Research Article

ESR1 rs9340799 Is Associated with Endometriosis-Related Infertility and In Vitro Fertilization Failure

Diego Davila Paskulin, 1,2 João Sabino Cunha-Filho, 3 Livia Davila Paskulin, 2 Carlos Augusto Bastos Souza, 3 and Patricia Ashton-Prolla 1,2,4

1 Department of Genetics and Molecular Biology, Federal University of Rio Grande do Sul, Avenida Bento Gonçalves 9500, 43323 M, 91501-970 Porto Alegre, RS, Brazil
2 Genomic Medicine Laboratory, Experimental Research Center, Hospital de Clinicas de Porto Alegre, Avenida Ramiro Barcelos 2350, 90035-903 Porto Alegre, RS, Brazil
3 Department of Obstetrics and Gynecology, Federal University of Rio Grande do Sul, Avenida Ramiro Barcelos 2350, 90035-903 Porto Alegre, RS, Brazil
4 Medical Genetics Service and National Institute of Science and Technology in Population Medical Genetics (INAGEMP), Hospital de Clinicas de Porto Alegre, Avenida Ramiro Barcelos 2350, 90035-903 Porto Alegre, RS, Brazil

Correspondence should be addressed to Diego Davila Paskulin; diegopaskulin@gmail.com

Received 30 September 2013; Revised 30 November 2013; Accepted 5 December 2013

Academic Editor: Irene Rebelo

Copyright © 2013 Diego Davila Paskulin et al. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Estrogen receptor alpha has a central role in human fertility by regulating estrogen action in all human reproductive tissues. Leukemia inhibitory factor (LIF) expression, a cytokine critical for blastocyst implantation, is mediated by estrogen signaling, so we hypothesized that ESR1 gene polymorphisms might be candidate risk markers for endometriosis-related infertility and in vitro fertilization (IVF) failure. We included 98 infertile women with endometriosis, 115 infertile women with at least one IVF failure and also 134 fertile women as controls. TaqMan SNP assays were used for genotyping LIF (rs929271), MDM2 (rs2279744), MDM4 (rs1563828), USP7 (rs1529916), and ESR1 (rs9340799 and rs2234693) polymorphisms. The SNP ESR1 rs9340799 was associated with endometriosis-related infertility \( (P < 0.001) \) and also with IVF failure \( (P = 0.018) \). After controlling for age, infertile women with ESR1 rs9340799 GG genotype presented 4-fold increased risk of endometriosis \( (OR = 4.67, 95\% CI 1.84–11.83, P = 0.001) \) and 3-fold increased risk of IVF failure \( (OR = 3.33, 95\% CI 1.38–8.03, P = 0.007) \). Our results demonstrate an association between ESR1 rs9340799 polymorphism and infertile women with endometriosis and also with women who were submitted to IVF procedures and had no blastocyst implantation.

1. Introduction

Endometriosis is a benign gynecological estrogen-dependent inflammatory condition defined by the presence of endometrial-like tissue in extraterine locations [1]. Endometriosis affects up to 10% of women of reproductive age and is responsible for infertility and pelvic pain [2]. Due to its complexity, endometriosis is usually referred to as exhibiting a polygenic and multifactorial basis [3]. Estrogen plays a significant role in the pathogenesis of the disease by promoting endometriotic tissue cell survival, maintenance, and differentiation [2, 3]. Estrogen activates a wide array of tissue- and organ-specific physiological responses by binding to its receptor ESR1, mostly located at the thecal layer, and modulating uterine events preparing the endometrium for embryo attachment and implantation [4].

Though many studies suggest that genetic polymorphisms of estrogen receptor α gene (ESR1) modify susceptibility to women’s disorders including osteoporosis, preeclampsia, and breast cancer, limited studies have demonstrated associations of ESR1 polymorphisms in women with endometriosis-related infertility [5–7]. Previous reports have shown associations of ESR1 genetic variants with susceptibility to endometriosis and fertility status [6, 8–13], but many...
studies failed to achieve an association regarding ESR1 variants and endometriosis-related infertility [9, 14–16]. Interestingly, Lamp et al. linked ESR1 SNPs only to endometriosis without infertility [12], while Wang et al. associated ESR1 rs3798573 with risk of both endometriosis and infertile endometriosis in Han Chinese women [13]. ESR1 rs2234693 (PvuII) polymorphism was significantly more prevalent in infertile women at premature ovarian aging [17] and was predictive of an improved controlled ovarian stimulation [18]. Both rs9340799 (XbaI) and ESR1 rs2234693 (PvuII) polymorphisms are associated with differences in the response to ovarian stimulation bestowing an indirect role that might affect implantation rates [19].

