

## Y-STR Haplotype Diversity and Population Data for Al-Anbar in Iraq

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### ABSTRACT

In this study, a sample of 112 Al-Anbar Iraq population was analysed using 17 Y- chromosome short tandem repeat (STR) polymorphisms. A total of 112 different haplotypes were identified, 67 unique haplotype and 45 shred. The high haplotype diversity (0.988) supports the usefulness of Y-STR markers in Al-Anbar of Iraq population diversity investigation. DC was 0.598214 Our results suggest a close genetic relationship between Al-Anbar and other populations of the Arabian Peninsula, and an even more pronounced similarity of Kuwaiti populations and Yemenis and Saudi Arabians.

**Keywords-** Haplotype, Y chromosome, population structure.

Investigation (FBI) announced a core of 13 autosomal STR loci required for the National DNA Index System (NDIS), a subset of the U.S. National Combined DNA Index System (CODIS), These markers were chosen to be highly informative, easily amplified using PCR, and on different chromosomes or opposite ends of the same chromosome so that they are almost genetically unlinked (6).

A DNA analysis of markers located on the Y chromosome is valuable in certain cases, particularly in sexual assault cases, in which evidence often contains a mixture of DNA from both a female victim and a male perpetrator. The forensic community has agreed on a core set of eight Y-STR (Y-chromosome short tandem repeat) markers, named the minimal haplotype DNA Y-chromosome(unique) segment(DYS) (DYS19, DYS389 I, DYS389 II, DYS390, DYS391, DYS392, DYS393 and DYS385ab) to use in routine Y-STR analysis(7).

The routine Y-STR analysis is based on multiplex amplification of the minimal haplotype loci as well as the loci recommended by **Scientific Working Group on DNA Analysis Methods (SWGDM)** (DYS439 and DYS439). The amplified fragments are then size separated using CE. (8, 9).

Highly variable regions within the DNA termed Short Tandem Repeats (STRs) are widely used for characterizing population structure and estimating human genetic diversity (10,11).

Such DNA-based data also provide leads in disease susceptibility studies, paternity and individual identification. Population genetic analyses utilizing such variable markers have identified bidirectional human migration through the Middle East, linking movement through Africa, Asia and Europe (12,13 ).

### I. INTRODUCTION

Humans' origin and migrations is an active topic in the field of anthropology and molecular genetics (1,2). Separate genetic profiles can be constructed using standard marker sets for the autosomal (chromosomes one through 22), Y-chromosome, and mitochondrial DNA. The types of genetic marker currently used in forensic genetic practice are length polymorphisms called short tandem repeats (STRs). STRs used forensically typically have a simple or more complex tetranucleotide repeat structure. The markers are highly polymorphic, with between 15 and more than 100 alleles observed at a locus, giving these markers, taken together, high power to discriminate between individuals (3).

In 1997, an effort was made to standardize the nomenclature for STR alleles, based on the number of repeated motifs or overall length polymorphism size(4,5). That same year, the Federal Bureau of

The Y chromosome is less variable than the other chromosomes. Many markers are thus needed to obtain a high degree of discrimination between unrelated males (14,15).

Research in DNA technologies has helped law enforcement agencies such as the police in the investigation of crimes such as murder, attempted murder, physical assault, and sexual assault. Sexual assault such as rape is one of the most violent crimes and is a serious problem which is faced by many countries (16).

The Y-chromosome haplotype is commonly constructed using Short Tandem Repeat (STR) markers. As the Y-chromosome is subject to rapid genetic drift, haplotypes can be used to study the geographical distribution of ethnic groups (17). The Y-chromosome contains the largest non-recombining section within the human genome, providing informative haplotypes for genetic analyses of populations (18). The main source of information about Y-STR is the website YHRD.org. In practice, the frequency of the Y-STR profile in question can be estimated by referencing databases with large numbers of Y-STR profiles and counting the number of matching profiles within the population of interest. This provides an estimate of the profile probability for the Y-haplotype. One of these databases is the Y Chromosome Haplotype Reference Database (YHRD) (19). Following the guidelines of the International Society of Forensic Genetics (ISFG) for the publication of genetic population data, submissions to YHRD are required to consist of at least 17 Y-STR markers (including the eight-marker minimal Y-haplotype: DYS19, DYS389I, DYS389II, DYS390, DYS391, DYS392, DYS393, DYS385)5 for 200 or more individuals from a population.44 As of 2015, YHRD contained more than 84,000 Y-profiles of 17 markers or greater in 572 populations.

