MERIT RESEARCH JOURNALS
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Merit Research Journal of Medicine and Medical Sciences (ISSN: 2354-323X) Vol. 5(8) pp. 403-410, August, 2017
Available online http://www.meritresearchjournals.org/mms/index.htm

Original Research Article

G1733A Polymorphism of the Androgen Receptor Gene and Recurrent Spontaneous Abortion in Saudi's Women

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Abstract

Various gene polymorphisms have been reported to influence the risk of recurrent spontaneous abortion (RSA). In females, androgen receptor (AR) is expressed in reproductive tissues; however, the causal association of androgens receptors (AR: G1733A) in RSA has not been investigated in Saudi's women with RSA. The aim of the present study was to investigate the potential effect of the G1733A polymorphism of the AR gene in the occurrence of RSA in Saudi women. The study group comprised of 65 Saudi women with unexplained RSA. The reference population consisted of 65 Saudi women who had at least 3 children, and were without known pregnancy losses or any known medical illnesses. Peripheral venous puncture, DNA extraction, PCR, and sequencing were employed to genotype women for the presence of a polymorphism at position G1733A of androgen receptor gene. For the G1733A polymorphism, the frequency of the A allele in the Saudi healthy females is 16.2%. The frequencies of homozygous AA were 6.2% and 0% in the control and RSA group, respectively, but the difference was not significant (p>0.05). In addition the frequency of heterozygous GA was 20% for the control group and 26.2% for the patient group but the difference was not significant (P>0.05). Regarding the G1729A polymorphism, the frequency of the A allele in the Saudi females was 3.1%. The frequencies of GA+AA were 6.2% and 1.5% in the control and patient groups, respectively, and the difference was not significant (p>0.05). These findings confirmed that there were no causal association between the occurrence of the A allele of the G1733A and G1729A polymorphisms of the AR gene and increased risk of RSA in Saudi's women.

Keywords: Recurrent spontaneous abortion, Androgen receptor, Gene polymorphism, Risk factor.

INTRODUCTION

Recurrent spontaneous abortion (RSA) is a clinical problem and occurs in women of reproductive age with a frequency of 1%–3% (Regan, 1998; Pandey et al., 2005; Shin et al., 2010). According to Royal College of Obstetrician and Gynaecologists, recurrent abortion is defined as 3 or more consecutive fetal loss before 20 weeks of gestation (green top guideline 17) (Li et al., 2002; Jahaninejad et al., 2013; Regan et al., 2010). The pathogenesis of RSA is very complicated (Xiao et al., 2014). The risk of miscarriage in a subsequent pregnancy is about 40 % to 50 %in women with a history of recurrent miscarriage (Jahaninejad et al., 2013). It is enhanced by a variety of factors including advanced maternal age, anatomic abnormality, chromosome abnormalities, hormonal disorders, hereditary thrombophilia, immune-logic factors, infections, and nutritional and environmental

factors (Regan et al., 2010; Xiao et al., 2014; Rai and Regan, 2006; Redline, 2006; Sereshki et al., 2014). Although various studies have been conducted to elucidate the factors responsible for this disorder (Suzumori and Sugiura-Ogasawara, 2010; Beaman et al., 2012), up to 50 % of RSA cases are not clearly understood (Wilczynski et al., 2012). Various gene polymorphisms have been hypothesized to influence the risk of RSA, and research interest has been persistent on association studies between different genetic polymorphisms and RSA but the results from different studies are often controversial (Hefler et al., 2002; Buchholz et al., 2004; Papazoglou et al., 2005; Zammiti et al., 2006; Karvela et al., 2010; Kim et al., 2014).

Androgens are lipophilic hormones with several physiologic effects in both sexes (Quigley et al., 1995). In females, androgens production is very important for decidualization, a process that controls embryo implantation and placentation (Guay et al., 2004). Androgen receptor (AR) is commonly expressed in female reproductive tissue such as endometrium (Apparao et al., 2002). AR gene encodes a ligand-activated nuclear transcription factor that is activated by steroid hormones such as testosterone or dihydrotestosterone (Roy et al., 1999; Gelmann, 2002). Activated AR is transferred into the nucleus, where it regulates the transcription of androgen-responsive genes (Burger, 2002).

The AR gene is highly conserved, located on the chromosome Xq11-12 locus and is composed of eight exons (Brown et al., 1989). In humans, AR is a 110 kD protein composed of 919 amino acids (Trapman et al., 1988). The AR contains four domains: the amino terminal activation domain (NTD); the DNA-binding domain (DBD); the hinge region (HR); and the carboxyl ligand-binding domain (LBD) (Chang et al., 1995; Yong et al., 1998). AR exon 1 encodes the entire N-terminal domain (NTD) (a.a. 1-556) which comprises the bulk of the AR and is the least conserved of the four domains. This variability allows AR to differentially recruit co-regulators conferring androgen specific transactivation (Eisermann et al., 2013). Exon 1 of the gene harbours two microsatellites, two polymorphic trinucleotide repeats (CAG and GGC). Between these two repeats, a single nucleotide polymorphism (SNP) determined as a G to A substitution (G1733A; rs6152) in the third nucleotide position of the 211 codon has been previously described (Esteban et al., 2005). This codon resides in the N-terminal domain of the AR, that harbors the major transcription activation functions (Grad et al., 2001), raising the possibility that any variation in this codon may affect AR transactivational activity (5). G1733A polymorphism is a synonymous change as the glutamic acid remains unchanged (35), and it occurs in 13% to 20% of Caucasian populations (Lu and Danielsen, 1996; Ellis et al., 2001).