In a recent investigation, gene-array analysis revealed more than 300 genes downregulated in patients with repeated in vitro fertilization (IVF) failure, with at least 8% of them being estrogen dependent [20]. Numerous factors as folliculogenesis, endometrial receptivity, and oocyte maturation have been associated with failure of in vitro fertilization (IVF) failure, but the lack of estrogen responsiveness might be a great challenge in these situations [20]. The embryonic implantation process requires a receptive endometrium and both estrogen and TP53 present essential roles during implantation through the regulation of leukemia inhibitory factor (LIF), a polyfunctional glycoprotein cytokine critical for blastocyst implantation [21]. LIF expression is continuous in the uterus; however, it shows a transient expression peak during pregnancy and this peak coincides with the onset of implantation at the 12th day after fertilization in humans [22]. LIF has been described as an important gene in differentiation, proliferation, and cell survival pathways [23] and its expression is reduced in endometrium from women with unexplained infertility [24].

To our knowledge, no study has focused on ESR1 polymorphisms and infertile women who were submitted to conventional in vitro fertilization (IVF) procedures with unsuccessful blastocyst implantations. Meanwhile estrogen functions are so important to blastocyst implantation and to the pathogenesis of endometriosis; ESR1 gene variants might be one of the causative factors for these conditions in infertile women. We then hypothesized that genetic variants in ESR1, MDM2, MDM4, USP7, and LIF genes may differ between fertile women and two groups of infertile women: first, women with endometriosis-related infertility and second, women with failure of in vitro fertilization procedures.

2. Material and Methods

2.1. Subjects. Patients and subjects were invited to participate and signed a consent form at inclusion. The research project was approved by the Hospital de Clínicas de Porto Alegre (HCPA) Ethics Committee (GPPG 05-182; GPPG 09-430). Infertile patients with and without endometriosis and controls were divided into three study groups as previously described [25]. Infertility was defined as the inability of a couple to achieve pregnancy after 1 year of regular unprotected sexual intercourse [26]. The IVF Failure Group consisted of 115 infertile women with at least one IVF failure, submitted to conventional IVF with 35 years or less. Patients with endometriosis, previous thyroid disease, positive antilupus or anticardiolipin antibodies, and thrombophilias were excluded from our sample. Controlled ovarian hyperstimulation was performed with the use of recombinant human FSH and pituitary suppression with GnRH antagonist (fixed day-6 protocol). Ovulation was induced by 6500 IU recombinant hCG when at least three follicles had reached a diameter of 17 mm, and transvaginal follicle aspiration was performed 36 hours later under ultrasound guidance. Embryos were classified according to the cumulative embryo classification, taking into account cleavage speed, blastomere symmetry, extent of fragmentation, and the presence or absence of multinucleated blastomeres. The Endometriosis Group comprised 98 infertile women with minimal or mild endometriosis as diagnosed by laparoscopy according to the classification proposed by the American Society for Reproductive Medicine recruited at the Gynecology Service of HCPA, in Southern Brazil [26]. Other causes of infertility were excluded by hysterosalpingography, sperm evaluation, and hormonal measurements whenever necessary. The Fertile Group consisted of 134 women with no history of infertility, who already had two or more children without any difficulties or assisted reproduction and underwent laparoscopy for tubal ligation at HCPA.

2.2. Genotyping. Genomic DNA was extracted from peripheral blood leukocytes using the Illustra blood genomic Prep Mini Spin Kit (GE Healthcare, Piscataway, NJ, USA) as described by the manufacturer. DNA concentration was measured with Nano-Drop 1000 (Thermo Scientific, Wilmington, USA) and diluted to a final concentration of 10 ng/μL.

