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## FORENSIC Y-STR STUDY IN Y-CHROMOSOME ABNORMALITIES

**Abstract:** Y-chromosome infertility is usually caused by deletions of genetic material in Azoospermia Factor (AZF) regions, localized in three nonoverlapping Yq regions – AZFa, AZFb and AZFc. However, some of the Y-STRs routinely studied in Forensic Genetics with multiplex kits are localized in these three regions giving rise to microdeletions observed in some *loci*. Rare alleles were also encountered in these samples with Y-chromosome structural changes. A complete Y-STR profile in a sample from a female individual was detected which may raise ethical implications. The aim of this work is to assess the validity in the use of Y-STR multiplex systems in structurally abnormal Y-chromosomes, which may occur in forensic casework analysis.

**Keywords:** AZF regions; Y-STRs; microdeletions.

### Introduction

Y-chromosome infertility is usually caused by deletions of genetic material in AZF region, localized in three no overlapping Yq regions – AZFa, AZFb and AZFc (1-3). Microdeletions are rare in AZFa region, concerning 1-2% of total deletions in AZF region. The most frequent microdeletions are observed in AZFc region where we encountered 80% of total AZF region deletions (4,5). However, some of the Y-STRs routinely studied in Forensic Genetics are placed in these three regions (6). The aim of this work is to assess the validity in the use of Y-STRs in structurally abnormal Y-chromosomes, which may occur in forensic casework.

### Material and Methods

Y-STRs were studied with AmpFISTR® Yfiler® in 46 samples previously screened for AZF region deletions and with informed consent. DNA samples from these individuals were provided as blind samples, not being known how many individuals had AZF microdeletions. Samples of 50 individuals also with informed consent and with clinical indication of male infertility but with no Y chromosome deletions (control samples) were also studied. Control samples were also previously characterized at the molecular level, using Y chromosome specific STS (Single Tagged Site) concerning these

three AZF regions: AZFa – DFFRY3', DBY; AZFb – sY1227, sY1224, sY134, sY119, sY134, RBMY1, sY143; AZFc – sY1192, sY254, RRM3, sY1291, sY283, sY1201.

DNA extraction was done by “Salting in salting out” method with Wizard Genomic DNA kit (Promega) and, more recently, automatic extraction with MagNa Pure LC DNA Isolation Kit (Roche). AmpFI STR® Yfiler® PCR Amplification kit (Applied Biosystem) was used for studying the following 16 *loci* – DYS456, DYS389I, DYS390, DYS389II, DYS458, DYS19, DYS385, DYS393, DYS391, DYS439, DYS635, DYS392, GATAH4, DYS437, DYS438 and DYS448. Capillary electrophoresis was performed in a 3130 Genetic Analyser with 36 cm capillaries with POP<sup>TM</sup>4 (Applied Biosystems), using specific software GeneMapper IDv3.2.

## Results

Forty-six samples were previously screened for AZF region deletions by using Y chromosome specific STS (Single Tagged Site) (Table1). DNA samples from these individuals were provided as blind samples, not being known how many individuals had AZF microdeletions. Four cases with no amplification of DYS385, DYS392 and DYS488 *loci* were detected (Fig.1 and Table 2), suggesting microdeletions in the AZFb region (Table2). From the Y-chromosome map positions, DYS385, 392 and 448 are located in the AZFb region, while DYS434, 437, 435, 439, 389I/II, 388, 438 and 436 are located in the AZFa region. DYS 391, 393 and 19 are not assigned to these regions. Using Yfiler multiplex system, rare alleles were observed in DYS458 (allele17.2), DYS385 (allele12.1) and GATAH4 (allele14.1) *loci*. Concerning control samples, there was no suggestion of microdeletions although some rare alleles were also present in DYS458 (alleles16.2, 17.2 and 18.2) and DYS635 (allele17). A complete Y-STR profile was detected in a sample belonging to a female individual, although with a 46, XY karyotype (Fig.2).

## Discussion and Conclusions

The existence of Y-chromosome microdeletions is a phenomenon with which Forensic Geneticists may have to deal. In case of such an occurrence, is of great importance to know how to interpret the nature of these results, given the structural changes of the Y-chromosome. In all control samples, a full Y-STR profile was obtained, confirming molecular information previously obtained with STS analysis. Four cases of non amplification of DYS385, DYS392 and DYS488 *loci* were detected, suggesting microdeletions in the AZFb region which was confirmed by single tagged site studies. Rare alleles not previously detected in Portuguese population studies were also encountered in several Y-STR *loci*.

A complete Y-STR profile was detected in a sample belonging to a female individual, although with a 46, XY karyotype. This is a well known clinical situation, even though of rare occurrence. Complete Y-STR profiles in samples from female gender individuals may raise ethical implications and, above all, increase the difficulty in the interpretation of physical evidence in criminal investigation casework.

