

Tracing the genetic history of the people living in the Rolwaling valley (East-Central Nepal)

In 2011, the Earth Mater Team led by Davide Peluzzi explored the remote Rolwaling Kumbu Valley located in the Eastern-Central part of Nepal, along the Tibetan border. During the scientific expedition called “Himalaya Remota: Rolwaling”, the explorers and researchers of ExPlora Nunaat, collected some buccal swab samples from the people inhabiting the area. Thanks to the collaboration between the ExPlora Nunaat group and the Laboratory of Molecular Anthropology of the University of Bologna (Italy), those samples have been processed to extract and analyse the whole genome DNA, with the aim to investigate the genetic variability and to reconstruct the genetic history of the people living in this remote valley of Nepal.

The Molecular Anthropology, is a scientific field aimed at reconstructing the genetic history of human population, by looking at the composition of its DNA. The study of DNA allows us to unravel the population history, because DNA contains and stores the genetic footprints (mutations) left by our ancestors, transferred from one generation to another. The DNA of a person is more than 99% the same as the DNA of any other unrelated person. But no two people have exactly the same DNA except the identical twins. Differences in DNA are called genetic variations or *mutations*. Therefore, by looking at the present and going back through the past, it is possible to reconstruct the genealogy of current living populations until a common genetic ancestor, by searching for specific patterns of mutations shared among individuals.

Because of the specific way of inheritance, two “portions” of human genome, have especially demonstrated to be extremely useful in population genetics study, to trace and reconstruct the history of human populations. These parts of our genome are also called “*uniparental markers*” and correspond respectively to Y-Chromosome and Mitochondrial DNA (mtDNA). The first one (Y-chromosome), it is present only in males and is therefore inherited on the *paternal side* from the father to sons. The second one (mtDNA) is instead inherited from the mother, along the *maternal side*. (Fig. 1)

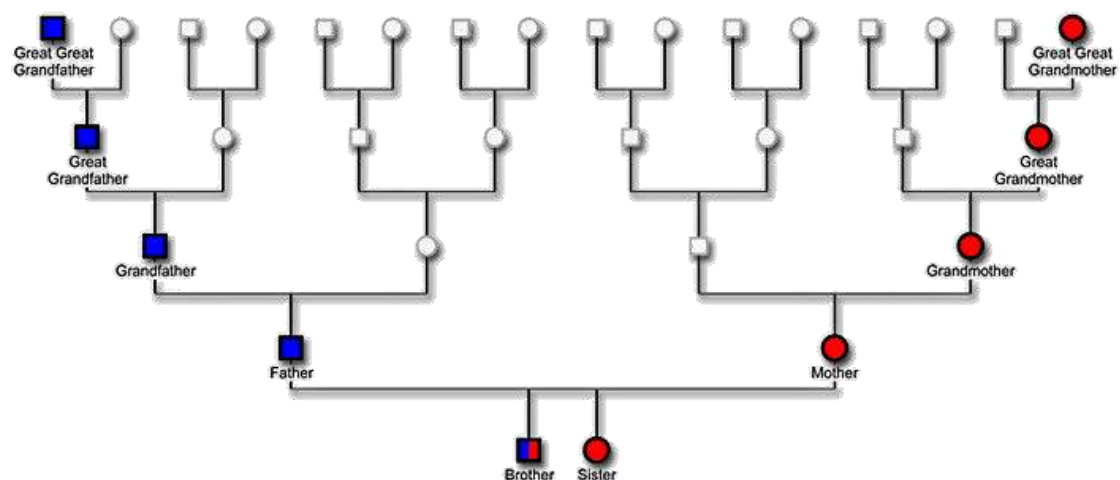


Fig 1. Y-line, paternal (blue) and mitochondrial, maternal (red)

These genetic markers can be used to trace paternal and maternal genealogical lines from the present to the distant past, because they are transmitted identical through generations despite for the occurrence of the genetic mutations. These occasional random mutations allow us to reconstruct the ancestry of a population because different population have a different combination of these variable sites. Therefore, by following the spread of the descendants of a specific ancestry, we can genetically retrace the patterns of human movements through the different geographic areas.

