

Y

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3  
1 . 1 . 1 . 2 . 2  
3 . 3 . 3 . 1

### Relationship between Microdeletions on the Y Chromosome and Defect of Spermatogenesis

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**Objectives:** To estimate the frequency of Y chromosome microdeletions in the Korean population of infertile men and to evaluate the relationship between microdeletion on the Y chromosome and clinical phenotypes of infertile men with idiopathic azoospermia and oligozoospermia.

**Materials and Methods:** Genomic DNA was extracted from blood samples collected from 330 infertile men attending the Infertility Clinic at Samsung Cheil Hospital, Korea. Six sequence tagged sites (STSs) spanning the azoospermia factor (AZF) regions of the Y chromosome were amplified by polymerase chain reactions (PCRs).

**Results:** Microdeletions on Y chromosome were detected in 35 (10.6%) of the 330 infertile men. Most of the microdeletions (91.4%) involved AZFb or AZFc. The high incidence of microdeletions were found in AZFc region (57.1%), but the low in AZFa (8.6%) and AZFb (5.7%). Larger microdeletions involving two or three AZF regions were detected in 28.6% of cases. All patients (6 patients) with deletion of AZFa region showed no germ cell phenotypes, Sertoli cell only syndrome or Leydig cell hyperplasia in histopathologic examinations.

**Conclusion:** Microdeletions on the Y chromosome, especially, at AZFc/DAZ regions may be the major cause of azoospermia and severe oligozoospermia. We suggest that idiopathic infertile men have genetic counselling and microdeletion analysis on the Y chromosome before IVF-ET and ART program.

**Key Words:** Y chromosome, Microdeletion, Azoospermia factor (AZF), Genetic counselling

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(R01-2000-000-00147-0)

2% , screening

10~15% 가 , (intracytoplasmic sperm injection, ICSI)

50% , (testicular sperm extraction, TESE)

Y 가 ,

1-7 Y 가

Zuffardi<sup>8</sup>가 Y (Yq11) Tiepolo 가

oospermia Factor (AZF) , , Az<sup>12,13</sup> Y 가

Y STS-based mapping strategy

Y

AZF 가 가

Ma<sup>9</sup> interval 6 , *YRRM1*

*YRRM2* RNA recognition motif 가

RNA processing

translational control 1.

Y 2000 1 2002 8

AZF

Reijo<sup>10</sup> (<math>2 \times 10^6</math>)

*Deleted in Azoospermia (DAZ)* ml) 330

*YRRM1 YRRM2* Y

가 RNA binding motif 10 , 가

30

Vogt<sup>3</sup> AZFa, AZFb AZFc

3 subregion , 3 subregion

2. STS Primers

Y primers

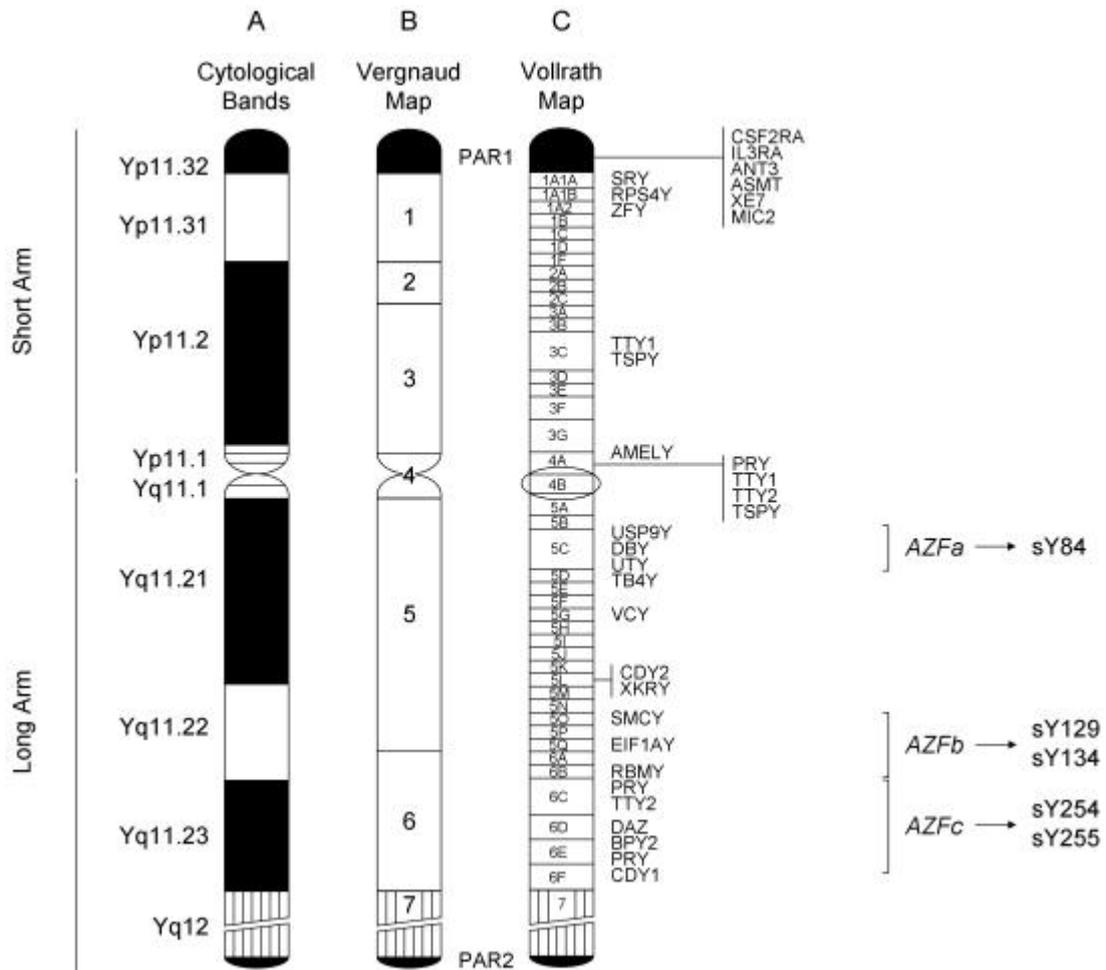
cell only syndrome (SCOS) Sertoli Vollrath<sup>14</sup> Reijo<sup>2,20</sup>