It was reported that there is a statistically significant association between the A allele of the G1733A polymorphism and the increased risk of prostate cancer (Medeiros et al., 2003; Kucerova et al., 2014). Other studies showed that women with pre-eclampsia have significantly higher androgen levels when compared to controls (Carlsen et al., 2005; Saarela et al., 2005). Furthermore, it has been shown that androgens play a significant role in the development of hypertension (Bachmann et al., 1991; Mantzoros et al., 1995) and obesity (Gustafson et al., 2003). Around 1,000 mutations are reported in the AR (Gottlieb et al., 2012). While the number of mutations reported continues to rise, the relevance of these mutations in RSA requires more studies. Few studies suggested that G1733A polymorphism is associated with a higher risk for RSA in some populations (Jahaninejad et al., 2013; Karvela et al., 2010). However the causal relationship between the risk of RSA and G1733A polymorphism has not been poorly understood.

In view of the review presented above. The aim of our study was to determine whether the AR G1733A polymorphism is associated with an increased risk for RSA in Saudi women.

MATERIALS AND METHODS

Patients

This study was approved by the Institutional Review Board (IRB), College of Medicine, King Saud University.

The patients participating in this study were referred to the Recurrent Abortion Clinic, King Khaled University Hospital, Riyadh, Kingdom of Saudi Arabia from January 2010 to January 2011. There were no ethnic differences between cases and controls. 60 % of the invited cases and controls declined to participate. The study group comprised of 65 women (mean age: 34.1 ± 6.2 years, range: 15-45 years) with unexplained RSA, consecutively referred. The reference population (controls) consisted of 65 women who had at least 3 children, and were without known pregnancy losses or any known medical illnesses. Routine analysis at the hospital laboratory were performed to exclude known causes of abortion: parental karyotypes; hormone levels; toxoplasmosis; cytomegalovirus; rubella; antiphospholipid antibodies; protein C; protein S; glucose level; hysteroscopy; hysterosalpingography; and serial ultrasound when needed. The criteria for inclusion were: females presenting with unexplained RSA after all the tests mentioned above were normal. The protocol of this investigation was approved by the Medical Ethics Committee of King Khalid University Hospital and the Ethical Committee of King Saud University, Riyadh, Saudi Arabia. All women were required to sign the informed consent form to participate.

Table 1. Demographic and clinical characteristics of unexplained recurrent spontaneous abortion patients in comparison with the control group

Parameter	Patients	Controls	P value
	Mean ± standard error of the mean		
Age, years	34.1 ±0.77	34.6 ±0.97	0.6
Height, m	156.97 ±0.75	158.56 ±0.68	0.414
Weight, Kg	75.15 ±2.45	71.34 ±1.64	0.126
BMI, Kg/m2	30.66 ±1.01	28.28 ±0.62	0.047*
No. of pregnancies	6.5 ±0.38	3.9 ±0.22	0.0001*
No. of children	2.1 ±0.27	3.8 ±0.22	0.04*

*statistically significant

Table 2. Genotype and allele frequencies of G1733A polymorphism of AR gene among patients with unexplained recurrent spontaneous abortion and controls.

Genotype/Allele	Patients (n= 65)	Controls (n= 65)	OR (95% CI)	*P* value
Genotype	*n (%)*	*n (%)*		
GG	48 (73.8)	48 (73.8)	1.00 (0.46-2.19)	1.00
GA	17 (26.2)	13 (20.0)	1.42 (0.62-3.22)	0.53
AA	0 (0)	4 (6.2)	0.11(0.01-1.98)	0.12
Allele				
G	113 (86.9)	109 (83.8)	0.78 (0.39-1.56)	0.60[a]
A	17 (13.1)	21 (16.2)		

[a]Fisher exact test.

Genotyping

Ten ml blood samples were extracted by venipuncture in ethylenediaminetetraacetic acid tubes from each case and control. Genomic DNA was extracted using the DNA Pure gene purification kit (Qiagen, Venlo, Netherlands). DNA was stored at 4°C until analysis. The purified DNA was amplified by PCR using sequence specific primers designed using the Primer3 program. The PCR amplification primers were as follows: forward primer: 5'-GAGGATGGTTCTCCCCAAG-3'; reverse primer: 5'-GGACTCAGATGCTCCAACG-3'.PCR amplification was carried out in 25 µl volume containing 5U/µl of hot star Taq-polymerase (Qiagen, Venlo, Netherlands), 2.5 mMdNTPs (Amersham Pharmacia Biotech, NJ, USA), 10 µM/µl of each primer (Invitrogen, CA, USA), and ~50 ng DNA. PCR conditions comprised an initial denaturing step at 95°C for 15 min, followed by 36 cycles of 95°C for 30sec, 60°C for 30 sec, and 72°C for 2 min, and a final extension at 72°C for 10 min. After PCR, The generated 500 bp amplicons were subjected to sequencing using Applied Biosystems Integrated (ABI) 3130xl Genetic Analyzer following the kit manufacture instructions.

