

## Profiling of 17 Y-STR loci in Mazandaran and Gilan provinces of Iran

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**Background/aim:** The Y-chromosome mainly consists of heterochromatin regions that have a father-to-son inheritance. Short tandem repeat polymorphic (STRP) markers distributed all over the chromosome provide the opportunity for investigations in forensic medicine and ancestral lineage studies. Due to the existence of wide varieties of geographical and ethnic groups in Iran, studying Y-STRP markers is necessary for further applications. Here we investigated the provinces of Mazandaran and Gilan for the first time.

**Materials and methods:** Samples included 119 and 90 unrelated males from Mazandaran and Gilan, respectively. Using a PCR amplification kit, 17 Y-STRP markers were amplified and genotyping was conducted by capillary electrophoresis. Allele frequency, haplotype diversity (HD), and haplotype discrimination capacity (DC) were calculated. The populations were compared together and to neighboring countries including Afghanistan and Azerbaijan by  $F_{ST}$  index.

**Results:** A total of 204 unique haplotypes were observed. No uniqueness was observed between the two provinces. HD was 0.9993 and 0.9998 in Mazandaran and Gilan, respectively. DC was 0.9666 and 0.9888 for Mazandaran and Gilan, respectively. DYS385b and DYS391 had the most and least polymorphic content in both provinces, respectively. There was not a significant difference between these two provinces ( $F_{ST} = 0.0006$  and  $P = 0.00$ ) and neighboring countries.

**Conclusion:** The results highlight the effectiveness of these Y-STRP markers for male discrimination in the north of Iran. Using additional markers along with extended sample size would provide a better opportunity for removing matched haplotypes and introducing the best polymorphic markers in this specific population.

**Key words:** DYS385b, DYS391, haplotype diversity, Y-chromosome

### 1. Introduction

The Y-chromosome is highly heterochromatic consisting of repetitive DNA sequences, which makes sequencing difficult (1). It mainly carries genes involved in sex determination and spermatogenesis (2). During meiosis small regions at extreme ends of short and long arms of the Y chromosome, PARI and PARI, undergo recombination while other parts which contain about 95% of the Y-chromosome tend to be transmitted from father to son intact (3). This phenomenon makes the Y-chromosome interesting for studying male identification.

Short tandem repeat polymorphic (STRP) markers are a class of multiallele sequences distributed frequently and randomly all over the genomes of animals and plants.

They normally consist of 1–6 bp tandemly repeated units. STRP markers are variable in length among individuals except for monozygotic twins. They are mostly located in noncoding regions of the genome (4,5). The STRP markers have been widely used in medical genetics for tracking disease genes in Mendelian inherited disorders in families but not in mutation identification (6). When several STRP markers are used simultaneously following haplotype reconstruction, they have the ability to identify individuals. Then they are commonly utilized in forensic medicine for paternity testing, identification in sexual abuse cases, criminal subjects, and proving male kinship with the survivors in mass disasters (7,8). Furthermore these markers are useful for investigating migration and common ancestral lineage of a specific population (9).

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More than 3000 Y-STRP markers have been found up to date (10), which raises the need of finding informative markers in specific populations for forensic applications, and finding ancestral lineages. Iran, a large country with more than 80 million people, consists of several ethnic groups, and needs its own ethnic-specific databases for allele frequencies and heterogeneity of STRP markers. Although the eastern and central regions have been studied previously (11–14), there is not much information on Y-STRP markers and haplotype heterogeneity in the north of Iran. Therefore, in this project, we analyzed 17 Y-STRP markers in male populations in two northern populations of Iran via multiplex PCR and capillary electrophoresis following statistical analysis.

## 2. Materials and methods

The present investigation was approved by review boards and the ethics committee of Islamic Azad University of Rasht and the Iranian Legal Medicine Research Center. Interviewing the male clients and volunteers, blood samples from 119 unrelated subjects from Mazandaran Province and 90 unrelated cases from Guilan Province were obtained on Whatman FTA Classic cards after obtaining informed written consent. Subjects enrolled in the study included males who did not belong to Sadat ancestry and did not experience blood transfusion in the last 24 h. A punch of 1.2 mm<sup>2</sup> was used for DNA extraction according to the manufacturer's standard procedure.

Using the AmpFLSTR Yfiler PCR amplification kit (Applied Biosystems, Foster City, CA, USA) 17 STRP markers were amplified in a multiplex PCR reaction following the manufacturer's instruction. Characteristics of the markers are shown in Table 1. The PCR products were separated using capillary electrophoresis on an ABI 3500 genetic analyzer (Applied Biosystems, Foster City, CA, USA). The alleles were assigned by GeneMapper ID-X v.1.3 software (Applied Biosystems, Foster City, CA, USA). Alleles have been named following the update of the recommendation of the DNA Commission of the International Society of Forensic Genetics (15). Allele frequency for each locus and haplotype frequency were calculated by simple gene counting method. Gene and haplotype diversity (HD) was calculated according

to Nei's formula ( $h = \frac{N}{N-1(1 - \sum P_i^2)}$ ) using the

Arlequin Software package v3.5.2.2 and GenAEx 6.5 software. Haplotype match probability (HMP) was calculated according to  $HMP = 1 - HD$  formula. Haplotype discrimination capacity (DC) was calculated by dividing the total number of unique haplotypes in the analyzed population by the total number of individuals in the sample size. Genetic distance between these two provinces

and neighboring countries, Afghanistan and Azerbaijan, were calculated via  $F_{ST}$  value.

