

Association study of nine polymorphisms in eight different genes (FASLG, JMJDIA, LOC203413, TEX15, BRDT, OR2W3, INSR and TAS2R38) with male infertility

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INTRODUCTION

The analysis of polymorphisms in genes involved in spermatogenesis represents an exciting area of research in genetics of male infertility. Polymorphisms in these genes are considered potential risk factors which may contribute to the severity of spermatogenic failure. Here we have investigated the possible association of nine single nucleotide polymorphisms (SNPs) in eight different genes (FASLG, JMJDIA, LOC203413, TEX15, BRDT, OR2W3, INSR and TAS2R38) with male infertility. The SNPs were selected because they were found to be associated with azoospermia and/or oligozoospermia in a recent study (Aston KI et al, Hum Reprod, 25:1383-97, 2010).

MATERIALS AND METHODS

We have analysed a total of 136 men with idiopathic infertility (60 azoospermic and 76 oligozoospermic) and 161 fertile controls. Ninety three of the infertile men were Macedonians, 32 were Albanians and 11 men were of other ethnic origin. The control group was composed of 125 Macedonian and 36 Albanian men. The methodology included multiplex PCR/SNaPshot method, followed by capillary electrophoresis on ABI3130 Genetic Analyzer (Figure 1).

RESULTS

The allele frequencies of the nine studied SNPs among the whole group of infertile men are given in Table 3, while those in azoospermic and oligozoospermic men are given in Tables 4 and 5, respectively. To exclude the possible ethnic differences in the distribution of the studied SNPs, Macedonian and Albanian infertile men were also considered separately in comparison with Macedonian and Albanian fertile controls, respectively. No deviations from Hardy Weinberg equilibrium was observed within control groups for all studied SNPs.

Of the nine SNPs evaluated, we have found significant association ($p < 0.05$) for three. The most significant association was found for rs5911500, an intergenic SNP located on the X chromosome. This SNP as well as rs3088232 in BRDT gene showed association with azoospermia both among Macedonians and Albanians, while rs11204546 in OR2W3 gene was also associated with azoospermia, but only among Macedonian men. Another SNP (rs34605051 in JMJDIA gene) was present with higher frequency among all groups of infertile men in comparison to fertile controls, but the differences were not significant.