Statistical analysis

All statistical analyses were carried out using the Statistical Package for Social Sciences version 12 for Microsoft Window (SPSS Inc, Chicago, IL, USA) and the genotype and allele frequencies of the patient were compared with control using X2 and Fisher exact test analysis and the difference was considered statistically significant if $p<0.05$.

RESULTS

The demographic and clinical characteristics of the patient and control groups are shown in Table 1. There were no significant differences between patients and controls concerning average age ($p = 0.6$). The average BMI ($P= 0.047$) and number of pregnancies ($P=0.0001$) of patients are significantly higher compared to the control women. The control women have significantly more live born children on average compared to patients ($P=0.04$).

This study was done to investigate G1733A polymorphism of the AR gene for 65 patients and 65 controls, the genotype and allele frequencies of G1733A polymorphism of AR gene among patients with RSA and controls are shown in Table 2. In the present study, 48 patients had the GG genotype, and the other 17 patients had the GA genotype. 48 control subjects had the GG genotype, 13 had the GA genotype, and the other 4 had the AA genotype. No mutant homozygous (AA) carriers were identified in the patient group. Figure 1 is showing the sequence diagrams of G1733A locus in AR gene. The frequencies of the GG and GA genotypes of the G1733A polymorphism were 73.8%, and 26.2%,

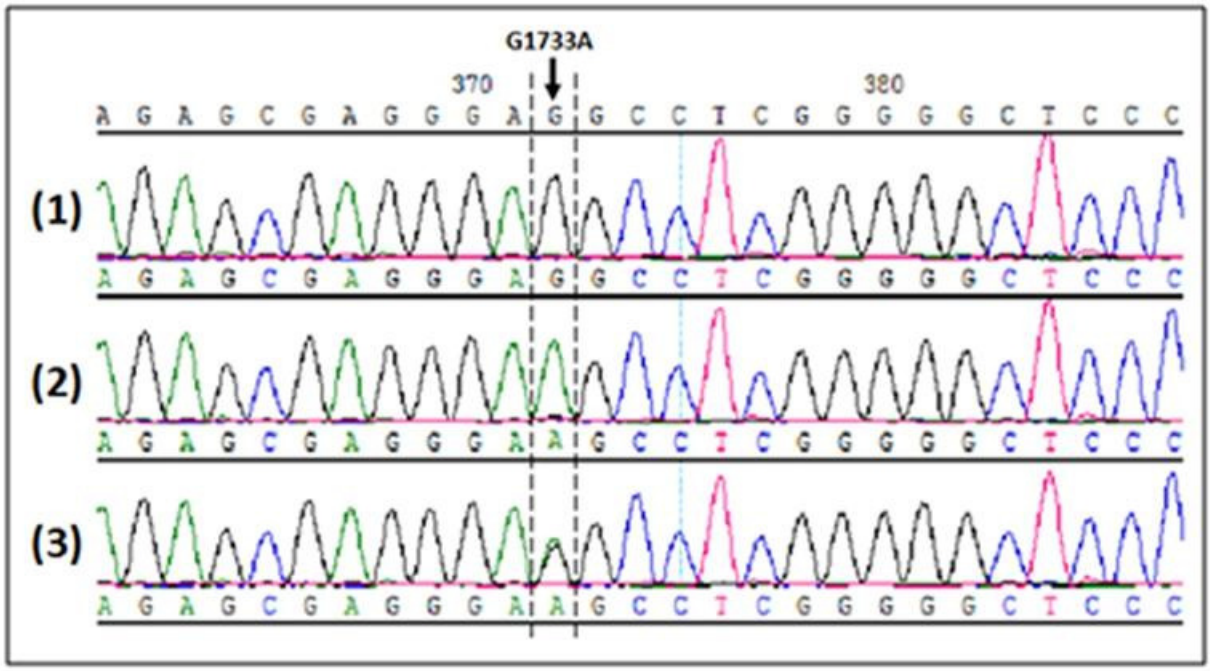

Figure 1. Sequence diagrams of G1733A locus in AR gene. (1) Represented G allele, (2) represented A allele (homozygous), (3) represented genotype of GA (heterozygote).