TaqMan allelic discrimination analyses were performed according to Applied Biosystems standard protocols (Applied Biosystems, Carlsbad, USA). The analyzed SNPs were as follows: MDM4 rs1563828 (C>G, rs1563828), USP7 rs1529916 (C>T, rs1529916), LIF rs9292721 (C>T, rs9292721), ESR1 rs9340799 (C>T, rs9340799), ESR1 rs2234693 (G>A, rs2234693), and MDM2 rs2279744 for which a custom-made TaqMan assay was made, using forward primer 5'-CGGGAGTCTAGGTAAGGT-3', reverse primer 5'-CAAGGCACCTCGATCATC-3', VIC probe 5'-CTCCCGCGCGCAAG-3' and FAM probe 5'-TCCCGCGCCGAG-3' (Applied Biosystems). PCR cycling reactions were performed on an ABI StepOne System (Applied Biosystems) and consisted of initial denaturation at 95°C for 15 min, 40 cycles with denaturation 95°C for 15 s, and then annealing and extension at 60°C for 1 min.

2.3. Statistical Analysis. Clinical features of women in all study groups were compared by t-test. Differences in genotype distribution were assessed by chi-square analysis, which was also used to test for Hardy-Weinberg equilibrium. Logistic regression analysis was carried out to estimate the odds ratios with 95% confidence intervals (CIs) in order to assess the influence of ESR1 rs9340799 genotypes on endometriosis-related infertility and IVF failure. Statistical analyses were performed using the SPSS 20.0 statistical package. All reported P values are two-tailed and were considered statistically significant when equal to 0.05 or less.
3. Results

The clinical and demographic characteristics of the women enrolled in the study are shown in Table 1. Mean age at recruitment was higher in the Fertile Group (42.6 ± 12.88 years) than in both the Endometriosis (32.87 ± 4.7 years) and IVF Failure (31.65 ± 3.24 years) groups since only women of 35 years or less were included in these two latter groups. The population-based fertile control women presented a mean of 3.62 ± 1.94 pregnancies reflecting the average number of pregnancies in the normal population from Southern Brazil. Both Endometriosis and IVF Failure groups presented low frequencies of pregnancy, abortion, and caesarean due to their infertility status. Patients and healthy study subjects did not differ significantly regarding self-attributed skin color as a self-denomination of “white” color predominated in all study groups as previously described in [25].

Hardy-Weinberg equilibrium was achieved for all SNPs in the three study groups (data not shown). Table 2 presents genotype frequencies of the SNPs included in the study. No association was found between LIF, MDM2, MDM4, and USP7 SNPs and endometriosis-related infertility or in vitro fertilization failure. However, a strong association was found between the ESR1 rs9340799 polymorphism and clinical phenotype in both case groups (Endometriosis, P < 0.001 and IVF Failure, P = 0.018) when compared with the Fertile Group. Interestingly, no association was found between ESR1 rs2234693 and the outcomes.

To evaluate the effects of the ESR1 rs9340799 polymorphism, we carried out a logistic regression analysis, controlled by age, with endometriosis-related infertility and IVF failure as outcomes. Results are summarized in Table 3 and show a statistically significant effect of AG (OR 2.67, 95% CI 1.49–4.78, P = 0.001) and GG (OR 4.67, 95% CI 1.84–11.83, P = 0.001) genotypes with endometriosis-related infertility. Regarding the IVF Failure Group, genotype GG contributed significantly to the outcome as women with genotype GG had 3-fold-increased risk of IVF failure (OR 3.33, 95% CI 1.38–8.03, P = 0.007).

4. Discussion

In the present study, we have analyzed common SNPs in ESR1, MDM2, MDM4, USP7, and LIF genes in infertile women with endometriosis or failure of in vitro fertilization procedures. Our results demonstrate an association between ESR1 rs9340799 polymorphism with infertile women with endometriosis and also with women who were submitted to IVF procedures and had no embryo implantation.

TP53 regulates maternal reproduction through the expression of LIF [27]. At 12 days of pregnancy, LIF is expressed at high levels making the uterus receptive to the blastocyst [27]. Both TP53 and estrogen are essential for LIF expression in the endometrial glands, and impaired function of these proteins are clearly associated with failure of blastocyst implantation [27]. Different studies have demonstrated that SNPs modulate the activity of TP53, and also in its regulators MDM2, MDM4, and USP7 are more frequent in IVF patients [25, 28]. We have previously shown that TP53 polymorphisms are associated with both endometriosis-related infertility and IVF failure in patients from Southern Brazil [25]. Using the same cohort, we expanded the analysis to other TP53 signaling network genes [29], and in contrast with previous findings, our results demonstrated no association of MDM2, MDM4, USP7, and LIF polymorphisms with endometriosis-related infertility or IVF failure patients.

