Intercellular adhesion molecule-1 (ICAM-1) gene polymorphisms in endometriosis

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Endometriosis is a gynaecological disease with a certain genetic background, but the locations of possible genomic aberrations are still poorly clarified. Intercellular adhesion molecule-1 (ICAM-1), which is a surface glycoprotein that promotes adhesion in immunological and inflammatory reactions, seems to play a role in this condition. The aim of this study was to examine the potential associations of ICAM-1 gene polymorphisms with endometriosis and its severity. Specifically, we have studied two polymorphic sites located in codons 241 (G/R241) and 469 (E/K469) of the ICAM-1 gene. Three hundred and sixty-three Italian Caucasian women of reproductive age who underwent laparoscopy for benign pelvic conditions were enrolled in the study. Endometriosis was documented and staged in 188 women while 175 subjects, in whom endometriosis was laparoscopically ruled out, served as the control group. The frequency of the R241 allele was only marginally higher in endometriosis patients than in controls [5.8 versus 2.9%, P = 0.05; odds ratio (OR), 2.1; 95% confidence interval (CI), 1–4.5]. However, a strikingly high frequency of this allele was found in patients with Stage IV endometriosis versus controls (8.6 versus 2.8%, P = 0.008; OR, 3.2; 95% CI, 1.3–7.9). In contrast, the allele and genotype frequencies of the E/K469 polymorphism did not differ significantly between endometriosis and control groups. While the functional correlate of the G/R241 polymorphism remains unclear, this finding indicates that a genetic polymorphism in the ICAM-1 gene domain may contribute to the susceptibility to endometriosis.

Key words: endometriosis/ICAM-1/polymorphism

Introduction

Endometriosis is defined as a condition in which tissue histologically similar to endometrium is found outside the uterine cavity. Nowadays, the most widely invoked hypothesis to explain its origin is the transtubal refluxed theory that implies the ability of endometrial cells, regurgitated in peritoneum with the menstruation, to survive, implant and proliferate (Oral and Arici, 1997; Witz, 2002). This critical phenomenon does not explain why retrograde menses, a semiphysiological occurrence, results in the development of endometriosis in only a minority of women. Moreover, the mechanisms by which regurgitated endometrial cells are cleared from the peritoneal cavity in the majority of individuals are still poorly understood (Braun and Dmowski, 1998).

In this regard, it has been suggested that a peritoneal immunosurveillance network involving different cell types may subservice this role with the consequence that the immune system appears critical for the development of the disease (Braun and Dmowski, 1998; Lebovic et al., 2001). We have previously shown that one of the molecular mediators that may be involved in the complex events that allow the interaction between endometrial and lymphoid cells is intercellular adhesion molecule-1 (ICAM-1) which is a member of the immunoglobulin (Ig) superfamily of adhesion molecules (Vigano et al., 1994, 1998). ICAM-1 is a cytokine-inducible or, in certain cell types, a constitutively expressed but cytokine-up-regulatable single chain transmembrane glycoprotein expressed on a variety of cells of various lineages (van der Stolpe and van der Saag, 1996; Hayflick et al., 1998). The molecule is also shed from the cell, most likely through proteolytic cleavage and is present as a soluble form (sICAM-1) in plasma and other biological fluids (van der Stolpe and van der Saag, 1996). The surface form plays a key role in trans-endothelial migration of neutrophils and in lymphocyte activation, throughout its interaction with two leukocyte integrins, leukocyte function associated antigen-1 (LFA-1) and Mac-1 (Diamond et al., 1991; van der Stolpe and van der Saag, 1996; Hayflick et al., 1998). The soluble form has been shown to retain biological activity with regard to ligand binding and activation, thereby potentially hindering the adhesion between immune cells and their targets (van der Stolpe and van der Saag, 1996). Both the surface and the soluble forms of ICAM-1 have been proposed to be involved in the aetiopathogenetic mechanisms underlying various autoimmune and immune-mediated diseases including disorders of the female reproductive system, such as pre-eclampsia, ovarian stimulation syndrome and endometriosis (Somigliana et al., 1996, 2002; Airoldi et al., 1998; Wu et al., 1998; Daniel et al., 2000; Abramov et al., 2001). In previous studies, our group has demonstrated that both eutopic endometrium and ectopic endometriotic implants express surface ICAM-1 and release the soluble form of the molecule (Somigliana et al., 1996; Vigano et al., 1998). However, endometrium from women with endometriosis has been shown to release higher amounts of sICAM-1 when compared to that derived from women without the disease (Somigliana et al., 1996; Vigano et al., 2000). Finally, increased serum levels of the soluble protein have been detected in women with endometriosis (De Placido et al., 1998; Wu et al., 1998).
Functional or quantitative alterations of ICAM-1 protein in different pathologies have been related to the expression of specific gene variants (Yang, 1997; Mycko et al., 1998; McLaren et al., 1999; Macchioni et al., 2000). Two single-base polymorphisms of this gene have been described and are located in exons 4 and 6, modifying codons 241 and 469 respectively (Macchioni et al., 2000). Both result in amino acid changes and can potentially lead to different interactions with ICAM-1 ligands. The polymorphism at codon 241 has been significantly associated with rheumatoid arthritis (Macchioni et al., 2000), inflammatory bowel disease (Braun et al., 2001) and giant cell arteritis (Salvarani et al., 2000). The polymorphism at codon 469 has been significantly associated with Bechet’s disease (Verity et al., 2000), chronic renal allograft failure (McLaren et al., 1999) and multiple sclerosis (Mycko et al., 1998).