Table 3. Genotype and allele frequencies of G1729A polymorphism of AR gene among patients with unexplained recurrent spontaneous abortion and controls

Genotype/Allele	Patients (n= 65)	Controls (n= 65)	OR (95% CI)	*P* value
Genotype	*n (%)*	*n (%)*		
GG	64 (98.5)	61 (93.8)	4.20 (0.46-38.63)	0.36
GA + AA	1 + 0 (1.5)	4 + 0 (6.2)	0.23 (0.03-2.19)	0.36
Allele				
G	129 (99.2)	126 (96.9)	0.24 (0.03-2.22)	0.37[a]
A	1 (0.8)	4 (3.1)		

[a]Fisher exact test.

respectively, for the patient group and 73.8%, 20%, and 6.2%, respectively, for the control group. Allele frequencies of the G1733A polymorphism among patients and controls were 86.9% and 83.8%, respectively, for the most frequent allele (G) (wild type) and 13.1% and 16.2%, respectively, for the A allele (mutant). Statistical analysis of the genotype frequencies showed that there are no significant differences between the two groups. Furthermore, as shown in Table 2, the analysis of allele frequencies indicated insignificant differences (P = 0.60; odds ratio 0.78) between women with RSA and controls. The $\chi 2$ values of Hardy-Weinberg equilibrium test of G1733A locus for the patient group were 1.29 (P = 0.53), while $\chi 2$ values of the control group were 4.43 (P = 0.11). These results suggested that the 2 groups in our study were in Hardy-Weinberg equilibrium and were demographically representative.

In addition, we identified, unexpectedly, a rare variation (G1729A) upstream the G1733A locus. The genotype and allele frequencies of G1729A variation of AR gene among the same patients with RSA and same controls are shown in Table 3. 64 patients had the GG genotype, and the other patient had the GA genotype. 61 control subjects had the GG genotype, and the other 4 had the GA genotype. No mutant homozygous (AA) carriers were identified in either the patient or control group. Figure 2 is showing the sequence diagrams of G1729A locus in AR gene. The frequencies of the GG and (GA+AA) genotypes of the G1729A variation were 98.5% and 1.5%, respectively, for the patient group and 93.8% and 6.2%, respectively, for the control group. Allele frequencies of the G1729A variation among patients and controls were 99.2% and 96.9%, respectively, for the most frequent allele (G) (wild type) and 0.8% and 3.1%, respectively, for the A allele (mutant). Statistical analysis of the genotype frequencies showed that there are no

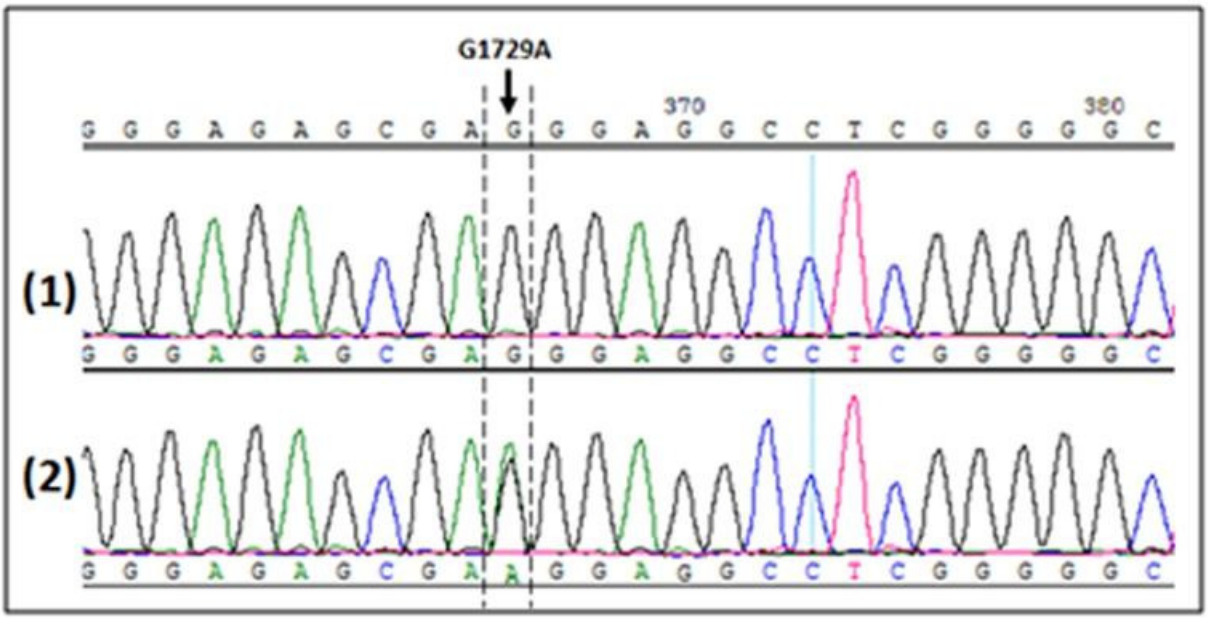

Figure 2. Sequence diagrams of G1729A locus in AR gene. (1) Represented G allele, (2) represented genotype of GA (heterozygote).

significant differences between the two groups. Furthermore, as shown in Table 3, the analysis of allele frequencies indicated insignificant differences (P = 0.37; odds ratio 0.24) between women with unexplained recurrent spontaneous abortion and controls.