## 3. Results

In this investigation, the genotype of 17 STRP markers was determined by the multiplex PCR followed by capillary electrophoresis in 119 males from Mazandaran and 90 males from Gilan. All loci were successfully amplified in both populations. Observations displayed a total of 204 unique haplotypes in both provinces. We found 115 unique haplotypes in Mazandaran, none of which matched the haplotypes found in Gilan (Table 2). Haplotype reconstruction unraveled 89 unique haplotypes in Mazandaran without any similarity to those seen in Gilan (Table 3). Unique haplotypes had frequencies of 0.0083 and 0.0111 in Mazandaran and Gilan, respectively. Haplotypes were submitted to the Y-Chromosome Haplotype Reference Database (YHRD: [www.yhrd.org](http://www.yhrd.org)) and are available with the accession number YA004258 for Mazandaran and YA004257 Gilan. HD was 0.9993 (HMP = 0.0007) and 0.9998 (HMP = 0.0002) in Mazandaran and Gilan, respectively. We obtained DC = 0.9663 in Mazandaran and DC = 0.9888 in Gilan. The genetic distance between these two provinces was 0.003. There was a significant difference between Mazandaran and Gilan ( $F_{ST} = 0.0006$ ,  $P = 0.00$ ). We compared Mazandaran Province with available data from neighboring countries, Afghanistan and Azerbaijan, the statistical analysis of which revealed  $F_{ST} = 0.003$ ,  $P = 0.00$  and  $F_{ST} = 0.0086$ ,  $P = 0.00$ , respectively. These statistics were  $F_{ST} = 0.0026$ ,  $P = 0.00$  and  $F_{ST} = 0.0082$ ,  $P = 0.00$  for Gilan Province in comparison with Afghanistan and Azerbaijan, respectively. The STRP markers showed a genetic diversity ranging from 0.480 to 0.863 in Mazandaran. Genetic diversity ranged from 0.379 to 0.824 in Gilan. DYS389b had the most polymorphic content in both provinces while DYS391 was the least polymorphic STRP marker (Table 4). Allele frequency per marker in both populations is exhibited in Table 5. The mean gene diversity in Mazandaran and Gilan was 0.665 and 0.683, respectively.

## 4. Discussion

Studying Y-STRP markers in specific populations and geographical regions is valuable for identifying ancestral lineage and forensic applications. In the present study, we aimed to study 17 Y-STRP markers in two northern provinces of Iran, Mazandaran and Gilan, using STRP markers. Here we found 204 unique haplotypes and introduced DYS385b as the most polymorphic marker in both populations. No shared haplotype was observed between these two provinces. DYS391 had the least polymorphic content in both populations. Comparing these two populations together and with neighboring countries showed low genetic differences.

**Table 1.** Characteristic of Y-STRP markers used in this investigation.

marker STRP	Repeat motif	Allele range
DYS456	AGAT	13–18
DYS389 I	(TCTG) <sub>3</sub> (TCTA) <sub>n</sub>	9–17
DYS390	(TCTA) <sub>2</sub> (TCTG) <sub>n</sub> (TCTA) <sub>n</sub> (TCTG) <sub>n</sub> TCA(TCTA) <sub>2</sub>	17–28
DYS389 II	(TCTG) <sub>n</sub> (TCTA) <sub>Nn</sub> 28(TCTG) <sub>2</sub> (TCTA) <sub>n</sub>	23–34
DYS458	GAAA	14–20
DYS19 / DYS394	(TAGA) <sub>3</sub> TAGG(TAGA) <sub>n</sub>	10–19
DYS385a/b	GAAA	7–25
DYS393	AGAT	8–16
DYS391	(TCTG) <sub>3</sub> (TCTG) <sub>n</sub>	6–14
DYS439	AGAT	8–15
DYS635	TSTA	20–26
DYS392	(TAT) <sub>n</sub>	7–18
Y GATA H <sub>4</sub>	TAGA	8–13
DYS437	TCTR	13–17
DYS438	TTTTTC	8–13
DYS448	AGAGAT	17–27