It contains a description of the Y-STR loci used in forensic analysis, mutation rates, and the biggest global database available (more than 194,000 sampled individuals to this date). Moreover, one of the *f*-estimating methods is implemented on the website, and its estimates are freely available. The only drawback is that, since many researchers publish anonymously on the website and do not want their data to be globally known, the haplotypes in the database are not accessible. For a haplotype H, an expert can only access the number of occurrences of H in the database, by subpopulation, and the *f*-estimate mentioned earlier. (19).

## II. MATERIALS AND METHODS

### Collection samples

In this study, the buccal swab was collected from unrelated individual living in central of Iraq. 178 samples divided into Baghdad 178 sample from (Baghdad University and Al- Nahrain University), All samples after collected left to dry for 30 minutes and

then after to dry put inside a letter envelope until they are extraction. All samples were DNA extraction in laboratory Al-Nahrain University in forensic DNA center research and training.

### Genomic DNA extraction methods

DNA was extracted from buccal samples using organic extraction method (manual method), extraction done according to the manufacturer protocol, this method depended in forensic DNA laboratory. **Organic extraction method(20).**

### Estimate quantification of DNA

The quantity of DNA was determined according to methods described by quantifier Y Human male quantification kit (21,22).

### Amplification Y chromosome STR by using AmpFSTR<sup>R</sup> Y Filer kit.

Commercial kit AmpFSTR<sup>R</sup> Y Filer PCR amplification kit (applied bio systems)

### Allele frequency

The allele frequency of the multicopy locus DYS385a/b was analyzed as combination of both alleles (16,23).

Allele frequencies for Y- Chromosomal STR were calculated by direct counting therefore:

$$\text{Allele frequencies} = \frac{\text{Total no. of alleles}}{\text{Total no. of samples}}$$

### Haplotype Frequency

Haplotype frequency it means of each haplotype of the sample found with any sample size. Haplotype Frequency was calculated by using the Excel program.

### Gene Diversity (GD)

Gene diversity (GD) was calculated for each Y-STR according to the formula supplied by (24,25):

$$HD = \frac{N}{N-1} (1 - \sum x^2)$$

Where *n* is the sample size and *x* the relative allele frequency. Gene diversity among populations occurs if there are differences in allele frequencies between those populations

### Haplotype Diversity (HD)

Haplotype Diversity (HD) was calculated using the same equation as calculating gene diversity using haplotype frequencies instead of allele frequencies (25).

### Discrimination Capacity (DC)

Discrimination Capacity (DC) was calculated using the following formula (25)

### Y-STR Haplotype Reference Database (YHRD)

The largest and most widely used forensic and general population genetics Y-STR database, known as the Y-STR Haplotype Reference Database (YHRD), was created by Willuweit and Roewer (2007).

### III. RESULTS AND DISCUSSIONS

Samples collected for the purpose to we will work were collected (Y-STR) of the provinces of Iraq of Arab Individuals lives in included the central governorates and southern Iraq (Baghdad, Al-Anbar, Diyala, south of Iraq).

The number of samples collected from the Baghdad area 178 (University of Baghdad and Al-Nahrain),

#### Allele, Genotype frequencies and Gene Diversity for Al-Anbar of Iraq males (n=112)

The GD value for DYS385 a/b among the males from Al-Anbar community was 0.898 (Table 4.3). ). This was the least diversity of genes observed anywhere between all four subgroups that were investigated.

The highest GD value for a single-copy-locus was 0.794 (DYS458). The lowest GD value was 0.230 (DYS392).

Allele frequency of 112 from Al-Anbar of Iraq Arab males' individuals that scored for the 17 Y-STR chromosome STRs are shown in (Table 4.3) and (Appendix) DYS458, DYS456, DYS3891, and DYS635 have 7 and 6 number of alleles respectively (Table 4.3) with genetic diversity of 0.794, 0.652, 0.526 and 0.714 respectively.

Loci which exhibited lower polymorphisms were DYS437 and R-Y-GATA-H4 which have a total number of 3 alleles with genetic diversity 0.346 and 0.543 respectively. DYS3891, DYS38911, DYS19, DYS393, DYS439, DYS438 and DYS448 have 4 total number of alleles with 0.526, 0.623, 0.471 , 0.518 , 0.612 , 0.595 and 0.445 genetic diversity.

