Genetic Analysis of Y-chromosomal STRs in Khandayat Population of Odisha, INDIA

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Abstract
This study was conducted to analyze haplotype diversity Khandayat Community of Odisha, INDIA using Y-chromosome specific short tandem repeat (Y-STR) loci. We collected blood samples from 106 unrelated healthy men of Khandayat Community and isolation of DNA was carried out by standard phenol-chloroform extraction method. Genomic DNA samples were amplified by multiplex-PCR using AmpFl STR Yfiler PCR Amplification Kit for 12 Y-STR loci followed by genotyping. A total of 97 haplotypes were identified among 106 individuals, out of which 93 were unique. Allelic frequency and gene diversity (GD) were calculated for each Y-STR locus. The resulting haplotype diversity (HD) was found to be 0.9798. Discrimination capacity (DC) was observed to be 0.9135. Haplotypes of Khandayat population of Odisha were compared with that of other Indian populations using AMOVA (Analysis of molecular variance) tool in order to measure the genetic relatedness between different populations.

Keywords: Y-STR, Haplotype diversity, Gene diversity, Khandayat population.

Introduction
Population genetics studies based on DNA polymorphism are being extensively used over last two decades aiming to analyze human evolutionary history and migration patterns. Microsatellites or short tandem repeats (STRs) are polymorphic DNA markers, which are used for studying genetic polymorphism. Y-chromosome is inherited paternally and Y-chromosome specific STRs are used in population and forensic genetics studies. Y-STRs have significant role in forensic science due to their use in solving sexual assault cases and cases of disputed paternity1-7.

Previous population genetics studies have revealed various diverse and heterogeneous population groups inhabiting in the Indian sub-continent8-22. Odisha is a coastal state located on the south-east region of India, which is inhabited by various population groups belonging to different strata of hierarchical caste systems like Brahmin, Khandayat, Karan and Gope. Khandayats, also known as Kshatriya, belong to an ancient warrior group and currently constitute over 35% of the state population. The current study was carried out with an aim to analyze the haplotype diversity of Khandayat population of Odisha using 12 Y-STR loci. The Y-STR loci included in the current study are DYS 19, DYS 389I, DYS 389II, DYS 390, DYS 391, DYS 392, DYS 393, DYS 385a/b, DYS 437, DYS 438, DYS 439 and DYS 448. Furthermore, the Y-STR haplotypes of Khandayats were compared with that of other Indian populations using AMOVA to find out the genetic relatedness.

Material and Methods
Whole blood samples were collected using standard procedure in 2ml EDTA vacutainers (BD Biosciences, NJ, USA) from 106 healthy unrelated male individuals of Khandayat Community, Odisha (India) stored at 4°C till further analysis. DNA extraction was carried out standard Organic (Phenol - Chloroform extraction) method23. The extracted DNA samples were quantified by spectrophotometer and quality of DNA was checked by agarose gel electrophoresis (0.8% agarose gel. The DNA samples were amplified in Thermal Cycler (PTC 200, MJ Research Inc., USA) using AmpFl STR Yfiler PCR Amplification Kit™ (Applied Biosystems, Foster City, CA, USA) for 12 Y-STR loci simultaneously by Multiplex PCR as per the manufacturer’s instructions24. The analyzed Y-STR loci include DYS 19, DYS 389I, DYS 389II, DYS 390, DYS 391, DYS 392, DYS 393, DYS 385a/b, DYS 437, DYS 438, DYS 439 and DYS 448. Amplicons were analyzed on ABI Prism 3130 xl Automated Genetic Analyzer (Applied Biosystems, Foster City, CA, USA). Allelic designations for different loci were obtained by GeneMapper ID software (v. 3.2).

Statistical Analysis: Allele frequencies were calculated by direct counting. Gene diversity (GD) and Haplotype diversity (HD) were calculated25.

Gene diversity (GD) was calculated using formula:

\[ GD = \frac{n}{n-1} \left[ 1 - \sum_{i=1}^{n} P_i^2 \right] \]
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Where, $P_i$ is the frequency of $i$th allele and $n$ is number of samples analyzed.

Haplotype diversity (HD) was calculated as:

$$HD = \frac{n}{n-1} \left[ 1 - \sum_{i=1}^{n} X_i^2 \right]$$

Where, $X_i$ represents haplotype frequency.

We used the online AMOVA tool for analysis of molecular variance\textsuperscript{26,27}. Haplotypes of Khandayat population (Odisha) were compared with 10 other Indian population samples with 873 haplotypes.

Following population samples were included in this study, Andhra Pradesh, India [Brahmin], population sample with 109 haplotypes, Himachal Pradesh, India [Saraswat Brahmin], population sample with 61 haplotypes, Jharkhand, India [Sakaldwipi Brahmin], population sample with 65 haplotypes, Karnataka, India [Brahmin], population sample with 103 haplotypes, Kashmir, India [Saraswat Brahmin], population sample with 58 haplotypes, Madhya Pradesh, India [Kanyakubja Brahmin], population sample with 78 haplotypes, Punjab, India [Saraswat Brahmin], population sample with 60 haplotypes, Rajasthan, India [Saraswat Brahmin], population sample with 60 haplotypes, Southern India, India [Tamil], population sample with 126 haplotypes, West Bengal, India [Rajbanshi], population sample with 39 haplotypes. Population pairwise distances between Khandayat population of Odisha and other Indian populations (Rst value) were calculated. Online AMOVA tool of YHRD (Y-chromosome haplotype reference database) was used for molecular variance analysis with 10,000 permutations for calculating $P$ value.

**Results and Discussion**

DNA samples of 106 healthy unrelated male individuals of Khandayat community of Odisha (India) were analyzed. A total of 97 haplotypes were identified among 106 individuals. The total number of alleles observed in this study was found to be 84. Maximum numbers of alleles were observed at the bi-allelic marker DYS385a/b. Allele frequency of Khandayat population of Odisha varied from 0.0088 to 0.7719. Gene diversity (GD) per locus ranged from 0.3659 to 0.9435 with mean GD value of 0.5683. The lowest gene diversity (0.3659) has been found at locus DYS391. The highest gene diversity (0.9635) has been found in case of the bi-allelic marker DYS385a/b.

A total of 97 different haplotypes were observed by combined analysis of 12 Y-STR loci in the studied samples, out of which 93 were unique. Haplotype diversity (HD) value for Khandayat population of Odisha was found to be 0.9798. Discrimination capacity (DC) value for the studied samples was calculated to be 0.9135.

Haplotypes of the Khandayats were compared with haplotypes of 10 other populations of India. Analysis of molecular variance pairwise distances based on Rst values between the Khandayats and other Indian populations are mentioned in Table-1. These observations revealed that, Khandayat population is distant from other Indian populations.

**Conclusion**

The observed results indicate that 12 Y-STR loci used in the current study are highly polymorphic among Khandayat community. Thus, this set of Y-STRs can be used for the forensic purposes like paternity testing, individual identification, genetic mapping etc. Hence this will add to the databank of various studies conducted on Indian population.

**References**