**Table 2.** Haplotype structure of Y-STRP markers in 119 unrelated men in Mazandaran.

Sample	Locus	DYS 19	DYS 389I	DYS 389II	DYS 390	DYS 391	DYS 392	DYS 393	DYS 385a	DYS 385b	DYS 438	DYS 439	DYS 437	DYS 448	DYS 456	DYS 458	DYS 635	GATA H4
	Pop																	
1	Mazandaran	16	13	30	25	11	11	13	11	14	11	10	14	20	15	16	23	12
2	Mazandaran	14	13	29	23	10	11	12	13	16	9	12	15	21	15	17	21	11
3	Mazandaran	14	13	27	23	10	11	12	15	17	9	12	14	23	16	15	22	12
4	Mazandaran	15	13	30	22	11	11	14	12	14	10	13	15	22	14	17	21	11
5	Mazandaran	14	13	29	24	9	11	12	13	16	10	12	14	20	15	18	21	10
6	Mazandaran	14	13	30	23	10	11	12	13	18	10	11	14	20	14	19	22	11
7	Mazandaran	15	14	32	22	9	11	12	13	16	9	11	15	19	15	17	23	11
8	Mazandaran	14	13	31	23	11	11	12	13	19	10	12	14	20	14	18	20	12
9	Mazandaran	15	13	29	24	11	13	14	15	20	10	11	14	20	15	17	22	11
10	Mazandaran	15	14	32	23	10	12	13	13	16	10	11	14	20	13	14	22	12
11	Mazandaran	14	14	31	22	10	11	13	14	14	10	11	16	18	15	18	21	12
12	Mazandaran	14	14	30	23	10	11	12	13	16	9	11	15	21	15	16	21	11
13	Mazandaran	15	12	28	23	10	13	12	11	14	10	11	14	20	15	19	25	11
14	Mazandaran	14	13	29	24	11	13	12	11	11	13	13	14	19	15	18	23	13
15	Mazandaran	13	14	30	22	10	15	13	12	17	8	11	15	19	16	18	24	10
16	Mazandaran	15	13	29	22	10	11	13	14	16	9	11	15	21	15	16	21	11
17	Mazandaran	14	14	30	23	10	11	12	12	18	10	11	14	21	15	19	21	11
18	Mazandaran	15	13	31	23	11	11	12	13	15	10	11	15	20	16	17	24	10
19	Mazandaran	14	13	29	22	10	11	12	14	16	10	12	14	21	15	15	21	11
20	Mazandaran	15	13	31	23	11	11	12	13	19	9	11	15	20	15	18	21	11
21	Mazandaran	14	13	29	24	10	13	12	12	15	12	12	15	19	15	18	23	12
22	Mazandaran	14	13	30	23	10	11	12	12	17	10	11	14	20	15	19	20	11
23	Mazandaran	14	13	31	23	10	11	12	12	18	10	11	14	20	15	17	20	11
24	Mazandaran	15	12	28	23	10	11	15	12	14	10	12	16	19	13	16	21	12
25	Mazandaran	14	13	29	23	10	11	12	12	17	9	12	14	20	15	15	21	12

Table 2. (Continued).