region maturation arrest (MA) , AZFb primers Lee<sup>4</sup>

가 sY84 (AZFa region), sY129, sY134 (AZFb region), sY254, sY255 (AZFc region, *DAZ*) (Figure 1 and Table 1).

10,11 Y

Y SRY primer



**Figure 1.** A. Representation of the cytological bands of the Y chromosome. B. The seven intervals of the Vergnaud map of the Y chromosome (Vergnaud *et al.*, 1986), where intervals 1 ~4 span the short arm and the centromere, intervals 5 and 6 span the euchromatic region, and interval 7 spans the heterochromatic region. C. The 43 interval map of the Y chromosome (Vollrath *et al.*, 1992). The list of genes mapped to the Y chromosome, the location of the AZF regions and the STS markers used in this study are shown.

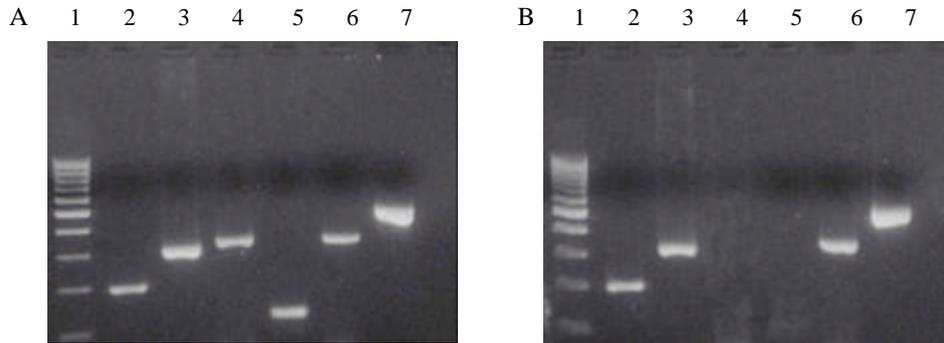
primer ( ) Bioneer -20

3. Genomic DNA EDTA가 va-

4. (Polymerase Chain Re - action, PCR)

primers sY84, sY129, sY134, sY254, sY255, SRY 20

10 mM Tris-HCl (pH 8.3), 50 mM KCl, 1.5 mM MgCl<sub>2</sub>, 0.2 mM dNTP, 20 pmol primer , 0.5 units Taq DNA polymerase (Boehringer Mannheim,



**Figure 2.** Microdeletion pattern of the AZF region in the normal and azoospermic patient. **A.** Normal fertile man. **B.** Azoospermia patient. Polymerase chain reaction products analyzed on the 2% agarose gel electrophoresis show deletion of sY254 and sY255 in the AZFc region. Lane 1, 100 bp ladder; lane 2, sY129; lane 3, sY134; lane 4, sY254; lane 5, sY255; lane 6, sY84; lane 7, *SRY*.