Endometriosis has a certain genetic component with an increased risk in siblings compared with the general population (Simpson et al., 1980). The locations of possible genomic aberrations are still poorly clarified. To contribute to the analysis of the genetic background of endometriosis, we have evaluated the frequency of ICAM-1 polymorphisms at codon 241 and 469 in a population of Italian origin affected by laparoscopic-proven disease. They were compared to a group of women from the same geographic area in whom diagnosis of endometriosis was laparoscopically ruled out.

Materials and methods

Patients and controls

ICAM-1 allele and genotype frequencies in codons 241 and 469 were studied in women who attended the endoscopic surgical service of the II Department of Obstetrics and Gynecology of the University of Milan to undergo gynaecological laparoscopy. The sample sizes used in this study for each polymorphism evaluated allowed us to detect at the usual level of study power (80% statistical power of avoiding a type II error and 5% level of significance) at least a doubling in the risk of developing the disease carrying the mutated allele.

Therefore, 363 women were enrolled in the study. All individuals were of Italian Caucasian origin and were of reproductive age (mean age ± SD, 32.2 ± 6.2 years). All women underwent complete pre-surgery clinical examination before the diagnostic-operative laparoscopy. Indications to laparoscopy included chronic pelvic pain, infertility, ovarian cysts and myomas. None of the women had been previously subjected to surgery for endometriosis (laparotomy or laparoscopy) or were taking medications except for non-steroidal anti-inflammatory drugs. In particular, they did not receive GnRH agonist or other ovarian suppressive drugs for at least 3 months prior to surgery.

Patients were excluded from the study if they were affected by an autoimmune disorder or if the laparoscopic visualization showed evidence of severe abdominal pelvic adhesions that could preclude the identification of endometriotic foci.

Endometriosis was always diagnosed during the laparoscopic intervention. Diagnosis of ovarian endometriotic cysts and deep peritoneal lesions were also confirmed histologically. Diagnosis of superficial peritoneal lesions was based on direct visualization when endometriotic implants presented as typical lesions. Lesions whose aspect was dubious were removed in order to confirm their endometriotic nature by histological examination. Three physicians active in the evaluation and treatment of endometriosis staged patients according to the revised American Society for Reproductive Medicine (rASRM) classification (American Society for Reproductive Medicine, 1997).

For the polymorphism at codon 241, we have evaluated the complete series of women enrolled in the study. Endometriosis was documented in 188 women (51.8%). Stage of the disease was found to be minimal (Stage I) in 40 cases (21.3%), mild (Stage II) in 16 cases (8.5%), moderate (Stage III) in 74 cases (39.4%) and severe (Stage IV) in 58 cases (30.8%). Other gynaecological benign pathologies were present in 48 of these 188 women with endometriosis (27 cases of uterine myomas, 20 cases of non-endometriotic benign adenexal cysts and one uterine malformation). The presence of endometriotic cysts (endometriomas) was observed in 119 of these 188 women. Deep endometriosis, defined as lesions infiltrating to a depth of at least 5 mm beneath the peritoneal surface, was observed in 36 of these 188 women. One hundred and seventy-five women who underwent laparoscopy and in whom no endometriosis was found served as the control group. Specifically, they included 32 cases with a regular pelvis, 26 cases of serous cysts, 24 cases of uterine myomas, 21 cases of dermoid cysts, 18 cases of functional cysts, 15 cases of uterine malformations, 15 cases of pelvic adhesions, eight cases of parovarian cysts, seven cases of mucinous cysts, eight cases of pelvic inflammatory disease and one case of ovarian fibroma. The mean age ± SD of patients of endometriosis and controls was 33.5 ± 7.7 and 32.4 ± 5.7 years respectively.