Table (1): Allele, Genotype frequencies and Gene Diversity for Al-Anbar of Iraq males (n=112)

Allele	DYS 456	DYS 3891	DYS 3892	DYS 3893	DYS 3894	DYS3 895	DYS3 896	DYS 3897	DYS 3898	DYS 3899	DYS 3900	DYS3 901	DYS3 902	DYS 3903	DYS 3904	Genoty pe	DYS3 85ab
6																10,19	0.009
7																11,12	0.009
8																11,14	0.161
9								0.027						0.161		11,15	0.045
10		0.009						0.375	0.179					0.580		11,18	0.018
11		0.098					0.045	0.500	0.571		0.875	0.625		0.214		12,12	0.009
12		0.634					0.652	0.089	0.170		0.009	0.205		0.045		12,14	0.009
13	0.027	0.259				0.036	0.232	0.009	0.080		0.036	0.170				12,15	0.018
14	0.429				0.045	0.696	0.071				0.071		0.786			12,17	0.027
15	0.393				0.089	0.205					0.009		0.196			12,18	0.036
16	0.116				0.259	0.063							0.018			13,13	0.027
17	0.027				0.089											13,14	0.009
18	0.009				0.313										0.009	13,15	0.018
19					0.170										0.143	13,16	0.036
20					0.036					0.089					0.723	13,17	0.054
21			0.018							0.473					0.125	13,18	0.134
22			0.054							0.134						13,19	0.232
23			0.625							0.188						13,20	0.027
24			0.152							0.063						13,21	0.009
25			0.12							0.05						14,15	0.018

			5							4							
26			0.027													14,16	0.009
27																14,17	0.009
28				0.054												14,18	0.018
29				0.313												14,19	0.009
30				0.518												15,16	0.027
31				0.116												16,17	0.009
32																17,19	0.009
33																18,19	0.009
34																	
Sum freq	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000		
N	112	112	112	112	112	112	112	112	112	112	112	112	112	112	112		112
NA	6	4	6	4	7	4	4	5	4	6	5	3	3	4	4	28	
GD	0.653	0.526	0.572	0.623	0.794	0.47	0.51882	0.606	0.612	0.715	0.23	0.5433	0.3468	0.595	0.445		0.898

\*N=number of samples; NA= number of Alleles of each Loci; GD = gene Diversity

Table (2): Gene diversity central of Iraq comparison with other countries

	South of Iraq <sup>a</sup>	Baghdad <sup>b</sup>	Al-Anbar <sup>c</sup>	Diyala <sup>d</sup>	Iraq <sup>e</sup>	Iraq – Kurd <sup>f</sup>	Iraq - Yazidis <sup>g</sup>	Sudia - arbia <sup>h</sup>	Kuwait <sup>i</sup>	Emirate <sup>s</sup>	Turkey <sup>k</sup>
Allele	GD	GD	GD	G.D	GD	GD	GD	GD	GD	GD	GD
DYS456	0.692	0.677	0.653	0.635	—	0.637	0.525	0.664	0.698	0.678	0.520
DYS3891	0.469	0.535	0.526	0.482	0.660	0.596	0.616	0.566	0.516	0.525	0.560
DYS390	0.598	0.663	0.572	0.647	0.657	0.708	0.732	0.663	0.533	0.707	0.700
DYS38911	0.675	0.715	0.623	0.603	0.714	0.728	0.726	0.726	0.699	0.684	0.710
DYS458	0.856	0.801	0.794	0.830	—	0.827	0.816	0.829	0.783	0.837	0.840
DYS19	0.572	0.532	0.472	0.603	0.665	0.634	0.721	0.600	0.593	0.643	0.640
DYS393	0.575	0.576	0.519	0.639	0.514	0.585	0.634	0.651	0.638	0.641	0.580
DYS391	0.534	0.548	0.606	0.588	0.580	0.493	0.464	0.493	0.574	0.455	0.550
DYS439	0.575	0.640	0.612	0.560	0.299	0.675	0.738	0.604	0.637	0.678	0.650
DYS635	0.668	0.688	0.715	0.693	—	0.764	0.770	0.678	0.717	0.707	0.700
DYS392	0.373	0.373	0.230	0.253	0.269	0.514	0.325	0.354	0.477	0.402	0.450
R-Y-GATA-H4	0.542	0.623	0.543	0.578	—	0.658	0.667	0.481	0.559	0.590	0.720
DYS437	0.497	0.449	0.347	0.567	—	0.547	0.526	0.353	0.409	0.437	0.440
DYS438	0.553	0.584	0.595	0.593	0.526	0.698	0.660	0.508	0.611	0.643	0.700
DYS448	0.517	0.586	0.445	0.631	—	0.659	0.525	0.627	0.577	0.625	0.670
DYS385a/b	0.939	0.949	0.898	0.933	0.839/0.65	0.958	0.952	0.921	0.957	0.949	0.950

**Y-STR-haplotype and haplotype frequency**

In this study 112 haplotypes each with 17 alleles were analyzed by used excel. analysis measures the Haplotype and Haplotype frequency The observed number of Haplotype and Haplotype frequency of any individual have been tabulated in Table (4). We identified 112 different haplotype in our study sample. 67 (59.82%) were unique Haplotype , 45 were (40.18%) were replicated haplotype among more individuals.