DISCUSSION

The progression of RSA is complicated and may be controlled by different hereditary pathways. It was reported that RSA is associated with proteins, encoded from different genes, involved in distinct biologic pathways (Buchholz et al., 2004; Tempfer et al., 2001). A number of studies were performed to explore whether polymorphisms of distinctive genes could contribute to RSA development. Polymorphisms in the genes coding for endothelial nitric oxide synthase (15, 43. 44), cytokine genes (16. 45), vascular endothelial growth factor (16), factor V (17), the angiotensinogen gene (14), Glutathione S-transferase A1 gene (46), prothrombin mutation and methylenetetrahydrofolatereductase genes (Lino et al., 2014), and folic acid metabolism-related genes (48) have been studied for the risk of RSA. Notwithstanding, the outcomes from diverse studies are frequently contentious, prompting indeterminate decisions about the role of these polymorphisms in RSA pathogenesis.

Steroid hormones and their receptors are involved as initiators or promoters in diverse ailments.AR CAG repeat length have been reported in several studies to be associated with prostatecarcinogenesis (Medeiros et al., 2003; Montgomery et al., 2001; Kim et al., 2008), benign prostatic hyperplasia (Giovannucci et al., 1999; Roberts et al., 2004), and infertility among men (Ochsenkuhn and De Kretser, 2003; Davis-Dao et al., 2007; Giagulli et al., 2014), in addition to ovarian (Terry et al., 2005) and endometrial cancer risk (McGrath et al., 2006), polycystic ovary syndrome (Kim et al., 2008), autoimmunity in women with lupus (Olsen et al., 2014), and osteoporosis (Langdahl et al., 2003) in females. Several reported mechanisms pointed out the involvement of androgens in the development of breast cancer in females (Chen et al., 2014). One suggested mechanism included the conversions of androgens to E2 or their coupling to the estrogen receptor and/or AR (Elhaji et al., 2001; Giguere et al., 2001; Cox et al., 2006). Besides, it was shown by two laboratories the important role of AR in normal female reproduction (64-65). They provided the evidence that female $AR^{-/-}$ mice are subfertile. Accordingly, it is possible that polymorphisms that may affect AR function could be associated with RSA.

In the present study we attempted to investigate the potential effect of the G1733A polymorphism in the 211 codon of the AR gene in the occurrence of RSA. In addition, we identified, unexpectedly, a rare variation (G1729A) in the second nucleotide position of the 210 codon that had not been previously reported. G1729A polymorphism is a non-synonymous change as the arginine is changed to lysine. Beside two other reports (5, 18), This is the third report concerning the association of G1733A polymorphism to RSA. Previous data indicated that the allele frequencies of the G1733A polymorphism vary among different populations. In Mediterranean populations, the A allele frequency increases from 8% in

coastal Sardinians to 22%in Spanish Basques (Esteban et al., 2005), whereas it has been found to be13% in white North Americans (Lu and Danielsen, 1996), 2% in Brazilians (Ribeiro et al., 2002), and approximately 27% in North African populations (Esteban et al., 2005).

For the G1733A polymorphism, our present results indicated that the frequency of the A allele in the Saudi females is 16.2%. The frequency of homozygous AA is 6.2% and 0% in the control and patient groups, respectively, whereas the frequency of heterozygous GA is20% for the control group and 26.2% for the patient group. For the G1729A polymorphism, our present results indicated that the frequency of the A allele in the Saudi females is 3.1%. The frequency of GA+AA is 6.2% and 1.5% in the control and patient groups, respectively. Statistical analysis indicated that there are no significant differences between the two groups and the two polymorphisms investigated in terms of both genotype distribution and allele frequencies. The presence of the A allele of the G1733A and G1729A polymorphisms seems to be not associated with an increased risk for recurrent miscarriage. Our results for G1733A polymorphism contradicted the results found in the Greece (18) and Iranian (5) populations. Thus, further studies and larger sample size are required to consider AR as a genetic marker for the assessment of a woman's risk for RSA.

CONCLUSIONS

Based on the findings of the present study, it can be concluded that there is no association between G1733A Polymorphism of androgen receptor and recurrent spontaneous abortion (RSA) in Saudi women. In addition, there was no association between AR G1729A polymorphism and RSA in Saudi's women.

ACKNOWLEDGEMENTS

This research project was supported by a grant from the "Research Center of the Female Scientific and Medical Colleges", Deanship of Scientific Research, King Saud University, Riyadh, Kingdom of Saudi Arabia.

Conflict of Interests

The authors declare that they have no conflict of interests.

REFERENCES

Apparao KBC, Lovely LP, Gui Y, Lininger RA, Lessey BA (2002). Elevated endometrial androgen receptor expression in women with polycystic ovarian syndrome. BiolReprod Feb;66(2):297-304.

Bachmann J, Feldmer M, Ganten U, Stock G, Ganten D (1991). Sexual dimorphism of blood pressure: possible role of the renin-angiotensin system. J Steroid BiochemMolBiol;40(4-6):511-5.

Beaman KD, Ntrivalas E, Mallers TM, Jaiswal MK, KwakGÇÉKim J, GilmanGÇÉSachs A (2012). Immune etiology of recurrent pregnancy loss and its diagnosis. Ame. J. Reprod. Immunol. 67(4):319-25.