For the polymorphism at codon 469, we have evaluated the first consecutive 100 women that were enrolled in the study. Endometriosis was documented in 46 women (46%). Stage of the disease was found to be minimal (Stage I) in 13 cases (28.3%), mild (Stage II) in seven cases (15%), moderate (Stage III) in 16 cases (35%) and severe (Stage IV) in 10 cases (21.7%). Other gynaecological benign pathologies were present in nine of these 46 women with endometriosis (four cases of uterine myomas and five cases of non-endometriotic benign adenexal cysts). The presence of endometriotic cysts was observed in 20 of these 46 women. Deep endometriosis was observed in six of these 46 women. Fifty-four women who underwent laparoscopy and in whom no endometriosis was found served as the control group. They included 10 cases with a regular pelvis, 11 cases of serous cysts, 10 cases of uterine myomas, 10 cases of dermoid cysts, three cases of functional cysts, five cases of uterine malformations, five cases of pelvic adhesions, two cases of pelvic inflammatory disease and one case of mucinous cysts.

Patients were informed that blood would be used for research purposes and gave written consent. Approval for this study was granted by the local Human Institutional Review Committee.

Molecular analysis of ICAM-1 polymorphisms

Genomic DNA was isolated from 1 ml of whole blood collected in ethylenediaminetetra-acetic acid (EDTA). To detect the substitution of arginine for glycine responsible for the ICAM-1 polymorphism at position 241 (G/R) in exon 4 of the ICAM-1 gene, we developed an allele-specific PCR method (AS-PCR). We designed allele-specific primers, ‘5-GTGGTCCTGTCCTGGAGCAGGCG-3‘ and ‘5-GTGGTCGTCCTGTCCTGGAGCAGGCGA-3‘, with the last nucleotide complementary to the allelic variant substitution base using published sequence information on the point mutation in question of the gene ICAM-1, and a common primer, ‘5-GCGGTACACTGACTGGCGCT-3‘. AS-PCR was performed in a total volume of 50 µl that contained 0.2 ng of genomic DNA, each primer pair consisting of 10 pmol of allele-specific primer and 10 pmol of common primer, 200 µM each dNTP, 10 µM Tris–HCl (pH 8.3); 50 mM KCl, 1.5 mM MgCl2 and 0.5 IU Taq DNA polymerase (Promega, Milano, Italy). PCs performed without DNA represented the negative controls. The reaction was carried out for 30 cycles each consisting of a denaturation step at 94°C for 20 s, 62°C for 20 s and 72°C for 20 s in a Progene (Techne, Cambridge, UK). The amplified PCR products of 137 bp were analysed by 2% agarose gel electrophoresis followed by ethidium bromide staining and ultraviolet visualization. Over 100 samples were genotyped twice in independent experiments to assess the consistency of the assay and in no case was a discrepancy observed. Cases with G/R or R/R genotypes were always confirmed at least twice. Moreover, for at least five of them, the nucleotide sequence of the PCR products was confirmed by sequence analysis. Sequencing reactions were prepared using the Big dye terminator chemistry (PE Applied Biosystems, Foster City, CA, USA) and analysed on the ABI Prism 310 Genetic Analyzer.

The second amino acid polymorphism, which consists of a substitution of lysine for glutamic acid (E/K469), was detected as described by Macchioni et al. (2000). The amplified fragment of 111 bp was digested with 5 IU BstUI, which cut product from the E469 allele into two fragments of 85 and 26 bp but not from the K469 allele. The fragments were analysed by 4% agarose gel electrophoresis.

Statistical analysis

Statistical analysis was performed using the SPSS statistical package. The frequencies of the alleles and genotypes among the patient and control groups
were determined and were then compared by \( \chi^2 \)-test. Odds ratios (ORs) were calculated, together with their 95% confidence intervals (CIs).