In the last years, many researches have been conducted on the Asia sub-continent, particularly involving the human populations neighbouring the region of Nepal. Several studies on the genetic variability of Tibetan, Chinese and Indian populations have been indeed published so far, whereas lower it seem to be the number of studies having specifically addressed the genetic history of Nepal. However, being located in the middle of the Asian sub-continent, moreover restricted on the north by the Himalayan mountain range, and bordering the "two ancient cultural giants of India and Tibet respectively" (Gayden, et al. 2007), the Nepalese population certainly represents a challenging case study in order to clarify the genetic origins of Himalayan populations. The study of the genetic history of Nepal indeed offers the opportunity 1) to understand the effects of a geographic barrier such as the Himalaya cordillera on the ancient and recent human dispersal routes through the continent and 2) to explore to what extent the cultural factors (such as language and/or religion) have influenced the genetic variability of the several ethnic groups currently residing in Nepal.

The more recent works on the genetic history of the Himalayan populations, have particularly analysed the genetic variability of uniparental markers (Y-Chromosome and Mitochondrial DNA) in two ethnic groups of Nepal, respectively the Tamang and the Newar, as well as in two general samples from Kathmandu and Tibet (Gayden, et al. 2007,2011,2013). These studies revealed a substantial contribution of Indian lineages in the Kathmandu sample and in the Newar Group of Tibeto-Burman origin. On the contrary, the Tamang (Tibeto-Burman origin) and the Tibetan samples exhibited a negligible or limited gene flow from India, instead showing almost exclusively genetic lineages of East-Asian origins.

In our laboratory, we have analysed the samples of 28 individuals living in the remote valley of Rolwaling (East-Central Nepal). The isolation of the valley, its high elevation and the proximity to the Tibetan border, makes particularly interesting try to reconstruct the genetic variability of this population. The samples analysed particularly consist of individuals belonging to three different ethnic groups: respectively Sherpa, Tamang and a third unspecified group with Indian influences. For both Y-chromosome (paternal side) and mitochondrial DNA (maternal side), we have analysed a set of specific markers characterised for having a high mutation rate. This means that these markers can help us to clarify the more recent events of population history (such as migration, isolation or founder effects).

Preliminary results highlight the presence of a high genetic variability in the Nepalese sample, which encompasses both different genetic lineages of East-Asian origin and, at a lower extent, also lineages with an Indian genetic background. These results are consistent with the historic rule of the region as an important ethnic and linguistic contact zone between populations of different genetic, cultural and linguistic matrices. (Fig 2, 3).

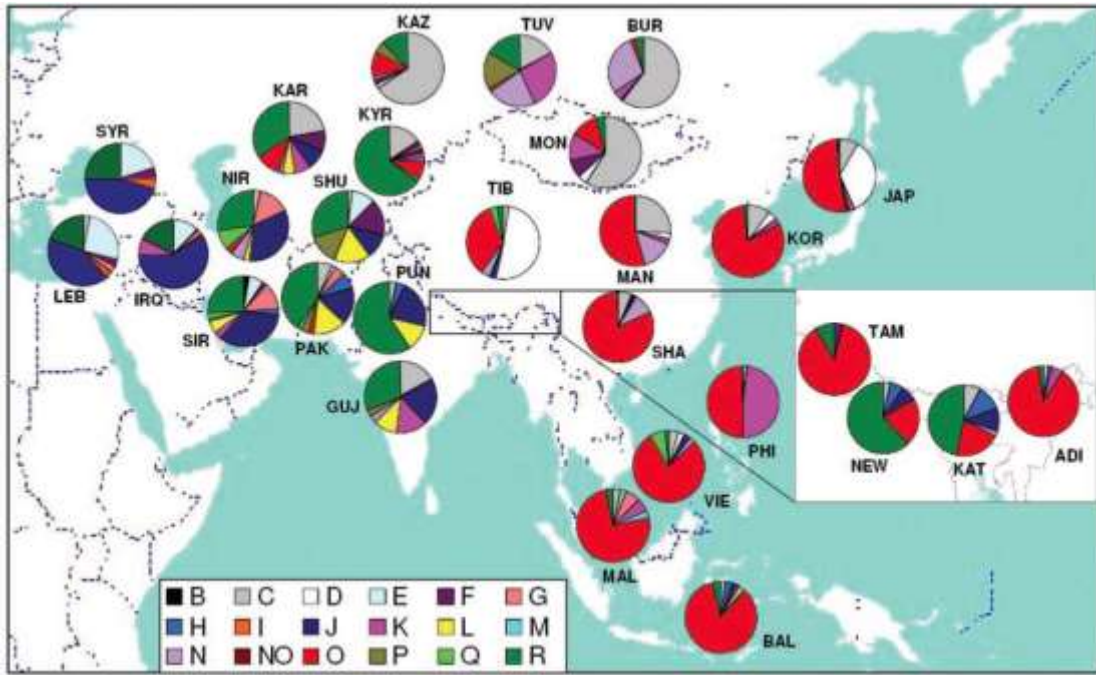


Fig 2. Geographic distributions of major Y-chromosome haplogroup frequencies in East Asia (Gayden et al. 2007)