Brown CJ, Goss SJ, Lubahn DB, Joseph DR, Wilson EM, French FS, et al. (1989). Androgen receptor locus on the human X chromosome: regional localization to Xq11-12 and description of a DNA polymorphism. Am J Hum Genet Feb;44(2):264-9.

Buchholz T, Lohse P, Kosian E, Thaler CJ (2004). Vasoconstrictively acting AT1R A1166C and NOS3 4/5 polymorphisms in recurrent spontaneous abortions (RSA). Am J ReprodImmunol May;51(5):323-8.

Burger HG (2002). Androgen production in women. Fertility and sterility;77:3-5.

Cao Y, Zhang Z, Zheng Y, Yuan W, Wang J, Liang H, et al. (2014). The association of idiopathic recurrent early pregnancy loss with polymorphisms in folic acid metabolism-related genes. Genes Nutr;9(3):1-8.

Carlsen SM, Romundstad P+, Jacobsen G (2005). Early secondGÇÉtrimester maternal hyperandrogenemia and subsequent preeclampsia: a prospective study. Actaobstetricia et gynecologicaScandinavica;84(2):117-21.

Chang C, Saltzman A, Yeh S, Young W, Keller E, Lee HJ, et al. (1995). Androgen receptor: an overview. Critical ReviewsGäó in Eukaryotic Gene Expression;5(2).

Chen HT, Wu YC, Chen ST, Tsai HC, Chien YC (2014). Androgen receptor CAG repeats, non-random X chromosome inactivation, and loss of heterozygosity at Xq25 in relation to breast cancer risk. BMC cancer;14(1):144.

Cox DG, Blanche H, Pearce CL, Calle EE, Colditz GA, Pike MC, et al. (2006). A comprehensive analysis of the androgen receptor gene and risk of breast cancer: results from the National Cancer Institute Breast and Prostate Cancer Cohort Consortium (BPC3). Breast Cancer Res;8(5).

Davis-Dao CA, Tuazon ED, Sokol RZ, Cortessis VK (2007). Male infertility and variation in CAG repeat length in the androgen receptor gene: a meta-analysis. J ClinEndocrinolMetab Nov;92(11):4319-26.

Eisermann K, Wang D, Jing Y, Pascal LE, Wang Z (2013). Androgen receptor gene mutation, rearrangement, polymorphism. Translational Andrology and Urology;2(3):137-47.

Elhaji YA, Gottlieb B, Lumbroso R, Beitel LK, Foulkes WD, Pinsky L, et al. (2001). The polymorphic CAG repeat of the androgen receptor gene: a potential role in breast cancer in women over 40. Breast Cancer Res Treat Nov;70(2):109-16.

Ellis JA, Stebbing M, Harrap SB (2001). Polymorphism of the androgen receptor gene is associated with male pattern baldness. Journal of investigative dermatology;116(3):452-5.

Esteban E, Via M, Gonzalez-Perez E, Santamaria J, Dugoujon JM, Vona G, et al. (2005). An unexpected wide population variation of the G1733A polymorphism of the androgen receptor gene: data on the Mediterranean region. Am J Hum Biol;17(6):690-5.

Gelmann EP (2002). Molecular biology of the androgen receptor. Journal of Clinical Oncology;20(13):3001-15.

Giagulli VA, Carbone MD, De Pergola G, Guastamacchia E, Resta F, Licchelli B, et al. (2014). Could androgen receptor gene CAG tract polymorphism affect spermatogenesis in men with idiopathic infertility? J. Assisted Reprod. Genetics;1-9.

Giguere Y, Dewailly E, Brisson J, Ayotte P, Laflamme N, Demers A, et al. (2001). Short polyglutamine tracts in the androgen receptor are protective against breast cancer in the general population. Cancer Res Aug 1;61(15):5869-74.

Giovannucci E, Stampfer MJ, Chan A, Krithivas K, Gann PH, Hennekens CH, et al. (1999). CAG repeat within the androgen receptor gene and incidence of surgery for benign prostatic hyperplasia in U.S. physicians. Prostate May;39(2):130-4.

Gottlieb B, Beitel LK, Nadarajah A, Paliouras M, Trifiro M (2012). The androgen receptor gene mutations database: 2012 update. Human mutation;33(5):887-94.

Grad JM, Lyons LS, Robins DM, Burnstein KL (2001). The androgen receptor (AR) amino-terminus imposes androgen-specific regulation of AR gene expression via an exonic enhancer. Endocrinology Mar;142(3):1107-16.

Guay A, Munarriz R, Jacobson J, Talakoub L, Traish A, Quirk F, et al. (2004). Serum androgen levels in healthy premenopausal women with and without sexual dysfunction: Part A. Serum androgen levels in women aged 20GÇô49 years with no complaints of sexual dysfunction. International journal of impotence research;16(2):112-20.

Gustafson DR, Wen MJ, Koppanati BM (2003). Androgen receptor gene repeats and indices of obesity in older adults. Int J ObesRelatMetabDisord Jan;27(1):75-81.

Hefler LA, Tempfer CB, Bashford MT, Unfried G, Zeillinger R, Schneeberger C, et al. (2002). Polymorphisms of the angiotensinogen gene, the endothelial nitric oxide synthase gene, and the interleukin-1beta gene promoter in women with idiopathic recurrent miscarriage. Mol Hum Reprod Jan;8(1):95-9100.