LIF is regulated by both TP53 and estrogen. Estrogen signaling is mediated through its nuclear receptor alpha. Studies have demonstrated an association between ESR1 polymorphisms and endometriotic women with and without infertility [13, 17], but to our knowledge, no study has evaluated ESR1 polymorphisms in IVF failure. Our results demonstrate an association between ESR1 rs9340799 polymorphism (also known as ER-α Xbal) and endometriosis-related infertility. In regard to the association found here, a previous meta-analysis performed to derive a more precise association between the ESR1 polymorphisms and risk of endometriosis found no obvious associations [30]. However, it is important to note that even though the authors indicate that ethnicity (Caucasian or Asian), country (Japan, China, Korea, Germany, and Italy), and sample size could not explain heterogeneity across the fifteen studies included in the meta-analyses, only two studies included Caucasian populations (totalizing only 111 cases and 146 controls from a total of 1349 cases and 1411 controls). In addition, there was no uniformity in the classifications regarding “endometriosis” among the different studies. To minimize bias towards endometriosis classification, we only included in the present study infertile women with minimal or mild endometriosis as diagnosed by laparoscopy according to the classification proposed by the American Society for Reproductive Medicine [26]. The classification of endometriosis is changing from a local disorder to a complex disease as new molecular mechanisms are being...
Table 2: Genotype and allele frequencies of TP53 signaling pathway gene polymorphisms.

| Gene   | SNP      | Fertile n (%) | Endometriosis n (%) | P value* | IVF Failure n (%) | P value** |
|--------|----------|---------------|---------------------|----------|-------------------|-----------|
| MDM2   | rs2279744|               |                     |          |                   |           |
| TT     |          | 57 (42.5)     | 41 (41.8)           | 0.824    | 48 (41.7)         | 0.918     |
| TG     |          | 64 (47.8)     | 45 (45.9)           |          | 54 (47)           |           |
| GG     |          | 13 (9.7)      | 12 (12.2)           |          | 13 (11.3)         |           |
| G      |          | 0.67          | 0.65                | 0.765    | 0.65              |           |
| T      |          | 0.33          | 0.35                |          | 0.35              | 0.765     |
| MDM4   | rs1563824|               |                     |          |                   |           |
| CC     |          | 34 (25.4)     | 34 (34.7)           | 0.268    | 40 (34.8)         | 0.141     |
| CT     |          | 71 (53)       | 43 (42.9)           |          | 59 (51.3)         |           |
| TT     |          | 29 (21.6)     | 21 (21.4)           |          | 16 (13.9)         |           |
| C      |          | 0.52          | 0.57                | 0.477    | 0.6               | 0.254     |
| T      |          | 0.48          | 0.43                |          | 0.4               |           |
| HAUSP  | rs1529916|               |                     |          |                   |           |
| CC     |          | 73 (54.5)     | 53 (54.1)           | 0.977    | 53 (46.1)         | 0.224     |
| CT     |          | 52 (38.8)     | 39 (39.8)           |          | 48 (41.7)         |           |
| TT     |          | 9 (6.7)       | 6 (6.1)             |          | 14 (12.2)         |           |
| C      |          | 0.74          | 0.74                | 1        | 0.67              | 0.277     |
| T      |          | 0.26          | 0.26                |          | 0.33              |           |
| LIF    | rs929271 |               |                     |          |                   |           |
| TT     |          | 57 (42.5)     | 47 (48)             | 0.702    | 46 (40)           | 0.784     |
| TG     |          | 60 (44.8)     | 39 (39.8)           |          | 51 (44.3)         |           |
| GG     |          | 17 (12.7)     | 12 (12.2)           |          | 18 (15.7)         |           |
| G      |          | 0.65          | 0.68                | 0.653    | 0.62              | 0.659     |
| T      |          | 0.35          | 0.32                |          | 0.38              |           |
| ESRI   | rs9340799|               |                     |          |                   |           |
| AA     |          | 71 (53)       | 27 (27.6)           | <0.001   | 45 (39.1)         | 0.018     |
| AG     |          | 54 (40.3)     | 55 (56.1)           |          | 51 (44.3)         |           |
| GG     |          | 9 (6.7)       | 16 (16.3)           |          | 19 (16.5)         |           |
| A      |          | 0.73          | 0.55                | 0.008    | 0.61              | 0.071     |
| G      |          | 0.27          | 0.45                |          | 0.39              |           |
| ESRI   | rs2234693|               |                     |          |                   |           |
| CC     |          | 27 (20.1)     | 18 (18.4)           | 0.861    | 17 (14.8)         | 0.105     |
| CT     |          | 69 (51.5)     | 54 (55.1)           |          | 51 (44.3)         |           |
| TT     |          | 38 (28.4)     | 26 (26.5)           |          | 47 (40.9)         |           |
| C      |          | 0.46          | 0.46                | 1        | 0.37              | 0.196     |
| T      |          | 0.54          | 0.54                |          | 0.63              |           |