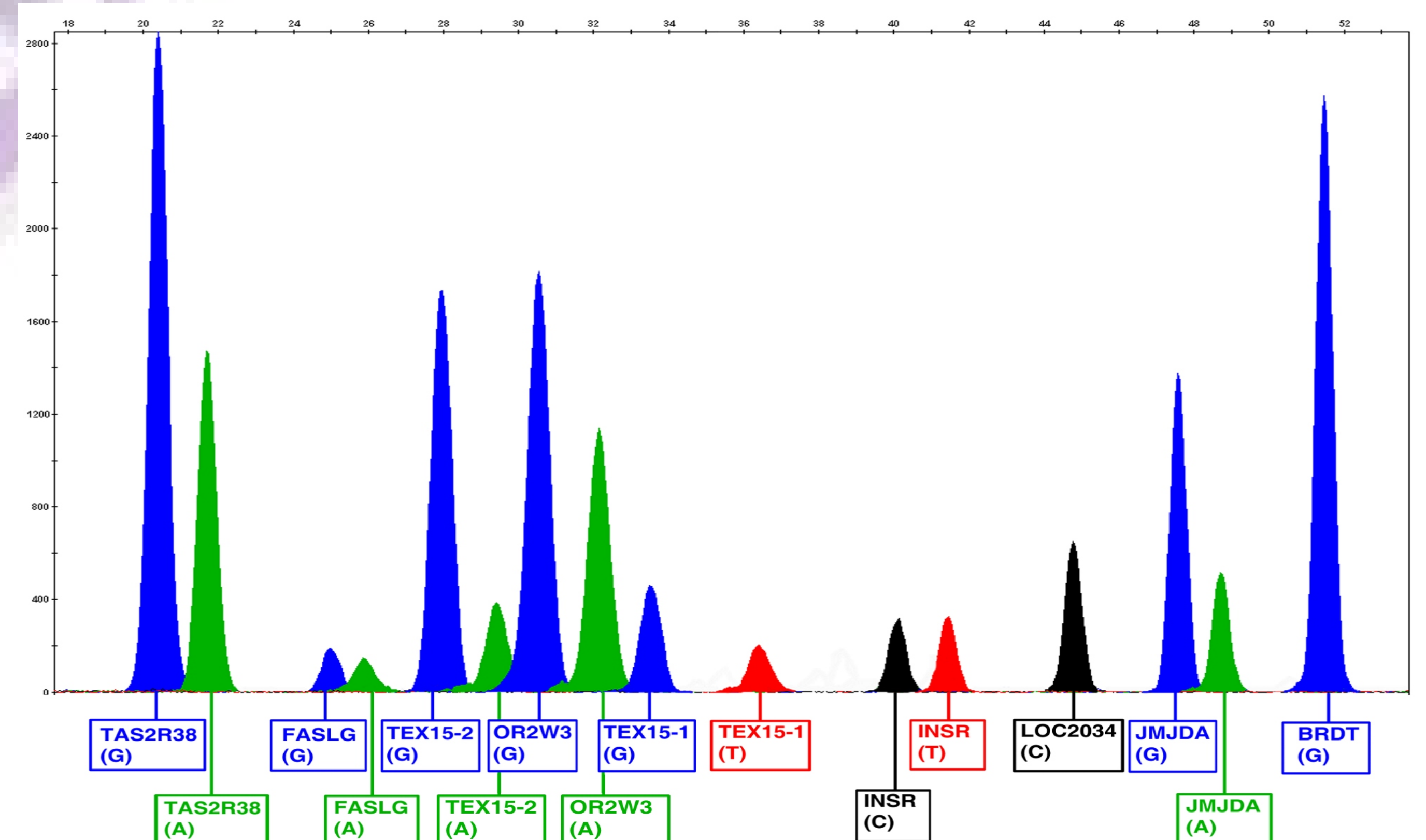


Figure 1. Electrophoreogram of the multiplex SNaPshot assay for the detection of nine SNPs in a DNA sample heterozygous for seven SNPs: rs10246939 in TAS2R38, rs763110 in FASLG G/A, rs323344 and rs323345 in TEX15, rs11204546 in OR2W3, rs2059807 in INSR and rs34605051 in JMJDIA.

Table 1. Allele frequencies of nine studied polymorphisms among infertile men

SNP	Allele	Infertile males						Fertile control males					
		Total (n=272)		Macedonians (n=186)		Albanians (n=64)		Total (n=322)		Macedonians (n=250)		Albanians (n=72)	
		No	%	No	%	No	%	No	%	No	%	No	%
LOC2034	C	216	79.4	146	78.5	48	75	282	87.6	212	84.8	70	97.2
	T	56	20.6	40	21.5	16	25	40	12.4	38	15.2	2	2.8
OR2W3	G	163	59.9	108	58.1	39	60.9	210	65.2	171	68.4	39	54.2
	A	109	40.1	78	41.9	25	39.1	112	34.8	79	31.6	33	45.8
INSR	C	178	65.4	122	65.6	43	67.2	204	63.4	150	60	54	75
	T	94	34.6	64	34.4	21	32.8	118	36.6	100	40	18	25
TAS2R38	A	141	51.8	95	51.1	37	57.8	173	53.7	130	52	43	59.7
	G	131	48.2	91	48.9	27	42.2	149	46.3	120	48	29	40.3
JMJDIA	A	213	78.3	143	76.9	52	81.3	267	82.9	205	82	62	86.1
	G	59	21.7	43	23.1	12	18.7	55	17.1	45	18	10	13.9
TEX15	T	250	91.9	167	89.8	61	95.3	289	89.8	221	88.4	68	94.4
	G	22	8.1	19	10.2	3	4.7	33	10.2	29	11.6	4	5.6
TEX15-2	A	250	91.9	167	89.8	61	95.3	289	89.8	221	88.4	68	94.4
	G	22	8.1	19	10.2	3	4.7	33	10.2	29	11.6	4	5.6
BRDT	C	204	75	137	73.7	50	78.1	268	83.2	206	82.4	62	86.1
	G	68	25	49	26.3	14	21.9	55	17.1	44	17.6	10	13.9
FASLG	G	165	60.7	117	62.9	38	59.4	202	62.7	157	62.8	45	62.5
	A	107	39.3	69	37.1	26	40.6	120	37.3	93	37.2	27	37.5

Table 3. Allele frequencies of nine studied polymorphisms among infertile men with oligozoospermia