26	Mazandaran	14	13	29	24	10	13	12	12	15	12	12	15	19	15	18	23	12
27	Mazandaran	14	15	31	23	10	11	12	14	16	9	11	15	21	15	16	21	11
28	Mazandaran	14	13	29	24	10	13	12	11	14	12	12	15	19	15	16	23	13
29	Mazandaran	15	14	31	23	10	10	14	14	16	11	13	14	19	15	19	25	11
30	Mazandaran	14	12	29	23	11	12	13	15	15	10	12	16	21	15	16	22	10
31	Mazandaran	14	13	30	24	11	13	12	11	14	13	13	15	20	15	16	23	13
32	Mazandaran	14	13	29	23	10	11	12	12	17	9	11	15	21	15	17	21	11
33	Mazandaran	14	13	30	23	11	13	12	11	14	12	12	15	20	17	16	23	12
34	Mazandaran	13	13	29	22	8	11	12	17	17	9	11	15	21	14	16	23	11
35	Mazandaran	16	13	30	24	11	11	13	11	14	11	10	14	20	16	15	24	12
36	Mazandaran	15	13	30	25	11	11	12	14	17	9	11	15	22	15	15	21	12
37	Mazandaran	14	13	29	23	10	11	12	14	17	9	11	14	19	15	16	21	11
38	Mazandaran	13	14	30	22	10	13	12	16	18	9	12	14	19	15	15	20	11
39	Mazandaran	14	13	28	25	11	13	12	10	16	12	12	15	20	16	16	24	11
40	Mazandaran	14	14	20	31	10	10	14	16	17	11	13	14	19	15	14	25	11
41	Mazandaran	14	13	29	23	10	11	12	13	16	9	11	15	22	15	16	21	11
42	Mazandaran	15	13	29	24	10	14	13	12	14	11	12	14	18	14	14	24	12
43	Mazandaran	13	13	28	22	10	13	12	14	16	9	12	15	19	15	15	21	12
44	Mazandaran	14	14	30	23	10	11	13	15	17	9	12	15	19	16	16	23	12
45	Mazandaran	14	14	30	23	10	11	12	14	17	9	11	14	19	15	16	21	11
46	Mazandaran	14	13	28	23	10	11	13	14	16	9	12	15	11	16	17	22	11
47	Mazandaran	15	14	31	23	10	14	11	12	17	10	12	16	19	15	17	21	11
48	Mazandaran	13	14	31	22	10	11	13	12	15	9	12	14	21	15	15	21	11
49	Mazandaran	15	13	30	23	11	11	12	13	16	9	12	14	21	15	14	22	12
50	Mazandaran	14	13	29	24	11	13	12	11	14	12	12	15	18	15	15	23	12
51	Mazandaran	14	13	30	23	10	11	12	13	18	10	11	14	20	14	15	21	11
52	Mazandaran	14	13	29	23	10	11	12	14	17	9	12	14	21	15	15	23	12
53	Mazandaran	14	13	29	24	10	11	12	14	14	9	11	15	21	15	16	23	11
54	Mazandaran	15	13	29	23	9	11	12	13	16	9	12	14	21	17	15	22	12
55	Mazandaran	14	13	30	24	11	13	12	11	15	12	12	15	19	15	17	23	12
56	Mazandaran	14	13	29	24	11	13	12	11	14	12	13	15	19	16	16	23	11
57	Mazandaran	14	13	29	24	11	13	12	11	11	12	14	14	19	15	15	23	13
58	Mazandaran	15	13	28	23	10	10	14	14	17	11	11	15	19	13	17	25	12
59	Mazandaran	16	13	30	22	9	12	13	15	16	10	11	15	19	17	15	21	12
60	Mazandaran	15	14	32	24	10	13	13	12	12	11	12	15	21	16	16	23	12
61	Mazandaran	14	13	29	23	11	11	12	13	17	10	11	15	21	14	15	21	12
62	Mazandaran	15	13	28	23	10	11	12	15	17	9	12	14	19	17	17	20	11
63	Mazandaran	17	13	32	25	10	11	13	11	14	11	11	14	20	16	15	23	13
64	Mazandaran	14	13	30	23	10	11	12	13	15	10	11	14	19	14	18	21	12
65	Mazandaran	16	13	31	25	11	11	13	11	14	11	10	15	20	16	15	23	13
66	Mazandaran	16	13	30	23	11	11	13	11	14	11	10	14	20	15	16	23	12
67	Mazandaran	15	14	31	23	10	10	13	12	20	12	11	15	20	15	17	25	12
68	Mazandaran	14	13	30	23	10	11	12	11	15	9	11	14	19	14	18	21	12
69	Mazandaran	14	13	29	24	11	13	12	12	14	12	13	15	19	16	16	23	11
70	Mazandaran	16	14	22	23	10	11	12	13	16	9	11	14	18	15	16	25	11
71	Mazandaran	14	13	31	23	10	11	12	13	19	10	11	14	20	14	18	20	11
72	Mazandaran	14	14	30	23	10	11	14	13	13	9	13	15	17	14	14	20	11
73	Mazandaran	13	13	32	24	10	11	13	15	16	11	12	14	20	15	17	21	12
74	Mazandaran	15	14	30	23	11	13	12	12	14	12	12	15	20	15	16	23	11
75	Mazandaran	15	13	30	22	10	11	13	13	14	10	12	16	22	15	19	21	11
76	Mazandaran	13	14	30	24	10	11	12	11	18	10	13	14	21	15	19	12	11
77	Mazandaran	15	12	28	23	10	11	14	12	15	10	11	15	21	16	17	21	12
78	Mazandaran	16	14	29	23	10	15	11	12	18	10	11	15	19	15	15	24	11
79	Mazandaran	16	14	32	26	11	11	13	11	14	11	11	14	19	16	16	23	14

Table 2. (Continued).