The highest haplotype were in sample 3 at frequency 0.0714 ( haplotype replicated 8 times ), while the haplotype number were replicated 6 times(frequency of haplotype 0.0625).

**Haplotype diversity and Discrimination Capacity**

DC of the 17 Y-STR Yfiler 0.598214 while HD was (0.988) in 17 Y-STR table (3)

Table (3): Discrimination Capacity and Haplotype diversity

	Y-STR haplotype
sample size	112
Number of haplotype	81
Unique haplotype	67
Repeating haplotype	45
HD	0.988
Discrimination capacity	0.598

Table (4): Haplotype frequency in Al-Anbar of Iraq

ID	DYS4 56	DYS3 89I	DYS 390	DYS3 89II	DYS4 58	DYS1 9	DYS3 85ab	DYS3 93	DYS3 91	DYS4 39	DYS6 35	DYS3 92	GAT AH4	DYS4 37	DYS4 38	DYS4 48
H1	14	13	23	29	19	14	13,19	12	12	11	21	11	11	14	10	20
H2	14	13	23	29	19	14	13,18	12	10	11	21	11	11	14	10	20
H3	16	13	25	30	16	15	11,14	13	10	10	23	11	13	14	11	20
H4	15	13	25	30	16	15	11,15	13	10	10	23	11	13	14	11	20
H5	14	14	23	31	19	14	13,19	12	11	11	21	11	11	14	10	20
H6	14	13	23	29	18	14	13,19	12	12	12	22	11	11	14	10	20
H7	14	13	23	30	18	14	13,19	12	11	11	21	11	11	14	10	20
H8	15	13	23	29	18	14	13,18	12	11	11	22	11	11	14	10	20
H9	14	13	23	30	19	14	13,18	12	11	11	21	11	11	14	10	21
H10	17	13	24	29	16	14	12,15	12	11	13	23	13	13	14	12	20
H11	14	13	23	29	18	14	13,19	12	11	11	22	11	11	14	10	20
H12	17	13	24	29	17	16	12,15	13	10	13	23	11	11	14	11	20
H13	14	13	23	30	19	14	13,19	12	12	12	21	11	11	14	10	20
H14	14	13	23	30	19	14	13,18	12	11	11	21	11	11	14	10	21
H15	15	14	23	30	16	14	14,17	11	11	13	21	11	11	14	9	20
H16	14	14	23	30	18	14	14,19	12	11	11	21	11	11	14	10	19
H17	18	14	23	31	17	15	14,15	13	10	11	22	11	11	15	9	20
H18	14	14	23	30	18	14	13,19	12	11	11	20	11	11	14	10	20
H19	16	14	24	30	15	14	11,15	12	11	12	25	13	12	15	11	20
H20	14	13	23	29	18	14	13,19	12	10	11	22	11	11	14	10	20
H21	15	13	23	29	16	14	11,12	13	10	10	24	14	13	14	10	18
H22	16	13	26	30	15	16	11,14	13	11	10	23	11	13	15	11	20
H23	14	13	23	29	20	14	12,18	12	11	11	21	11	11	14	10	20
H24	16	14	23	31	14	14	15,16	12	11	13	21	11	12	14	9	20
H25	16	13	23	29	14	16	13,16	12	9	12	21	11	12	14	9	20
H26	15	13	25	30	18	16	13,15	12	10	11	21	11	11	14	9	19
H27	16	13	24	29	16	14	11,14	12	11	12	25	14	12	15	12	19
H28	15	14	22	31	16	14	12,14	13	10	11	22	11	11	15	9	21
H29	15	13	24	29	15	16	13,17	13	11	11	22	13	11	14	9	20
H30	15	13	24	30	16	15	11,14	13	10	10	23	11	13	14	11	20
H31	14	13	23	29	19	13	13,17	12	11	11	22	11	11	14	10	20
H32	15	13	25	30	16	15	11,14	13	10	10	23	11	13	14	11	20

