b2/b3 deletion is associated with spermatogenic failure in men from the Republic of Macedonia

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INTRODUCTION

The b2/b3 deletion is a common Y chromosome partial deletion that is preceded by gr/gr inversion and which removes 1,8 Mb DNA segment (Figure 1), including 12 members of 8 testis-specific gene families. It was suggested that geographical and ethnic differences might affect the Y chromosomal background and phenotypic expression of this deletion. The association of the b2/b3 deletion with male infertility has been reported in Chinese men, whereas no predisposition was detected in several other populations. The b2/b3 deletion was found fixed in the Y haplogroup N which is widely distributed among men in the Northern Europe. It was present with low frequencies among other European populations.

MATERIALS AND METHODS

We studied the occurrence of partial AZFc deletions among 77 men with azoospermia, 93 with oligozoospermia, 44 normozoospermic individuals with unexplained couple infertility and 233 fertile controls (proven fathers). The methodology for detection and characterization of b2/b3 deletion included analysis of DAZ, MYPT2Y and CDY1 gene copy number by quantitative fluorescent (QF)-PCR analysis where the b2/b3 deletion is characterized by reduced DAZ/DAZL ratio, increased CDY2/CDY1 ratio and normal MYPT2Y/MYPT2 ratio (Figure 2), and PCR analysis of several sequence tagged site (STS) (Figure 4).

The Y chromosome haplogroups were determined by 28 Y-chromosome SNP markers, which were typed by multiplex PCR/SNaPshot reactions (Figure 3), organized in five different multiplexes.

RESULTS

We found 6 men with b2/b3 deletion (Table 1), one azoospermic men, 4 oligozoospermic and one fertile control. The b2/b3 deletion was significantly more frequent among men with impaired spermatogenesis (5 out of 170 men or 2,9%) than among fertile controls (1 out of 233 or 0,4%; p=0,04). The highest frequency was present in oligozoospermic men (4 out 93 or 4,3%; p=0,01). There was a difference in the distribution of the Y haplogroups among males with b2/b3 deletion; all five infertile men belonged to haplogroup E3b1, while the only fertile men belonged to haplogroup N3.

CONCLUSION

b2/b3 partial deletion is associated with impaired spermatogenesis in men from Republic of Macedonia.

ACKNOWLEDGMENTS

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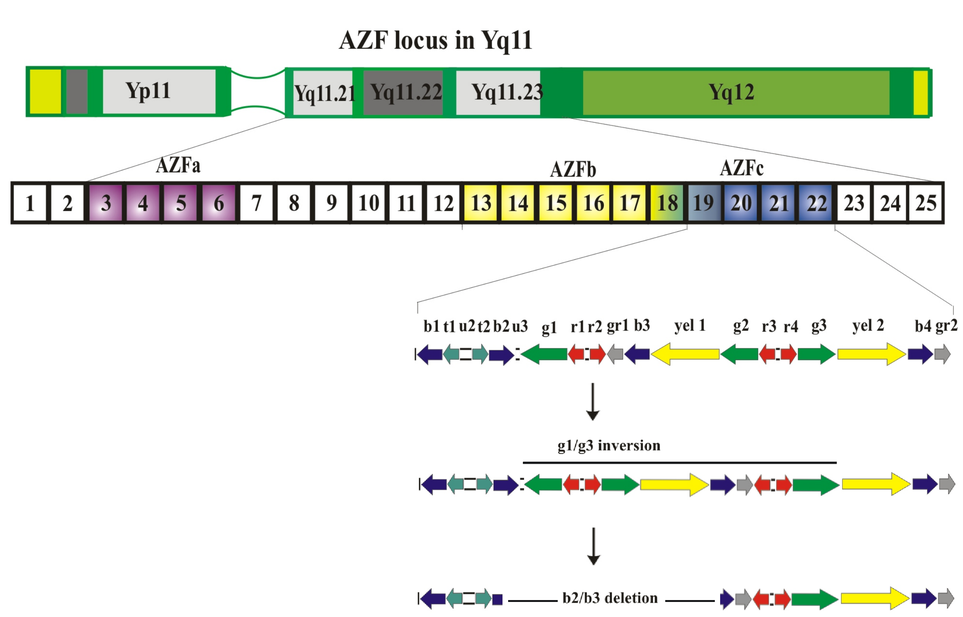

Figure 1. Schematic representation of the b2/b2 deletion in the AZF locus on Y chromosome.