80	Mazandaran	14	12	28	22	10	10	14	13	14	10	14	16	22	14	16	25	12
81	Mazandaran	14	13	29	23	10	11	12	13	16	9	11	15	21	15	17	20	11
82	Mazandaran	16	14	32	25	11	11	13	11	14	11	11	14	19	15	15	23	13
83	Mazandaran	16	14	32	26	11	11	13	11	14	11	11	14	19	16	16	23	14
84	Mazandaran	15	13	29	24	9	11	12	16	16	10	11	14	20	10	18	23	10
85	Mazandaran	16	12	29	22	10	10	16	14	16	11	11	16	21	14	17	20	12
86	Mazandaran	14	13	27	23	10	11	12	13	18	10	12	14	19	14	17	20	11
87	Mazandaran	14	13	29	24	10	13	12	11	13	12	12	15	19	15	18	23	13
88	Mazandaran	14	13	29	23	11	11	12	12	16	9	12	15	21	15	16	23	11
89	Mazandaran	14	13	31	23	11	11	12	13	19	10	11	14	20	14	18	21	11
90	Mazandaran	14	13	29	24	10	11	12	14	16	9	12	16	21	15	16	21	11
91	Mazandaran	17	13	32	25	10	11	13	11	14	11	11	14	20	16	15	23	13
92	Mazandaran	14	12	28	22	10	11	14	13	13	10	11	16	20	14	16	21	11
93	Mazandaran	14	13	31	23	10	11	12	14	21	9	11	15	19	15	16	21	13
94	Mazandaran	14	11	27	23	10	11	12	14	19	9	12	14	21	16	19	22	11
95	Mazandaran	14	13	29	23	10	11	12	13	14	10	11	15	19	15	16	21	12
96	Mazandaran	16	13	29	23	9	12	12	14	16	9	12	14	21	15	14	22	11
97	Mazandaran	14	14	29	23	10	11	12	13	17	9	11	14	19	15	16	21	11
98	Mazandaran	14	13	29	24	11	13	12	11	14	12	12	15	19	15	16	24	11
99	Mazandaran	14	13	31	23	10	12	13	15	15	10	12	16	21	15	15	21	11
100	Mazandaran	13	12	30	23	10	11	14	16	17	10	11	14	20	15	16	22	13
101	Mazandaran	15	13	29	24	10	11	12	14	17	9	12	15	18	14	16	21	12
102	Mazandaran	13	13	29	22	8	11	12	17	17	9	11	15	21	14	15	23	11
103	Mazandaran	14	13	29	24	10	13	12	12	15	12	12	15	19	15	18	23	12
104	Mazandaran	15	13	28	24	10	11	15	12	14	10	12	16	21	15	16	21	13
105	Mazandaran	14	13	29	24	10	11	13	13	18	9	12	15	21	15	16	22	11
106	Mazandaran	14	14	30	23	10	11	13	15	17	9	12	15	20	16	16	22	12
107	Mazandaran	15	12	28	22	10	11	14	12	15	10	11	16	21	16	17	22	12
108	Mazandaran	14	14	30	23	10	11	12	14	17	9	11	14	19	15	16	21	11
109	Mazandaran	15	14	31	23	10	10	14	12	20	12	11	15	20	15	17	25	12
110	Mazandaran	14	13	30	23	10	11	12	13	15	9	11	14	19	14	18	22	12
111	Mazandaran	16	12	29	22	10	10	14	15	16	10	12	14	21	15	17	21	12
112	Mazandaran	15	12	29	23	10	11	12	12	15	9	11	14	21	15	15	24	13
113	Mazandaran	14	12	29	23	10	12	13	15	15	10	12	16	21	16	16	24	11
114	Mazandaran	15	12	28	22	10	11	15	12	15	10	13	16	20	16	16	21	12
115	Mazandaran	14	13	30	22	10	11	12	12	16	9	12	14	20	15	15	23	11
116	Mazandaran	16	13	30	24	11	11	13	11	15	11	10	14	20	15	15	23	13
117	Mazandaran	15	12	30	22	11	11	13	13	15	10	12	15	21	15	16	23	12
118	Mazandaran	15	12	29	21	10	11	14	12	15	10	12	16	21	15	17	23	12
119	Mazandaran	15	12	29	21	10	11	14	12	15	10	12	16	21	15	17	22	12

Table 3. Haplotype structure of Y-STRP markers in 90 unrelated men in Gilan.

Sample	Locus	DYS 19	DYS 389I	DYS 389II	DYS 390	DYS 391	DYS 392	DYS 393	DYS 385a	DYS 385b	DYS 438	DYS 439	DYS 437	DYS 448	DYS 456	DYS 458	DYS 635	GATA H4
	Pop																	
1	Gilan	14	13	29	23	10	11	12	13	17	9	12	14	21	17	15	21	11
2	Gilan	15	12	28	22	10	11	13	13	14	10	9	16	23	14	19	20	13
3	Gilan	15	13	29	22	10	13	13	14	16	9	11	14	19	15	17	21	11
4	Gilan	16	13	28	22	10	11	13	12	18	10	11	14	19	15	17	20	10
5	Gilan	15	13	31	24	11	11	13	14	15	10	13	15	20	15	16	23	11
6	Gilan	14	14	29	24	10	14	12	11	15	12	13	15	20	16	16	23	13
7	Gilan	14	14	30	24	11	13	12	12	14	12	12	15	19	16	16	23	12
8	Gilan	15	13	28	23	9	12	12	13	16	12	12	15	20	14	14	23	13

Table 3. (Continued).