* Chi-square analysis for the difference between Fertile and Endometriosis groups.  ** Chi-square analysis for the difference between Fertile and IVF Failure groups.

The endometriotic process is classified as an estrogen-dependent inflammatory disease similar to cancer due to its capability to invade surrounding tissues, to promote angiogenesis, inflammation, and apoptosis in favor of the new endometriotic tissue survival [31–36]. Estrogen production plays a central role in the pathology of endometriosis enhancing the survival of the endometriotic tissue, and together with prostaglandins and cytokines, mediating pelvic pain and infertility [37, 38]. The fact that estrogen inhibitors such as GnRh analogues, oral, and aromatase inhibitors are used to
Failure of IVF was observed among the study population [39]. Although both SNPs are present in intron 1 and do not lead to any amino acid change, it is plausible that they may directly influence ESR1 gene expression or alternatively could be linked to some unidentified causative DNA sequence variants. Introns can significantly affect gene expression in a variety of ways, as they may contain enhancer elements or cis- and trans-regulatory elements that may lead to alternative splicing, as well as various cis- and trans-regulatory elements that may lead to different proteins isoforms [44–46].

Although, to our knowledge, this is the first study to report an association between ESR1 genetic variants and failure of in vitro fertilization, our study had limitations. First, only two common ESR1 polymorphisms were investigated, so haplotype analysis was not performed. Second, examination of endometrial tissue to evaluate the effect of the analyzed SNPs regarding TP53 and LIF expression was not performed. Endometrial samples are being collected at this time, so analyses of protein response at the implantation stage are underway. Lastly, it is known that allele frequencies are greatly affected by racial and ethnic backgrounds. Although ancestral informative markers were not used to infer individual ancestry, we used self-reported skin color as a control for ethnic background, and no significant difference in the distribution of self-denominated skin color was observed among the study groups as the majority of individuals self-denominated them as "white."
a Chinese population," *Fertility and Sterility*, vol. 92, no. 1, pp. 54–60, 2009.

[9] Y. Matsuzaka, Y. Y. Kikuti, S. I. Izumi et al., "Failure to detect significant association between estrogen receptor-alpha gene polymorphisms and endometriosis in Japanese women," *Environmental Health and Preventive Medicine*, vol. 17, no. 5, pp. 423–428, 2011.

[10] I. Georgiou, M. Syrrou, I. Bouba et al., "Association of estrogen receptor gene polymorphisms with endometriosis," *Fertility and Sterility*, vol. 72, no. 1, pp. 164–166, 1999.

[11] J. Kitawaki, H. Obayashi, H. Ishihara et al., "Oestrogen receptor-\(a\) gene polymorphism is associated with endometriosis, adenomyosis and leiomyomata," *Human Reproduction*, vol. 16, no. 1, pp. 51–55, 2001.

[12] M. Lamp, M. Peters, E. Reinmaa et al., "Polymorphisms in ESR1, ESR2 and HSD17B1 genes are associated with fertility status in endometriosis," *Gynecological Endocrinology*, vol. 27, no. 6, pp. 425–433, 2011.

[13] W. Wang, Y. Li, M. Mattituohet al., "Association of an oestrogen receptor gene polymorphism in Chinese Han women with endometriosis and endometriosis-related infertility," *Reproductive BioMedicine Online*, vol. 26, no. 1, pp. 93–98, 2013.