H33	14	13	23	29	18	14	13,18	12	11	11	21	11	11	14	10	20
H34	17	12	21	28	17	15	14,15	14	10	12	21	11	11	16	10	21
H35	15	14	23	31	19	14	13,19	12	11	11	21	11	11	14	10	20
H36	15	14	23	31	17	14	17,19	13	10	12	20	11	13	14	10	21
H37	15	14	23	31	16	14	14,18	12	11	13	22	11	11	14	9	20
H38	13	13	25	30	18	15	13,16	12	11	11	21	12	12	15	9	20
H39	14	13	23	29	18	13	13,17	12	11	11	21	11	11	14	10	20
H40	16	13	23	30	15	15	13,16	12	9	12	22	11	12	14	9	21
H41	14	13	25	31	15	15	11,15	14	11	10	23	11	12	14	11	20
H42	15	14	23	30	15	14	13,13	14	10	11	24	11	11	15	9	20
H43	14	13	23	30	19	14	13,18	13	11	11	20	11	11	14	10	20
H44	15	14	24	31	14	14	18,19	13	10	11	24	11	12	14	10	20
H45	15	13	24	30	16	14	13,14	12	10	11	20	11	12	14	10	19
H46	15	14	23	30	16	14	12,17	12	10	11	21	11	11	15	9	21
H47	15	14	22	31	17	14	10,19	12	10	11	21	11	12	14	9	20
H48	15	12	23	29	18	15	13,15	14	11	11	21	11	12	16	10	21
H49	15	13	24	31	16	13	16,17	13	10	12	21	11	11	14	10	20
H50	15	13	23	31	18	15	12,17	12	10	11	20	11	11	14	10	20
H51	15	12	22	28	17	14	13,17	11	9	12	24	14	11	15	10	19
H52	14	13	23	28	19	14	13,18	12	10	11	21	11	11	14	10	20
H53	14	13	23	30	19	14	13,18	12	10	11	21	11	11	14	10	20
H54	15	12	26	30	17	15	11,15	14	10	10	23	11	12	14	11	20
H55	15	12	23	29	17	14	11,18	12	11	12	20	11	11	15	10	19
H56	16	12	22	28	14	14	14,16	11	10	13	25	14	12	15	10	19
H57	14	14	23	30	18	14	13,19	12	12	11	21	11	11	14	10	20
H58	14	13	23	29	19	14	13,20	12	12	11	21	11	11	14	10	20
H59	13	10	22	29	18	14	13,18	12	11	12	21	11	11	14	10	20
H60	14	14	23	30	19	14	13,19	12	12	10	21	11	11	14	10	20
H61	14	14	23	30	19	14	13,19	12	12	11	21	11	11	14	10	20
H62	14	13	23	30	19	14	13,19	12	11	11	21	11	11	14	10	21
H63	15	12	22	28	15	14	13,17	11	10	13	23	15	12	15	11	19
H64	14	14	23	30	18	14	13,18	12	12	11	21	11	11	14	10	20
H65	14	14	23	30	18	14	13,20	12	11	11	21	11	11	14	10	19
H66	15	12	24	29	16	15	11,14	13	12	10	23	11	13	14	11	20
H67	14	13	23	29	19	14	13,19	12	11	11	22	11	11	14	10	19
H68	15	13	21	30	18	16	15,16	14	10	12	21	11	12	14	11	21
H69	15	12	24	28	18	14	12,18	12	10	12	20	14	13	15	11	20
H70	14	13	23	29	18	14	13,20	12	12	11	21	11	11	14	10	20
H71	16	13	26	30	15	16	11,14	13	11	10	24	11	13	15	11	20
H72	14	14	23	30	20	14	13,19	12	13	11	22	11	11	14	10	20
H73	15	13	23	30	18	14	13,18	12	10	11	20	11	12	14	10	20
H74	13	13	24	29	18	14	13,19	12	11	11	22	11	12	14	10	20
H75	15	14	23	30	16	14	14,18	11	11	13	22	11	11	14	9	20
H76	15	13	24	30	18	14	13,21	12	10	11	20	11	11	14	10	20
H77	14	13	23	29	17	13	13,17	12	11	11	21	11	11	14	10	20

H78	14	13	23	30	19	14	13,16	12	11	11	21	11	11	14	10	21
H79	14	13	23	29	18	14	13,18	12	11	11	22	11	11	14	10	20
H80	15	12	23	29	16	15	12,12	13	10	12	23	13	12	15	11	19
H81	14	14	23	30	18	14	13,19	12	11	11	21	11	11	14	10	20

#### IV. CONCLUSIONS

1- This present study has shown the Al-Anabr of Iraq Arab population to be diverse with 67 unique haplotypes and 45 Repeating haplotype observed in 81 haplotype.

2- The DYS385b had the highest diversity (GD =0.898 while loci DYS392 the lowest(D = 0.23).

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