**Results**

In this prospective study, patients and controls were examined before the results of genetic analysis were known and laparoscopic surgeons were therefore masked to the ICAM-1 genotype of the individuals. Since diagnosis of endometriosis can be definitively proven by laparoscopy and the disease is often asymptomatic, controls consisted of women in which the disease was laparoscopically ruled out. The frequencies of the R241 and E469 alleles in our control population agreed with those reported by Macchioni *et al.* (Macchioni *et al.*, 2000) for a group of healthy Italian volunteer blood donors (3.1 and 43.0% respectively) which are also very similar to those observed in the controls of the Centre D’Etudes du Polymorphisme Humain.

### G/R 241 ICAM-1 polymorphism: allele and genotype frequency

For the point mutation polymorphism located in ICAM-1 exon 4, which contains the information for codon 241, two alleles were present (i.e. allele G and allele R) and three different genotypes (i.e. homozygotes G241 and R241 or heterozygote G/R241) (Figure 1). Table I shows the allele and genotype frequencies of the G/R241 ICAM-1 gene polymorphism in 188 cases of endometriosis and 175 controls. In both groups, there was a strong predominance of allele G. The frequency of allele R was 2-fold higher in patients with endometriosis, compared to controls but this difference was only marginally statistically significant (OR, 2.1; 95% CI, 1–4.5).

In both groups of patients, the codon 241 genotype frequency pattern showed dominance of the allele G homozygote. Results from this analysis are shown in Table I. There was no statistically significant difference in the distribution of the G/R241 genotype. We detected only two individuals homozygotes for the R241 allele and both were in the endometriosis group (1.1%). The first patient had severe endometriosis and was re-operated for a deep form of the disease infiltrating the rectum after 1 year. The second patient had an ovarian cyst and several surface peritoneal implants.

We then analysed the endometriosis group according to the presence or absence of the different forms and features of the disease.

### Table I. Allele and genotype frequencies of G/R241 polymorphism in endometriosis patients and controls

<table>
<thead>
<tr>
<th>Variable</th>
<th>Endometriosis</th>
<th>Controls</th>
<th>( P )</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Allele</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>R</td>
<td>22/376 (5.8%)</td>
<td>10/350 (2.9%)</td>
<td>0.05</td>
</tr>
<tr>
<td>G</td>
<td>354/376 (94.1%)</td>
<td>340/350 (97.1%)</td>
<td></td>
</tr>
<tr>
<td><strong>Genotype</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Codominant model</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>RR</td>
<td>2/188 (1.1%)</td>
<td>0/175 (0%)</td>
<td>0.06*</td>
</tr>
<tr>
<td>GR</td>
<td>18/188 (9.6%)</td>
<td>10/175 (5.7%)</td>
<td></td>
</tr>
<tr>
<td>GG</td>
<td>168/188 (89.3%)</td>
<td>165/175 (94.3%)</td>
<td></td>
</tr>
<tr>
<td><strong>Recessive model</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>GG</td>
<td>168/188 (89.3%)</td>
<td>165/175 (94.3%)</td>
<td>0.09</td>
</tr>
<tr>
<td>RR+GR</td>
<td>20/188 (10.7%)</td>
<td>10/175 (5.7%)</td>
<td></td>
</tr>
<tr>
<td><strong>Dominant model</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>GR+GG</td>
<td>186/188 (98.9%)</td>
<td>175/175 (100%)</td>
<td>0.5</td>
</tr>
<tr>
<td>RR</td>
<td>2/188 (1.1%)</td>
<td>0/175 (0%)</td>
<td></td>
</tr>
</tbody>
</table>

*\( \chi^2 \) for trend.

### Table II. Frequency of ICAM-1 R241 allele in patients with endometriosis according to different forms of the disease

<table>
<thead>
<tr>
<th>R241 allele</th>
<th>Peritoneal lesions</th>
<th>Deep endometriosis</th>
<th>Adhesion*</th>
<th>Ovarian cyst**</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>13/234 (5.6%)</td>
<td>4/72 (5.6%)</td>
<td>9/174 (5.2%)</td>
<td>2/56 (8.9%)</td>
</tr>
<tr>
<td></td>
<td>(0.9–4.6)</td>
<td>(0.6–6.6)</td>
<td>(0.7–4.7)</td>
<td>(1.1–10.1)</td>
</tr>
</tbody>
</table>

ORs were calculated comparing values to those of control group.

*\( \chi^2 \) for trend \( P = 0.025 \).