[14] Z. Wang, S. Yoshida, K. Negoro, S. Kennedy, D. Barlow, and T. Maruo, "Polymorphisms in the estrogen receptor \(\beta\) gene but not estrogen receptor \(\alpha\) gene affect the risk of developing endometriosis in a Japanese population," *Fertility and Sterility*, vol. 81, no. 6, pp. 1650–1656, 2004.

[15] B. Trabert, S. M. Schwartz, U. Peters et al., "Genetic variation in the sex hormone metabolic pathway and endometriosis risk: an evaluation of candidate genes," *Fertility and Sterility*, vol. 96, no. 6, article e3, pp. 1401–1406, 2011.

[16] S. P. Renner, R. Strick, P. Oppelt et al., "Evaluation of clinical parameters and estrogen receptor alpha gene polymorphisms for patients with endometriosis," *Reproduction*, vol. 131, no. 1, pp. 153–161, 2006.

[17] N. M’Rabet, R. Moffat, S. Helbling et al., "The CC-allele of the PvuII polymorphic variant in intron 1 of the alpha-estrogen receptor gene is significantly more prevalent among infertile women at risk of premature ovarian aging," *Fertility and Sterility*, vol. 98, no. 4, article e5, pp. 965–972, 2012.

[18] S. Altmäe, K. Haller, M. Peters et al., "Allelic estrogen receptor 1 (ESR1) gene variants predict the outcome of ovarian stimulation in *in vitro* fertilization," *Molecular Human Reproduction*, vol. 13, no. 8, pp. 521–526, 2007.

[19] D. Loutradis, C. Theoananis, E. Anagnostou, D. Mavergianni, and G. A. Partisnevelos, "Genetic profile of SNP(s) and ovulation induction," *Current Pharmaceutical Biotechnology*, vol. 13, no. 3, pp. 417–425, 2012.

[20] M. Koler, H. Achache, A. Tsafir, Y. Smith, A. Revel, and R. Reich, "Disrupted gene pattern in patients with repeated *in vitro* fertilization (IVF) failure," *Human Reproduction*, vol. 24, no. 10, pp. 2541–2548, 2009.

[21] W. Hu, Z. Feng, L. Ma et al., "A single nucleotide polymorphism in the MDM2 gene disrupts the oscillation of p53 and MDM2 levels in cells," *Cancer Research*, vol. 67, no. 6, pp. 2757–2765, 2007.

[22] C. L. Stewart, "Reproduction: the unusual suspect," *Nature*, vol. 450, no. 7170, p. 619, 2007.

[23] D. Metcalf, "Leukemia inhibitory factor—a puzzling polyfunctional regulator," *Growth Factors*, vol. 7, no. 3, pp. 169–173, 1992.

[24] S. M. Laird, E. M. Tuckerman, C. F. Dalton, B. C. Dunphy, T. C. Li, and X. Zhang, "The production of leukaeemia inhibitory factor by human endometrium: presence in uterine flushings and production by cells in culture," *Human Reproduction*, vol. 12, no. 3, pp. 569–574, 1997.

[25] D. D. Paskulin, J. S. Cunha-Filho, C. A. B. Souza, M. C. Bortolini, P. Hainaut, and P. Ashton-prolla, "TP53 P33N and PEX4 polymorphisms and infertility associated with endometriosis or with post-*in vitro* fertilization implantation failure," *Cell Death and Disease*, vol. 3, article e392, 2012.

[26] M. Canis, J. G. Donnez, D. S. Guzick et al., "Revised american society for reproductive medicine classification of endometriosis: 1996," *Fertility and Sterility*, vol. 67, no. 5, pp. 817–821, 1997.

[27] W. Hu, Z. Feng, A. K. Teresky, and A. J. Levine, "p53 regulates maternal reproduction through LIF," *Nature*, vol. 450, no. 7170, pp. 721–724, 2007.

[28] H. J. Kang, Z. Feng, Y. Sun et al., "Single-nucleotide polymorphisms in the p53 pathway regulate fertility in humans," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 106, no. 24, pp. 9761–9766, 2009.

[29] D. Paskulin, V. R. Paixao-Cortes, P. Hainaut, M. C. Bortolini, and P. Ashton-prolla, "The TP53 fertility network," *Genetics and Molecular Biology*, vol. 35, supplement 1, no. 4, pp. 939–946, 2012.