## References

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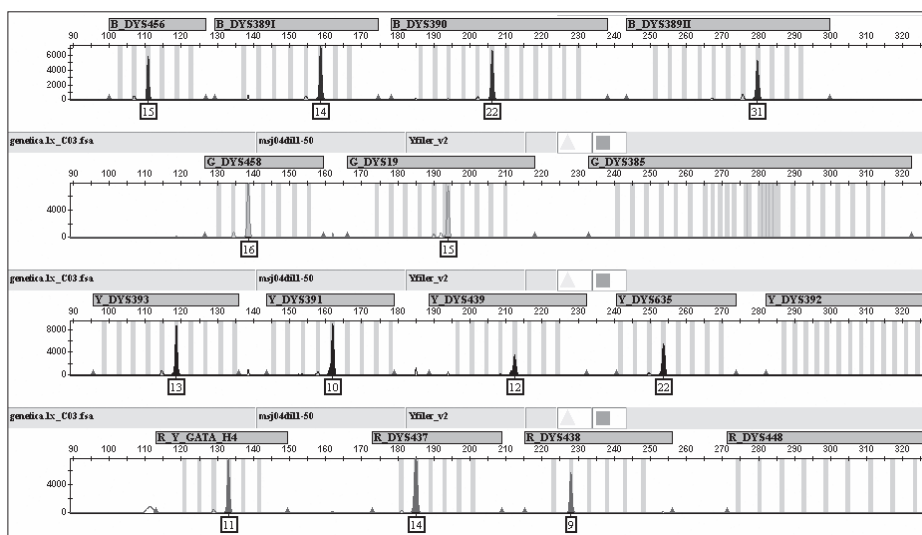


Figure 1 – Y-STR profile performed with Yfiler showing no amplification in *DYS385*, *DYS392* and *DYS448* loci

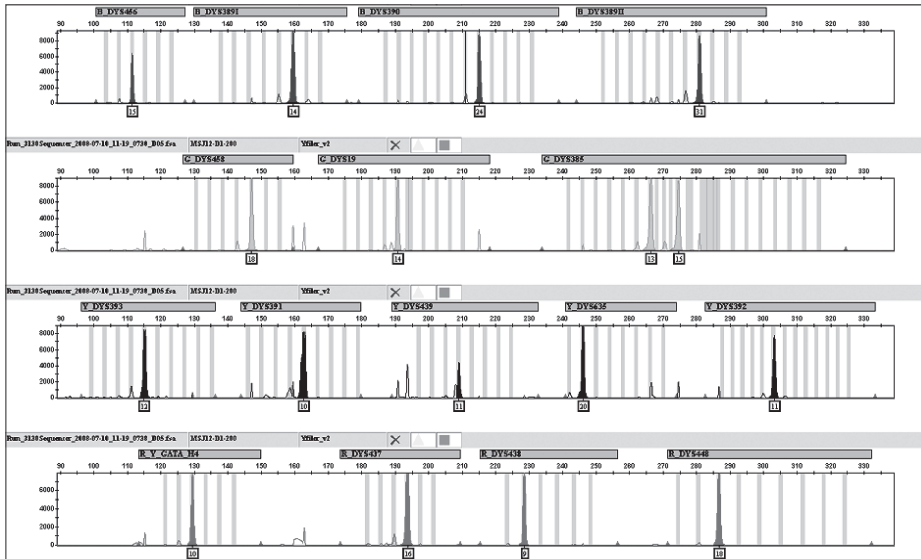


Figure 2 – A complete Y-STR profile was detected in a sample belonging to a female individual, although with a 46, XY karyotype.

AZF Region	STS's	Sample1	Sample2	Sample3	Sample4
AZFa	<i>DFFRY-3'</i>	Present	Present	Present	Present
	<i>DBY</i>	Present	Present	Present	Present
AZFb	sY1227	-	-	-	-
	sY1224	-	-	-	-
	sY119	-	-	-	-
	sY134	-	-	-	-
	<i>RBMY1</i>	-	-	-	-
AZFc	sY1192	Present	-	-	-
	sY254	Present	-	-	-
	<i>RRM3</i>	Present	-	-	-
	sY1291	Present	-	-	-
	sY283	Present	-	Present	-
	sY1201	Present	-	Present	-

Table 1 – Four examples of microdeletion results obtained with single tagged sites concerning AZF regions.

Amostras	Sexo	DYS19	DYS389I	DYS389II	DYS456	DYS390	DYS458	DYS385	DYS393	DYS391	DYS439	DYS635	DYS392	GATAH4	DYS437	DYS438	DYS448
1	M	14	13	29	15	23	18	11-14	13	11	12	23	13	12	15	12	19
	M	14	13	29	15	23	17.2	14-17	12	10	12	20	11	11	14	10	20
	M	14	13	29	16	23	18	?	13	11	13	23	?	11	15	12	?
	M	14	13	29	16	24	17	11-13	13	11	12	24	13	12	15	12	19
2	M	14	13	30	16	24	16	11-12.1	12	11	12	23	13	12	15	12	19
	M	14	14	31	16	25	17	11-15	13	11	10	23	13	12	15	12	19
	M	15	12	29	15	22	16	14	14	10	11	20	11	13	16	10	21
	M	15	13	29	15	24	17	?	13	12	14	23	?	14.1	15	12	?
3	M	15	13	30	15	24	17	11-14	13	11	12	23	13	12	15	12	19
	M	15	13	31	15	24	15	?	12	10	11	21	?	10	14	10	?
	M	15	14	29	17	24	16	11-15	13	10	11	23	13	12	15	12	17
	M	15	14	30	15	24	17	11-14	13	11	11	23	13	12	15	12	19
4	M	15	14	31	15	22	16	?	13	10	12	22	?	11	14	9	?
	M	15	14	31	15	22	16	?	13	10	12	22	?	11	14	9	?

Table 2 – Examples of Y-STR profiles showing 4 samples with no amplification in *DYS385*, *DYS392* and *DYS448* loci (samples in yellow).