9	Gilan	14	13	29	25	10	13	12	14	14	12	11	15	20	16	16	23	13
10	Gilan	14	12	29	24	10	11	12	13	14	9	13	15	19	14	17	21	11
11	Gilan	15	13	30	22	10	11	13	14	14	11	12	16	18	14	17	21	11
12	Gilan	18	13	30	24	10	11	14	17	18	10	14	14	20	14	15	21	12
13	Gilan	14	12	28	23	10	11	12	13	15	9	11	15	18	16	17	20	11
14	Gilan	14	13	29	23	10	14	13	12	13	10	10	14	18	15	15	25	12
15	Gilan	14	13	31	23	10	11	12	13	20	10	11	14	21	15	17	20	11
16	Gilan	14	14	30	24	10	10	12	13	15	9	11	15	20	15	15	22	12
17	Gilan	14	13	29	23	10	13	12	11	14	12	12	15	19	17	15	24	12
18	Gilan	13	13	29	22	10	15	13	15	16	11	11	14	20	15	17	23	10
19	Gilan	14	13	29	24	10	13	12	12	14	12	12	15	19	16	15	23	12
20	Gilan	15	13	31	23	10	11	12	13	20	10	12	14	20	15	19	20	11
21	Gilan	14	13	30	22	11	11	12	13	17	10	11	14	20	14	18	21	11
22	Gilan	16	14	32	25	10	11	13	11	14	11	11	14	20	18	15	23	13
23	Gilan	14	12	28	23	10	11	12	14	16	9	12	15	22	15	17	23	11
24	Gilan	14	13	29	23	10	11	13	13	17	9	11	15	21	16	18	23	11
25	Gilan	15	13	29	22	10	11	14	12	20	14	11	14	20	14	17	21	10
26	Gilan	14	13	30	24	11	13	12	12	14	12	12	15	19	15	16	23	12
27	Gilan	14	13	29	24	11	13	12	11	14	12	12	15	19	15	15	23	11
28	Gilan	15	13	30	22	10	11	13	14	14	10	11	16	18	14	18	21	12
29	Gilan	15	13	29	22	10	11	13	12	18	10	12	14	20	16	18	19	10
30	Gilan	14	13	28	24	10	14	12	11	14	12	14	15	19	15	18	23	12
31	Gilan	13	14	30	22	10	15	13	12	17	11	11	15	19	15	18	23	10
32	Gilan	12	13	30	25	10	10	15	14	16	10	11	16	21	17	16	21	10
33	Gilan	16	12	29	23	10	10	14	12	14	10	11	16	20	16	17	21	12
34	Gilan	15	13	29	22	10	11	13	12	16	9	11	14	21	15	15	21	12
35	Gilan	15	13	29	22	10	11	15	12	14	10	12	16	21	17	15	21	11
36	Gilan	15	13	29	23	10	11	12	13	16	9	11	15	21	17	15	22	12
37	Gilan	13	13	31	24	9	11	14	15	18	10	12	14	20	16	17	22	12
38	Gilan	14	12	28	23	10	11	12	14	16	9	11	16	20	15	15	21	11
39	Gilan	14	12	28	24	10	11	12	14	16	9	12	15	20	16	15	21	11
40	Gilan	14	13	30	23	10	11	12	13	18	10	11	14	20	14	19	21	11
41	Gilan	15	12	29	22	10	11	14	13	14	10	11	16	20	15	18	21	12
42	Gilan	15	12	28	22	10	11	14	12	14	10	11	16	20	15	16	21	12
43	Gilan	13	14	31	24	10	13	12	11	14	12	15	15	19	16	16	23	12
44	Gilan	15	12	28	22	10	11	14	14	14	10	11	16	15	15	15	23	11
45	Gilan	14	13	29	24	11	13	12	11	14	12	11	14	20	14	17	23	12
46	Gilan	14	12	28	23	11	11	12	13	16	9	12	15	22	15	15	21	12
47	Gilan	14	13	29	24	10	11	13	13	13	9	12	15	21	15	17	23	12
48	Gilan	13	13	31	23	10	11	12	12	16	9	10	15	19	15	17	24	12
49	Gilan	14	12	30	24	10	15	13	14	14	11	11	14	19	15	16	22	12
50	Gilan	16	11	28	22	10	12	13	12	14	10	12	15	23	14	17	21	10
51	Gilan	15	13	29	23	10	11	13	12	18	10	11	14	20	16	17	20	10
52	Gilan	14	13	30	23	10	11	12	13	17	9	12	15	20	17	17	21	10
53	Gilan	15	14	30	24	11	13	13	12	13	11	13	14	18	14	14	24	12
54	Gilan	14	13	28	23	10	11	12	13	16	9	11	15	21	15	17	22	11
55	Gilan	16	13	30	21	10	11	15	13	14	10	11	17	21	15	16	21	11
56	Gilan	15	12	29	23	10	11	13	13	16	9	12	15	20	15	16	24	12
57	Gilan	15	13	29	23	10	13	12	11	14	12	12	15	19	16	16	24	12
58	Gilan	14	14	31	23	10	11	12	12	19	10	12	14	21	15	13	20	11
59	Gilan	15	14	32	23	10	11	13	13	14	10	12	16	19	14	17	23	12
60	Gilan	15	13	30	22	10	11	12	15	15	9	12	14	21	15	16	22	11
61	Gilan	15	14	31	25	11	11	13	11	15	12	10	14	20	16	15	23	12
62	Gilan	15	13	29	24	11	13	13	12	13	11	12	14	18	14	17	25	11

Table 3. (Continued).