[30] Y. Li, F. Liu, S. Q. Tan, Y. Wang, and S. W. Li, "Estrogen receptor-alpha gene PvuII (T/C) and XbaI (A/G) polymorphisms and endometriosis risk: a meta-analysis," *Gene*, vol. 508, no. 1, pp. 41–48, 2012.

[31] M. S. Abrao, S. Podgaec, J. A. Dias Jr. et al., "Deeply infiltrating endometriosis affecting the rectum and lymph nodes," *Fertility and Sterility*, vol. 86, no. 3, pp. 543–547, 2006.

[32] W. P. Dmowski, H. M. Gebel, and R. G. Rawlins, "Immunologic aspects of endometriosis," *Obstetrics and Gynecology Clinics of North America*, vol. 16, no. 1, pp. 93–103, 1989.

[33] K. G. Osteen and E. Sierra-River, "Does disruption of immune and endocrine systems by environmental toxins contribute to development of endometriosis?" *Seminars in Reproductive Endocrinology*, vol. 15, no. 3, pp. 301–308, 1997.

[34] R. N. Taylor, D. I. Lebovic, and M. D. Mueller, "Angiogenic factors in endometriosis," *Annals of the New York Academy of Sciences*, vol. 955, pp. 89–100, 2002.

[35] W. P. Dmowski, J. Ding, J. Shen, N. Rana, B. B. Fernandez, and D. P. Braun, "Apoptosis in endometrial glandular and stromal cells in women with and without endometriosis," *Human Reproduction*, vol. 16, no. 9, pp. 1802–1808, 2001.

[36] A. Béliard, A. Noël, and J. M. Foidart, "Reduction of apoptosis and proliferation in endometriosis," *Fertility and Sterility*, vol. 82, no. 1, pp. 80–85, 2004.

[37] I. P. Ryan and B. N. Taylor, "Endometriosis and infertility: new concepts," *Obstetrical and Gynecological Survey*, vol. 52, no. 6, pp. 365–371, 1997.

[38] K. L. Bruner, L. M. Matrisian, W. H. Rodgers, F. Gorstein, and K. G. Osteen, "Suppression of matrix metalloproteinases inhibits establishment of ectopic lesions by human endometrium in nude mice," *Journal of Clinical Investigation*, vol. 99, no. 12, pp. 2851–2857, 1997.

[39] D. L. Olive and E. Pritt, "Treatment of endometriosis," *The New England Journal of Medicine*, vol. 345, no. 4, pp. 266–275, 2001.

[40] R. Boudjenah, D. Molina-Gomes, A. Torre et al., "Genetic polymorphisms influence the ovarian response to rFSH stimulation in patients undergoing in vitro fertilization programs with ICSI," *PLoS One*, vol. 7, no. 6, Article ID e38700, 2012.
[41] E. Anagnostou, D. Mavrogianni, C. Theofanakis et al., “ESR1, ESR2 and FSH receptor gene polymorphisms in combination: a useful genetic tool for the prediction of poor responders,” Current Pharmaceutical Biotechnology, vol. 13, no. 3, pp. 426-434, 2012.

[42] Ö. U. Ayvaz, A. Ekmekçi, V. Baltaci, H. I. Önen, and E. Ünsal, “Evaluation of in vitro fertilization parameters and estrogen receptor alpha gene polymorphisms for women with unexplained infertility,” Journal of Assisted Reproduction and Genetics, vol. 26, no. 9-10, pp. 503-510, 2009.

[43] S. Altmae, J. Reimand, O. Hovatta et al., “Research resource: interactome of human embryo implantation: identification of gene expression pathways, regulation, and integrated regulatory networks,” Molecular Endocrinology, vol. 26, no. 1, pp. 203-217, 2012.

[44] P. Yenerall and L. Zhou, “Identifying the mechanisms of intron gain: progress and trends,” Biology Direct, vol. 7, article 29, 2012.

[45] B. R. Graveley, “Alternative splicing: increasing diversity in the proteomic world,” Trends in Genetics, vol. 17, no. 2, pp. 100-107, 2001.

[46] H. Le Hir, A. Nott, and M. J. Moore, “How introns influence and enhance eukaryotic gene expression,” Trends in Biochemical Sciences, vol. 28, no. 4, pp. 215-220, 2003.