**Table 1.** Primer sequences of Y-chromosome STSs analyzed

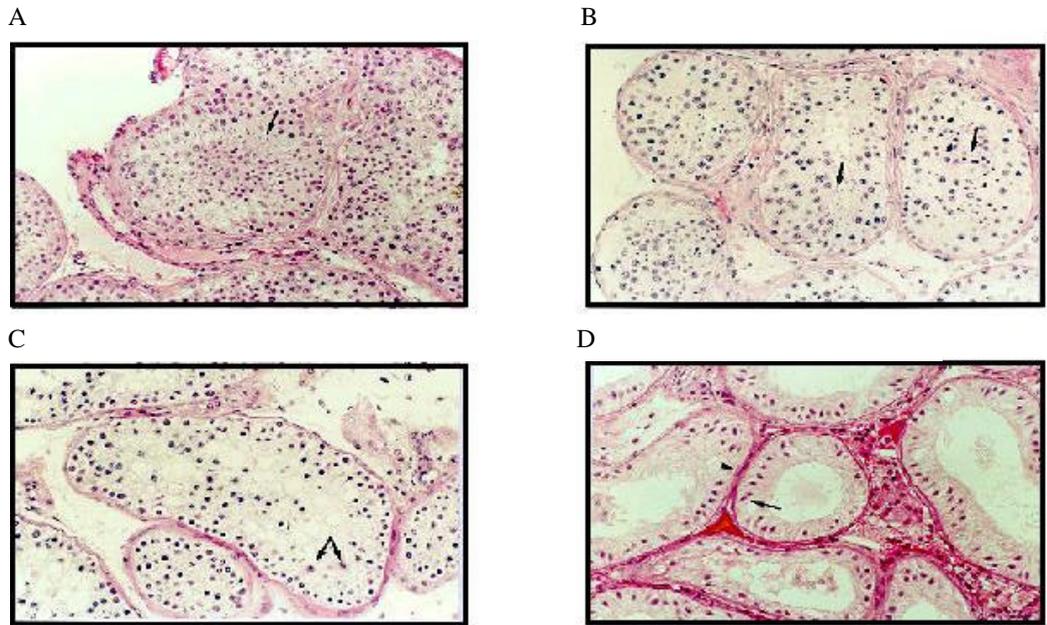
STSs*	Forward	Reverse
sY84	AgAAgggTCTgAAAgCAggT	gCCTACTACCTggAggCTTC
sY129	AgCTTCAggAggTTCAAAAC	AAgTgggACCTAAgCTACgA
sY134	gTCTgCCTCACCATAAAACg	ACCACTgCCAAAACCTTTCAA
sY254	gggTgTTACCAgAAggCAAA	gAACCGTATCTACCAAAgCAgC
sY255	gTTACAggATTCggCgTgAT	CTCgTCATgTgCAgCCAC
<i>SRY</i>	gAATATTCCCgCTCTCCggA	gCTggTgCTCCATTCTTgAg

\* STSs: sequence tagged sites.

**Table 2.** The incidence of Y chromosome microdeletion in 330 infertile men

	Total (n=330)	AZFa	AZFb	AZFc	AZFb, c	AZFa, b, c
Deletion (%)	35 (10.6)	3 (8.6)	2 (5.7)	20 (57.1)	5 (14.3)	5 (14.3)

Germany), 50~100 ng genomic DNA  
DNA thermal cycler (Strata-  
gene, USA) 94 2  
94, 40; 60 62, 1; 72, 1 cycle 1. Y  
35 72 10  
. 2% agarose gel primer 가  
가  
3 , 330



**Figure 3.** Testicular histology of normal and azoospermic patients. H&E staining (200X). **A.** normal spermatogenesis. Sperm indicated by arrow. **B.** Hypospermatogenesis. Arrow showed sperms. **C.** Maturation arrest. There are reduced numbers of spermatocytes, but no spermatids. Spermatocyte indicated by arrow. **D.** Germ cell aplasia (Sertoli cell only syndrome). Sertoli cell indicated by arrow and seminiferous tubules indicated by arrow head.

STS	Y	AZFa region	AZFb region	AZFc region	subregion
35	(10.6%)	3	(8.6%)	2	(5.7%)
20	(57.1%)	2		10	(28.6%)
32	(91.4%)				

2.

35 가

20 Y

(Figure

3) AZFa region 3

Sertoli cell only syndrome (SCO) AZFa,

AZFb, AZFc 3

2 SCO, 1 Leydig cell hyperplasia

. AZFb region 1 severe hypo-

spermatogenesis AZFb AZFc 7

4 3 SCO, 1 se-

vere hypospermatogenesis . AZFc

9 5 SCO, 3

severe hypospermatogenesis, 1 maturation arrest

(Table 3).

Y

**Table 3.** Testicular phenotypes of histological observations associated with microdeletions restricted to AZFa, AZFb, and AZFc

	Total	AZFa	AZFb	AZFc	AZFb, c	AZFa, b, c
Patients with microdeletion	35	3	2	20	5	5
Histological observations	20	3	1	9	4	3
SCO	13	3		5	3	2
MA	1			1		
Hypo	5		1	3	1	
Leydig cell hyperplasia	1					1

SCO: Sertoli cell only, MA: maturation arrest, Hypo: (severe) hypospermatogenesis

1~55%

. 1998

5,6 12.2%

3.4% .15

35 (10.6%)

TESE ICSI

(가, , ,

Y

)

3.5~20%

16-19

, ICSI

Y

가

Y

Y

Vogt 3

AZFa SCO, AZFb maturation arrest, AZFc SCO hypospermatogenesis

20

AZFa re-

gion 3 Sertoli cell only syndrome (SCO), AZFa region

AZFb AZFc가 3 2 SCO, 1 Leydig cell hyperplasia

AZFa region

SCO

Vogt 3

AZFb region

1 severe hypo spermatogenesis AZFb AZFc가

4 3 SCO, 1 se- vere hypospermatogenesis Vogt

. AZFc

9 5 SCO, 3 severe hypospermatogenesis, 1 maturation arrest

가 가

, 가

가

AZF subregion, genotype pheno- type

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