63	Gilan	15	14	31	24	10	13	12	14	22	10	12	14	21	13	18	21	12
64	Gilan	14	13	29	23	10	11	12	12	17	9	12	15	21	15	15	23	13
65	Gilan	13	13	31	24	10	11	12	12	16	9	10	15	19	15	19	24	13
66	Gilan	15	13	30	25	10	10	13	11	14	11	10	14	20	15	19	23	12
67	Gilan	14	13	29	25	10	13	12	11	14	12	14	14	19	15	15	23	12
68	Gilan	14	13	29	23	10	11	12	13	17	10	12	14	20	14	13	20	11
69	Gilan	14	12	28	23	10	11	13	13	17	9	13	15	21	15	15	22	12
70	Gilan	14	13	30	23	10	11	13	12	19	10	11	14	19	15	20	20	12
71	Gilan	14	12	28	23	11	11	12	12	17	9	12	15	22	16	15	21	12
72	Gilan	16	13	30	22	10	11	14	12	20	10	12	14	20	17	16	21	11
73	Gilan	14	13	28	24	10	14	12	11	14	12	13	15	20	16	17	23	13
74	Gilan	16	11	29	23	10	12	14	13	15	10	11	15	20	15	18	21	11
75	Gilan	13	14	31	24	10	12	12	12	17	9	10	15	19	15	17	24	12
76	Gilan	14	12	29	24	11	11	14	11	13	9	11	14	21	15	15	21	11
77	Gilan	15	12	29	22	10	11	14	12	15	10	12	16	21	15	16	21	12
78	Gilan	15	13	31	25	10	11	13	11	14	10	10	14	19	16	17	23	13
79	Gilan	15	13	29	23	10	11	13	12	18	11	11	14	20	15	13	21	10
80	Gilan	13	13	32	23	11	14	13	16	18	11	12	13	19	15	16	22	11
81	Gilan	14	13	30	24	11	13	12	11	15	12	12	14	19	15	17	23	11
82	Gilan	14	14	30	24	10	10	13	14	19	11	11	16	19	15	15	25	12
83	Gilan	14	13	29	23	10	11	12	13	17	9	12	14	21	15	15	22	12
84	Gilan	14	13	30	23	9	12	12	13	18	10	11	14	20	14	15	21	11
85	Gilan	14	12	30	24	11	11	14	14	14	9	12	14	20	15	16	24	12
86	Gilan	15	13	31	23	9	10	14	15	21	9	11	15	19	15	15	21	11
87	Gilan	15	13	29	24	10	13	13	12	13	11	12	14	17	15	14	26	12
88	Gilan	14	13	31	23	10	11	12	14	16	9	13	14	20	15	16	23	11
89	Gilan	13	13	30	22	11	16	11	12	16	10	11	16	21	16	16	21	11
90	Gilan	14	12	30	24	11	11	14	14	14	9	12	14	20	15	16	24	12

Table 4. Effective allele, Shannon's index, and genetic diversity in the studied populations.

Population	STRP marker	Population size	Number of alleles	Effective allele	Shannon's index	Genetic diversity
Mazandaran	DYS19	119	5	2.677	1.200	0.643
	DYS389I	119	5	2.030	0.917	0.549
	DYS389II	119	8	4.248	1.632	0.828
	DYS390	119	6	2.859	1.267	0.665
	DYS391	119	4	1.959	0.859	0.480
	DYS392	119	6	2.082	1.056	0.514
	DYS393	119	6	2.291	1.078	0.569
	DYS385a	119	8	5.186	1.757	0.806
	DYS385b	119	11	5.759	1.949	0.863
	DYS438	119	6	3.641	1.412	0.726
	DYS439	119	5	2.611	1.123	0.634
	DYS437	119	3	2.450	0.963	0.612
	DYS448	119	8	3.771	1.488	0.738
	DYS456	119	6	2.472	1.178	0.578
	DYS458	119	6	4.478	1.626	0.774
	DYS635	119	7	4.221	1.619	0.772



**Table 4.** (Continued).

	GATA H4	119	5	2.750	1.178	0.654
Gilan	DYS19	90	6	2.897	1.251	0.662
	DYS389I	90	4	2.207	0.988	0.553
	DYS389II	90	5	3.810	1.430	0.746
	DYS390	90	5	3.389	1.314	0.713
	DYS391	90	3	1.599	0.657	0.379
	DYS392	90	7	2.479	1.279	0.603
	DYS393	90	5	2.822	1.175	0.653
	DYS385a	90	7	4.245	1.563	0.773
	DYS385b	90	10	5.393	1.923	0.824
	DYS438	90	6	3.675	1.408	0.736
	DYS439	90	7	3.080	1.344	0.683
	DYS437	90	5	2.751	1.119	0.644
	DYS448	90	8	3.778	1.527	0.744
	DYS456	90	6	2.852	1.267	0.657
	DYS458	90	8	4.693	1.710	0.796
	DYS635	90	8	4.167	1.632	0.769
	GATA H4	90	4	2.971	1.204	0.671

**Table 5.** Allele frequencies per marker in Mazandaran and Gilan provinces.

Locus	Allele	Mazandaran	Gilan	Locus	Allele	Mazandaran	Gilan	Locus	Allele	Mazandaran	Gilan
DYS19	N	119	90	DYS385b	N	119	90	DYS458	N	119	90
	12	0.000	0.011		11	0.0168	0.000		13	0.000	0.033
	13	0.0756	0.100		12	0.008	0.000		14	0.050	0.033
	14	0.521	0.467		13	0.025	0.067		15	0.21	0.278
	15	0.268	0.333		14	0.226	0.333		16	0.344	0.222
	16	0.1178	0.078		15	0.168	0.100		17	0.193	0.267
	17	0.016	0.000		16	0.218	0.178		18	0.134	0.100
	18	0.000	0.011		17	0.184	0.122		19	0.067	0.056
DYS389I	N	119	90		18	0.075	0.100		20	0.000	0.011
	11	0.008	0.022		19	0.042	0.033	DYS635	N	119	90
	12	0.134	0.211		20	0.025	0.044		12	0.008	0.000
	13	0.621	0.622		21	0.008	0.011		19	0.000	0.011
	14	0.226	0.144		22	0.000	0.011		20	0.084	0.100
	15	0.008	0.000	DYS438	N	119	90		21	0.336	0.344
DYS389II	N	119	90		7	0.000	0.000		22	0.134	0.100
	20	0.008	0.000		8	0.008	0.000		23	0.294	0.300
	22	0.008	0.000		9	0.352	0.322		24	0.075	0.100
	27	0.025	0.000		10	0.344	0.344		25	0.067	0.033
	28	0.109	0.178		11	0.142	0.133		26	0.000	0.011
	29	0.369	0.367		12	0.134	0.178	GATA H4	N	119	90



Table 5. (Continued).