SNP	Allele	Infertile males with oligozoospermia						Fertile control males					
		Total (n=152)		Macedonians (n=104)		Albanians (n=34)		Total (n=322)		Macedonians (n=250)		Albanians (n=72)	
		No	%	No	%	No	%	No	%	No	%	No	%
LOC2034	C	122	80.3	82	78.8	26	76.5	282	87.6	212	84.8	70	97.2
	T	30	19.7	22	21.2	8	23.5	40	12.4	38	15.2	2	2.8
OR2W3	G	90	59.2	62	59.6	19	55.9	210	65.2	171	68.4	39	54.2
	A	62	40.8	42	40.4	15	44.1	112	34.8	79	31.6	33	45.8
INSR	C	104	68.4	70	67.3	24	70.6	204	63.4	150	60	54	75
	T	48	31.6	34	32.7	10	29.4	118	36.6	100	40	18	25
TAS2R38	A	81	53.3	52	50	23	67.6	173	53.7	130	52	43	59.7
	G	71	46.7	52	50	11	32.4	149	46.3	120	48	29	40.3
JMJDIA	A	120	78.9	83	79.8	26	76.5	267	82.9	205	82	62	86.1
	G	32	21.1	21	20.2	8	23.5	55	17.1	45	18	10	13.9
TEX15	T	142	93.4	95	91.3	33	97.1	289	89.8	221	88.4	68	94.4
	G	10	6.6	9	8.7	1	2.9	33	10.2	29	11.6	4	5.6
TEX15-2	A	142	93.4	95	91.3	33	97.1	289	89.8	221	88.4	68	94.4
	G	10	6.6	9	8.7	1	2.9	33	10.2	29	11.6	4	5.6
BRDT	C	120	78.9	81	77.9	28	82.4	268	83.2	206	82.4	62	86.1
	G	32	21.1	23	22.1	6	17.6	54	16.8	44	17.6	10	13.9
FASLG	G	91	59.9	63	60.6	22	64.7	202	62.7	157	62.8	45	62.5
	A	61	40.1	41	39.4	12	35.3	120	37.3	93	37.2	27	37.5

Table 2. Allele frequencies of nine studied polymorphisms among infertile men with azoospermia

SNP	Allele	Infertile males with azoospermia						Fertile control males					
		Total (n=120)		Macedonians (n=82)		Albanians (n=30)		Total (n=322)		Macedonians (n=250)		Albanians (n=72)	
		No	%	No	%	No	%	No	%	No	%	No	%
LOC2034	C	94	78.3	64	78	22	73.3	282	87.6	212	84.8	70	97.2
	T	26	21.7	18	22	8	26.7	40	12.4	38	15.2	2	2.8
OR2W3	G	73	60.8	46	56.1	20	66.7	210	65.2	171	68.4	39	54.2
	A	47	39.2	36	43.9	10	33.3	112	34.8	79	31.6	33	45.8
INSR	C	74	61.7	52	63.4	19	63.3	204	63.4	150	60	54	75
	T	46	38.3	30	36.6	11	36.7	118	36.6	100	40	18	25
TAS2R38	A	60	50	43	52.4	14	46.7	173	53.7	130	52	43	59.7
	G	60	50	39	47.6	16	53.3	149	46.3	120	48	29	40.3
JMJDIA	A	93	77.5	60	73.2	26	86.7	267	82.9	205	82	62	86.1
	G	27	22.5	22	26.8	4	13.3	55	17.1	45	18	10	13.9
TEX15	T	108	90	72	87.8	28	93.3	289	89.8	221	88.4	68	94.4
	G	12	10	10	12.2	2	6.7	33	10.2	29	11.6	4	5.6
TEX15-2	A	108	90	72	87.8	28	93.3	289	89.8	221	88.4	68	94.4
	G	12	10	10	12.2	2	6.7	33	10.2	29	11.6	4	5.6
BRDT	C	84	70	56	68.3	22	73.3	268	83.2	206	82.4	62	86.1
	G	36	30	26	31.7	8	26.7	54	16.8	44	17.6	10	13.9
FASLG	G	74	61.7	54	65.9	16	53.3	202	62.7	157	62.8	45	62.5
	A	46	38.3	28	34.1	14	46.7	120	37.3	93	37.2	27	37.5

CONCLUSIONS

In conclusion, we have studied nine SNPs that were previously found in association with azoospermia and oligozoospermia and we have confirmed the association for three of them (SNPs rs5911500 in LOC203413, rs3088232 in BRDT and rs11204546 in OR2W3 gene) among Macedonian and/or Albanian azoospermic and/or oligozoospermic patients.

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