Jahaninejad T, Ghasemi N, Kalantar SM, Sheikhha MH, Pashaiefar H (2013). StuI polymorphism on the androgen receptor gene is associated with recurrent spontaneous abortion. J Assist Reprod Genet Mar;30(3):437-40.

Karvela M, Papadopoulou S, Tsaliki E, Konstantakou E, Hatzaki A, Florentin-Arar L, et al. (2008). Endothelial nitric oxide synthase gene polymorphisms in recurrent spontaneous abortions. Arch GynecolObstet Oct;278(4):349-52.

Karvela M, Stefanakis N, Papadopoulou S, Tsitilou SG, Tsilivakos V, Lamnissou K (2010). Evidence for association of the G1733A polymorphism of the androgen receptor gene with recurrent spontaneous abortions. FertilSteril 2008 Nov;90(5):.e9-e12.

Kim HS, Lee BE, Jeon YJ, Rah H, Lee WS, Shin JE, et al. (2014). Transcobalamin II (TCN2 67A> G and TCN2 776C> G) and Transcobalamin II Receptor (TCblR 1104C> T) Polymorphisms in Korean Patients with Idiopathic Recurrent Spontaneous Abortion. American Journal of Reproductive Immunology.

Kim JJ, Choung SH, Choi YM, Yoon SH, Kim SH, Moon SY (2008). Androgen receptor gene CAG repeat polymorphism in women with polycystic ovary syndrome. FertilSteril Dec;90(6):2318-23.

Kucerova R, Bienova M, Kral M, Bouchal J, Trtkova KS, Burdova A, et al. (2014). Androgenetic alopecia and polymorphism of the androgen receptor gene (SNP rs6152) in patients with benign prostate hyperplasia or prostate cancer. J EurAcadDermatolVenereol Mar 1;n/a.

Langdahl BL, Stenkjaer L, Carstens M, Tofteng CL, Eriksen EF (2003). A CAG repeat polymorphism in the androgen receptor gene is associated with reduced bone mass and increased risk of osteoporotic fractures. Calcif Tissue Int Sep;73(3):237-43.

Li TC, Makris M, Tomsu M, Tuckerman E, Laird S (2002). Recurrent miscarriage: aetiology, management and prognosis. Hum Reprod Update;8(5):463-81.

Lino FL, Traina +, Barreto JA, Moron AF, Mattar R (2014). Thrombophilic Mutations and Polymorphisms, Alone or in Combination, and Recurrent Spontaneous Abortion. Clinical and Applied Thrombosis/Hemostasis Jan 24.

Lu J, Danielsen M (1996). A Stu I polymorphism in the human androgen receptor gene (AR).Clin Genet Jun;49(6):323-4.

Mantzoros CS, Georgiadis EI, Young R, Evagelopoulou C, Khoury S, Katsilambros N, et al. (1995). Relative androgenicity, blood pressure levels, and cardiovascular risk factors in young healthy women. American journal of hypertension;8(6):606-14.

McGrath M, Lee IM, Hankinson SE, Kraft P, Hunter DJ, Buring J, et al. (2006). Androgen receptor polymorphisms and endometrial cancer risk. Int J Cancer Mar 1;118(5):1261-8.

Medeiros R, Vasconcelos A, Costa S, Pinto D, Morais A, Oliveira J, et al. (2003). Steroid hormone genotypes ARStuI and ER325 are linked to the progression of human prostate cancer. Cancer Genet Cytogenet Mar;141(2):91-6.

Montgomery JS, Price DK, Figg WD (2001). The androgen receptor gene and its influence on the development and progression of prostate cancer. J Pathol Sep;195(2):138-46.

Ochsenkuhn R, De Kretser DM (2003). The contributions of deficient androgen action in spermatogenic disorders.Int J Androl Aug;26(4):195-201.

Olsen NJ, Benko AL, Kovacs WJ (2014). Variation in the androgen receptor gene exon 1 CAG repeat correlates with manifestations of autoimmunity in women with lupus. Endocrine connections;3(2):99-109.

Pandey MK, Rani R, Agrawal S (2005). An update in recurrent spontaneous abortion. Archives of gynecology and obstetrics;272(2):95-108.

Papazoglou D, Galazios G, Papatheodorou K, Liberis V, Papanas N, Maltezos E, et al (2005). Vascular endothelial growth factor gene polymorphisms and idiopathic recurrent pregnancy loss. FertilSteril Apr;83(4):959-63.

Quigley CA, De Bellis A, Marschke KB, el-Awady MK, Wilson EM, French FS (1995). Androgen receptor defects: historical, clinical, and molecular perspectives. Endocr Rev Jun;16(3):271-321.

Rai R, Regan L (2006). Recurrent miscarriage. The Lancet;368(9535):601-11.

Redline RW (2006). Thrombophilia and placental pathology.ClinObstetGynecol Dec;49(4):885-94.

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Regan L (1998). Overview of recurrent miscarriage. Gynaecology Forum;3:3-7.