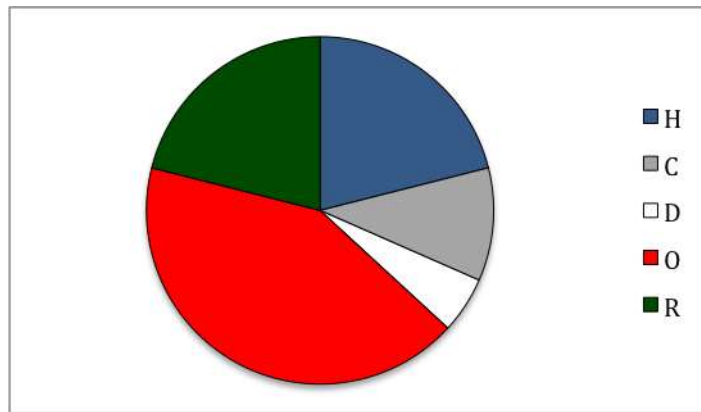


Fig 3. Y-chromosome haplogroup frequencies in **Rolwaling** samples

From the maternal point of view, the Nepalese population particularly reveals a closer genetic affinity with the Tibetan populations, and especially with those coming from the neighbouring Shigatse Prefecture (Zhao, et al. 2009) (Fig 4). From the paternal perspective, the Nepalese population, despite confirming the closer genetic relationship with the Tibetans, however reveals affinities also with the Indian-related groups of Kathmandu and Newar (Gayden, et al. 2007,2013) (Fig 5).

Consistently with previous studies (Gayden, et al. 2007,2011,2013; Wang, et al. 2012) the closer affinity between our Nepalese sample and the Tibetan population can be explained by the geographical proximity of Rolwaling valley to Tibet. However, at least partially different genetic histories and population dynamics can be hypothesised for males and females. Therefore the completion of the current analysis as well as the analysis of new collected samples, will promise to shed new lights into the genetic history of such an interesting population.

