site polymorphism information, haplogroup status has been assigned to the sequences (Torroni et al. 1996). Our study is based on four sequences out of which three belong to T1 haplogroup and one to rare haplogroup N1a (Table 1). The T1 haplogroup is characterized by founder motif 126-163-186-189-294. Interestingly, we identified this T1 haplogroup in Madia Gond,

Table 1: Mutation relative to the Cambridge Reference Sequence (minus 1600) observed in the mtDNA HVS-I region along with their haplogroup designation in the study population.

Sequence name	HVS-I motif	Gen bank acc. no.	Hap- logr- oup
Madia Gond	126-163-186-189-294	AY208753	T1
Baluchi	93-126-163-186-189- 292-294-302	AY524681	T1
Baluchi	147G-172-223-248- 295-355	AY524683	N1a
Maragatos	126-163-186-189-294	(As reported by Larruga J et al. 2001)	T1

Baluch and Maragatos groups. Previous study reveals that T1 haplogroup is one of the most common Neolithic haplogroup identified after J and U3 (Larruga J et al. 2001).Our phylogenetic analysis shows splitting of sequences into T1 and N1a haplogroups (Fig. 1). Madia Gond and one Baluch sequence cluster tightly in the T1 haplogroup. The T1 haplogroup include sequences, ranging eastward from Tibet to those from Spain (Maragatos and Hispanic) from Western Europe. The T1 haplogroup also includes sequences from Pakistan, Middle East and two from genetically distinct ethnic groups from Europe namely Basque and Hispanic. Of note, this clustering is well supported by high bootstrap support (Fig. 1).Similarly, the haplogroup N1a, which is considered as rare haplogroup (Haak et al. 2005) comprises of Baluch, Serbian and Neolithic farmer sequence. Notably, clustering of 7500 old sequence from Neolithic farmer with Baluch confirms its Neolithic status. The phylogenetic tree was rooted with Neanderthal man sequence as out group. The haplogroup M is the most common haplogroup found in throughout India. The age estimate of ~50,000 ybp for the haplogroup M confirms early exit of modern human out of Africa. Similarly, the age estimation of haplogroup T1 using  $\rho$  (rho) statistics yields a time depth of  $12108 \pm 3200$  ybp. The time of ~12000 ybp confirms that this haplogroup originated during the Neolithic most probably in the Fertile Crescent region and spread to Europe and India along with the movement of Neolithic farmers. Additionally, the presence of N1a haplogroup in one of the Baluch re-inforces the presence of Neolithic lineages and probable route taken by the Neolithic farmer. Study by Haak et al. (2005) on 7500 years old bones shows that 25% of the Neolithic farmers of Europe were harboring N1a haplogroup. The radio carbon dating method confirms that these bones were part of Linear Pottery culture which is considered as the first farming culture in Central Europe (Hibben 1958). Though N1a was found in high frequency in the Neolithic remains but currently it appears in only 0.18 to 0.20 percent of regional population thus making it rare haplogroup. Presently, N1a haplogroup is found in Eurasian as well in Northern African population. Based on specific genetic marker it exhibits European, Central Asian, and African and South Asian division. The origin and migration pattern of this haplogroup is still an enigma and a matter of debate among archeologists and evolutionary biologists. The occurrence of single N1a out of seven samples indicates that this haplogroup may be found in high frequency in the Fertile Crescent region compare to other parts of the world. Thus, the presence of common and rare haplogroups T1 and N1a in the present day Madia Gond and Baluch population respectively supports Neolithic origin. Living Megalithic practices amongst Madia Gond are already being reported by some social anthropologists (Geetali 2002).

Based on this, we argue that some present day tribe in India like Madia Gond despite their upper Paleolithic origin show some relic of Neolithic diffusion and probable Mediterranean links.

### CONCLUSION

Although majority of present day Indian population shows its root in the upper Pale-

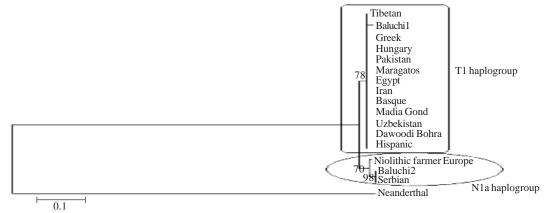


Fig. 1. Phylogenetic tree showing genetic relatedness of Baluch and Madia Gond sequences with global dataset

olithic, but the presence of Neolithic haplogroup T1 in the present day Madia Gond from India, Baluch from Baluchistan region of Iran, Maragatos from Iberian Peninsula supports eastward and westward movement of Neolithic farmers from the Fertile Crescent region. Further, the occurrence of rare haplogroup N1a in one Baluch individual reinforces possible Neolithic origin. Thus, besides "Cultural diffusion", the origin and spread of agro-pastoralistic societies by "Demic expansion" in the Indian subcontinent cannot be ruled out. In the future, vast sampling and probing by Y-chromosome and autosomal DNA markers of more tribes and caste population will cast light on the origin and diffusion of Neolithic lineages in India.

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