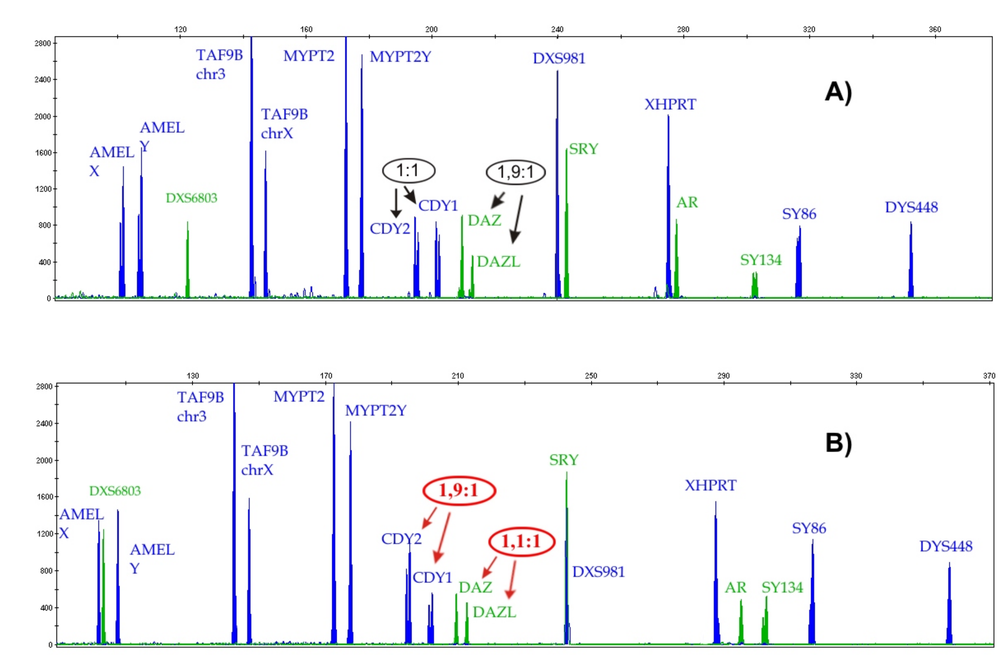

Figure 2. Electrophoreograms of QF-PCR analysis from normal male (A) and male with b2/b3 deletion (B).

Figure 3. Agarose gel electrophoresis of the duplex PCR of sY 1291 and sY1191 STS markers. 1) DNA ladder; 2-5, normal male DNA samples; 6, DNA from a men with b2/b3 deletion, 7) DNA from a men with gr/gr deletion.

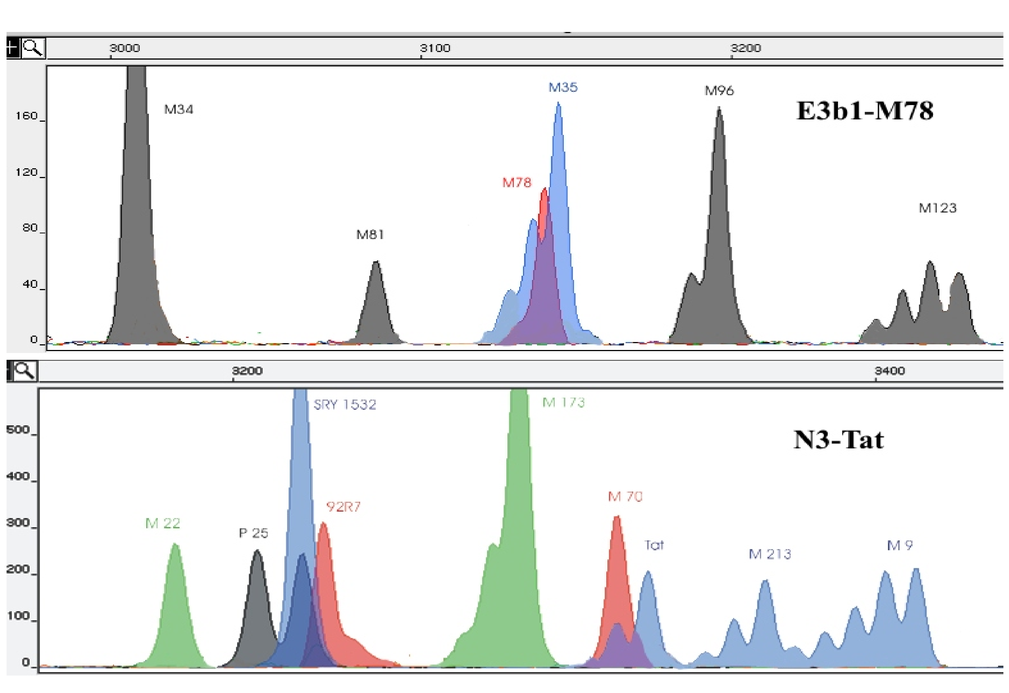

Figure 3. SNaPshot multiplexes from samples with E3b1 and N3 haplogroups