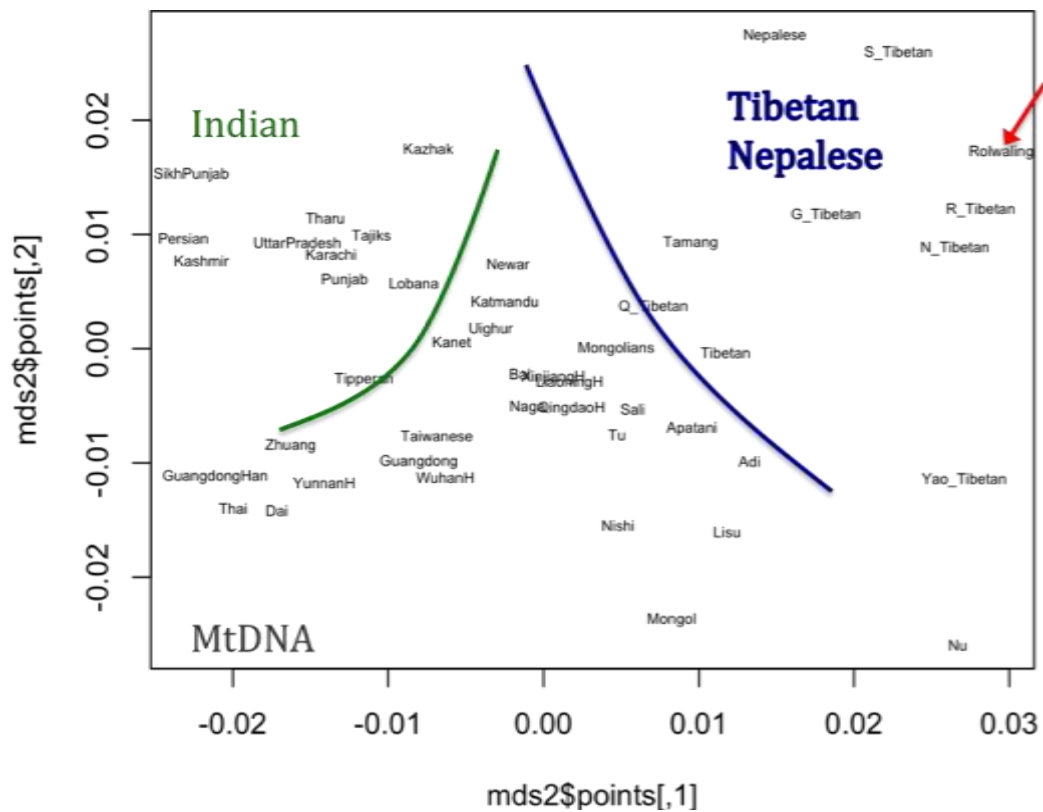


Fig 4. Mds plot showing genetic relationships between different Asian populations (Rolwaling samples indicated whit a red arrow) for the maternal line (Mitochondrial DNA). The two curves (green and blue) approximately separates the Indian populations (behind the green curve) from the Tibetan ones (behind the blue curve). The Rolwaling sample groups together whit the Tibetan populations, nearer to the one from Shigatse (R_Tibetan).

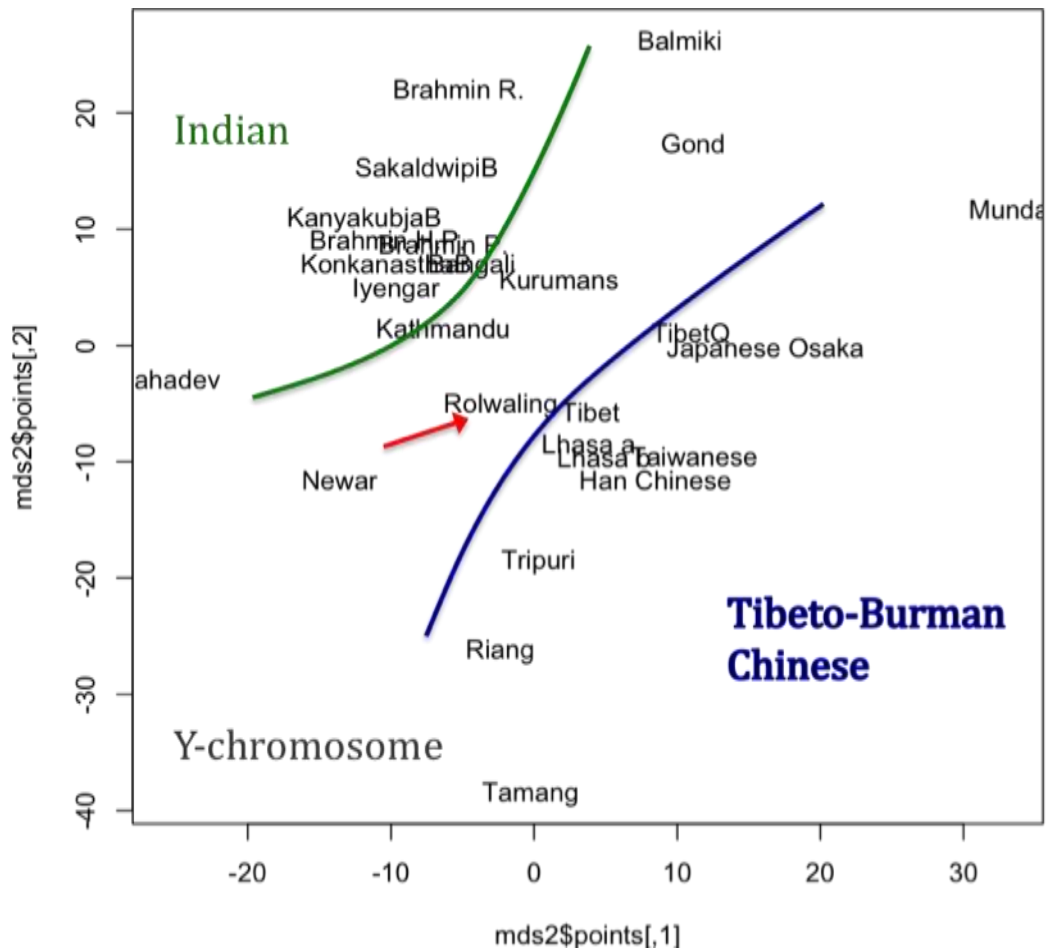


Fig 5. Mds plot showing genetic relationships between different Asian populations (Rolwaling samples indicated with a red arrow) for the paternal line (Y-Chromosome). The two curves (green and blue) approximately separates the Indian populations (behind the green curve) from the Tibeto-Burman and Chinese ones (behind the blue curve). The Rolwaling sample is localized between these two clusters, near to Tibetan populations as well as to other Nepalese samples (Newar and Kathmandu).