	30	0.134	0.267		13	0.016	0.000		9	0.000	0.000
	31	0.075	0.156		14	0.000	0.011		10	0.042	0.111
	32	0.083	0.033		20	0.000	0.011		11	0.453	0.356
DYS390	N	119	90	DYS439	N	119	90		12	0.361	0.433
	21	0.000	0.011		9	0.000	0.011		13	0.117	0.100
	22	0.016	0.222		10	0.042	0.078		14	0.0168	0.000
	23	0.193	0.378		11	0.445	0.389		15	0.000	0.000
	24	0.495	0.311		12	0.411	0.400				
	25	0.226	0.078		13	0.084	0.078				
	26	0.016	0.000		14	0.016	0.033				
	31	0.008	0.000		15	0.000	0.011				
DYS391	N	119	90		24	0.000	0.000				
	8	0.016	0.000	DYS437	N	119	90				
	9	0.050	0.044		10	0.000	0.000				
	10	0.672	0.767		13	0.000	0.011				
	11	0.265	0.189		14	0.445	0.433				
DYS392	N	119	90		15	0.421	0.389				
	10	0.006	0.067		16	0.134	0.156				
	11	0.672	0.600		17	0.000	0.011				
	12	0.050	0.056	DYS448	N	119	90				
	13	0.176	0.178		11	0.008	0.000				
	14	0.016	0.056		15	0.000	0.011				
	15	0.016	0.033		17	0.008	0.011				
	16	0.000	0.011		18	0.042	0.067				
DYS393	N	119	90		19	0.310	0.267				
	11	0.016	0.011		20	0.285	0.378				
	12	0.179	0.467		21	0.294	0.211				
	13	0.235	0.333		22	0.042	0.033				
	14	0.134	0.156		23	0.008	0.022				
	15	0.0252	0.033	DYS456	N	119	90				
	16	0.008	0.000		10	0.008	0.000				
DYS385a	N	119	90		12	0.000	0.000				
	9	0.000	0.000		13	0.025	0.011				
	10	0.008	0.000		14	0.159	0.178				
	11	0.193	0.167		15	0.605	0.522				
	12	0.243	0.311		16	0.168	0.200				
	13	0.243	0.278		17	0.03	0.078				
	14	0.168	0.178		18	0.000	0.011				
	15	0.092	0.044		20	0.000	0.000				
	16	0.033	0.011								
	17	0.016	0.011								

N: number of repeats.

Amplification of all 17 Y-STRP markers signifies the efficiency of these markers for forensic and molecular investigation of the Y-chromosome in the north of Iran. Haplotype analysis via 17 Y-STRP markers of 259 males from eastern provinces of Iran showed HD = 0.9999 (12). This value was declared to be 0.997 in Tehran and Isfahan provinces by analyzing 9 Y-STRP markers (11). By analyzing 8 Y-STRP markers in 103 unrelated males from Isfahan Province, Vatandoost et al. announced that HD = 0.986 (14). Here we obtained HD = 0.9993 in Mazandaran and HD = 0.9663 in Gilan. Both provinces showed a lower HD by using these STRP markers when compared with eastern provinces of Iran, while the value is greater for Gilan in comparison with Tehran and Isfahan. This value was reported as 0.9850 in Afghanistan (16) and 0.9992 in Turkish Cypriots (17). The lower HD obtained for Mazandaran demonstrates that there has been less migration to this province; on the other hand, the male population in this province is isolated. The calculated DC in Gilan was higher than the previously reported DC in

Tehran (DC = 0.9), Isfahan (DC = 0.938 and DC = 0.95) (11,14), and the eastern population of Iran (DC = 0.9884) (12), which illustrates the capability of these markers for male identity in this province. However, the DC value in Mazandaran was higher than the one in other provinces except for eastern provinces including Razavi Khorasan, South Khorasan, and Sistan and Baluchestan. Comparing Mazandaran and Gilan together and with neighboring countries, Afghanistan and Azerbaijan, could not establish evidence of a common ancestral lineage. It demonstrates the unique diversity among Iranian population, which is due to geographical verities and ethnic differences.

Our results elucidate that using Y-STRP markers has the capability of discriminating male identity among northern Iranian male subjects. Studying additional Y-STRP markers with greater sample size would help to remove matched haplotypes and determine valuable polymorphic markers in this region. This would provide a better circumstance in order to utilize ethnic-specific markers for forensic applications.

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