Regan L, Backos M, Rai R (2010). The Investigation and treatment of couples with Recurrent First-trimester and Second-trimester Miscarriage. RCOG Green-top Guideline No. 17 [http://www.rcog.org.uk/files/rcog-corp/GTG17recurrentmiscarriage.pdf].

Ribeiro ML, Santos A, Carvalho-Salles AB, Hackel C (2002). Allelic frequencies of six polymorphic markers for risk of prostate cancer. Braz J Med Biol Res Feb;35(2):205-13.

Roberts RO, Jacobson DJ, Rhodes T, Klee GG, Leiber MM, Jacobsen SJ (2004). Serum sex hormones and measures of benign prostatic hyperplasia. Prostate Oct 1;61(2):124-31.

Roy AK, Lavrovsky Y, Song CS, Chen S, Jung MH, Velu NK, et al. (1999). Regulation of androgen action. Vitamins and hormones;55:309-52.

Saarela T, Jaaskelainen J, Tenhola S, Voutilainen R, Heinonen S (2005). Preeclampsia and androgen receptor gene CAG repeat length: results from both children and women. J Assist Reprod Genet Aug;22(7-8):269-75.

Sereshki N, Gharagozloo M, Ostadi V, Ghahiri A, Roghaei MA, Mehrabian F, et al. (2014). Variations in T-helper 17 and regulatory T cells during the menstrual cycle in peripheral blood of women with recurrent spontaneous abortion. Int. J. fertility & sterility 8(1):59.

Shiina H, Matsumoto T, Sato T, Igarashi K, Miyamoto J, Takemasa S, et al. (2006). Premature ovarian failure in androgen receptor-deficient mice. ProcNatlAcadSci U S A Jan 3;103(1):224-9.

Shin SJ, Lee HH, Cha SH, Kim JH, Shim SH, Choi DH, et al., (2010). Endothelial nitric oxide synthase gene polymorphisms (GêÆ 786T> C, 4a4b, 894G> T) and haplotypes in Korean patients with recurrent spontaneous abortion. European Journal of Obstetrics & Gynecology and Reproductive Biology;152(1):64-7.

Suzumori N, Sugiura-Ogasawara M (2010). Genetic factors as a cause of miscarriage. Current medicinal chemistry;17(29):3431-7.

Tempfer C, Unfried G, Zeillinger R, Hefler L, Nagele F, Huber JC (2001). Endothelial nitric oxide synthase gene polymorphism in women with idiopathic recurrent miscarriage. Hum Reprod Aug;16(8):1644-7.

Terry KL, De Vivo I, Titus-Ernstoff L, Shih MC, Cramer DW (2005). Androgen receptor cytosine, adenine, guanine repeats, and haplotypes in relation to ovarian cancer risk. Cancer Res Jul 1;65(13):5974-81.

Trapman J, Klaassen P, Kuiper GGJM, Van der Korput JAGM, Faber PW, Van Rooij HCJ, et al. (1988). Cloning, structure and expression of a cDNA encoding the human androgen receptor. Biochemical and biophysical research communications;153(1):241-8.

Unfried G, Tempfer C, Schneeberger C, Widmar B, Nagele F, Huber JC (2001). Interleukin 1 receptor antagonist polymorphism in women with idiopathic recurrent miscarriage.FertilSteril Apr;75(4):683-7.

Walters KA, Allan CM, Jimenez M, Lim PR, Davey RA, Zajac JD, et al. (2007). Female mice haploinsufficient for an inactivated androgen receptor (AR) exhibit age-dependent defects that resemble the AR null phenotype of dysfunctional late follicle development, ovulation, and fertility. Endocrinology Aug;148(8):3674-84.

Wilczynski JR, Radwan P, Tch+¦rzewski H, Banasik M (2012). Immunotherapy of patients with recurrent spontaneous miscarriage and idiopathic infertility: does the immunization-dependent Th2 cytokine overbalance really matter? Archivumimmunologiae et therapiaeexperimentalis;60(2):151-60.

Xiao S, Lu X, Li X, Zhang L, Bao S, Zhao A (2014). Study on the pathogenesis of autoimmune-type recurrent spontaneous abortion by establishing a new mouse model. European Journal of Obstetrics & Gynecology and Reproductive Biology;178:84-8.

Yong EL, Ghadessy F, Wang Q, Mifsud A, Ng SC (1998). Androgen receptor transactivation domain and control of spermatogenesis. Reviews of reproduction; 3(3):141-4.

Zammiti W, Mtiraoui N, Mercier E, Abboud N, Saidi S, Mahjoub T, et al. (2006). Association of factor V gene polymorphisms (Leiden; Cambridge; Hong Kong and HR2 haplotype) with recurrent idiopathic pregnancy loss in Tunisia. A case-control study.ThrombHaemost Apr;95(4):612-7.

Zong C, Sha Y, Xiang H, Wang J, Chen D, Liu J, et al. (2014). Glutathione S-transferase A1 polymorphism and the risk of recurrent spontaneous abortion in Chinese Han population. J Assist Reprod Genet;31(3):379-82.