**\( \chi^2 \) for trend \( P = 0.017 \).

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*Figure 1. G/R genotyping by allele-specific PCR. DNA from selected patients with three different genotypes (GG, GR, RR) was amplified with both primers specific for the G241 and R241 alleles. PCR products were of the expected size (137 bp). The far left lane shows the DNA molecular weight marker.*
disease (presence of deep endometriosis, peritoneal surface implants, adhesions and endometriotic cysts) (Tables II and III). As shown in Table II, the frequency of the R241 allele appeared to be associated with some parameters indicative of a severe disease (i.e. presence of more than one cyst and a high rASRM adhesion score).

Ten out of the 22 (45%) R241 alleles demonstrated in the endometriosis group were detected in patients with a severe (Stage IV) form of the disease. More specifically, patients with severe endometriosis were over three times as likely to carry the R241 allele (OR, 3.2; 95% CI, 1.3–7.9). The frequency of allele R241 resulted in 8.6% of patients with severe endometriosis versus 2.9% in controls ($P$ = 0.008). The distribution of the G/R241 genotype was also statistically different between the two groups. The prevalence of heterozygosity (G241/R241) and homozygosity (R241/R241) was 13.8 and 1.7% respectively in patients with severe endometriosis compared to 5.7 and 0% respectively in women without the disease ($c^2$ for trend, $P$ = 0.01).

The analysis of patients according to disease duration and age at onset of endometriosis revealed no difference between those carrying or not carrying the R241 allele.

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The analysis of patients according to disease duration and age at onset of endometriosis revealed no difference between those carrying or not carrying the R241 allele.

E/K469 ICAM-1 polymorphism: allele and genotype frequency

For the point mutation polymorphism located in ICAM-1 exon 6, which contains the information for codon 469, two alleles were present (i.e. allele E and allele K) and three different genotypes (i.e. homozygotes E469 and K469 or heterozygote E/K469) (Figure 2).
Table IV shows the allele and genotype frequencies of E/469 ICAM-1 gene polymorphism in 46 cases of endometriosis and 54 controls.

In our population, we observed a predominance of allele K (62%). The differences in distributions of alleles K and E in the control and endometriosis groups were not statistically significant (63 versus 61% and 37 versus 39% respectively).

The codon 469 genotype frequency pattern showed dominance of the E/K heterozygote (49%), whereas the frequency patterns of the allele K homozygote and allele E homozygote were 38 and 13% respectively. As for allele frequencies, the differences in distributions of the three 469 genotypes in the control and endometriosis groups were not statistically significant.

Discussion

Endometriosis is a pathological condition in which multiple genetic components, in combination with not yet completely elucidated environmental risk factors, are important determinants of the disease process and clinical outcome (Campbell and Thomas, 2001). Indeed, genetic analysis revealed a correlation of the disorder with common polymorphisms in low-penetration genes coding for enzymes responsible for the metabolism of a broad range of xenobiotics and carcinogens (Baranova et al., 1999; Arvanitis et al., 2001; Baxter et al., 2001). Moreover, isolated reports indicate genetic variants of genes involved in hormone response as all plausible risk factors (Campbell and Thomas, 2001). Thus far, little is known about other candidate genes involved.

We report here, for the first time, evidence of the possible involvement of the ICAM-1 gene polymorphism at codon 241 in the genetic predisposition to endometriosis. ICAM-1 is a protein with a core of 55 kDa with five extracellular Ig-like domains (Diamond et al., 1991; Hayflick et al., 1998) coded by the second to the sixth exons. The human ICAM-1 gene is a single copy gene located on chromosome 1p13 and is known to contain at least two polymorphic sites, situated in codons 241 (G/R241) and 469 (E/K469) (Vora et al., 1994; Wenzel et al., 1996). These two polymorphisms are single base mutations. At position 241, located in exon 4, the mutation GGG-AGG results in the exchange of glycine for arginine in the protein Ig-like domain 3. Similarly, at position 469, located in exon 6, the mutation AAG-GAG also lead to an amino acid change (lysine–glutamine) in the Ig-like domain 5 (Macchioni et al., 2000).

In our study, in a total of 363 women, we found a higher frequency of the R241 allele of ICAM-1 gene in a group of women with endometriosis (5.8%) compared to a control population without the disease (2.9%). A great proportion of this polymorphism was present in patients with a severe form of the disease in which the frequency of the R241 allele was strikingly higher (8.6%). Therefore, this ICAM-1 gene variant might have a certain aetiological significance in endometriosis.

The functional role of these point mutations should be related to the altered amino acid coding sequence. In this regard, it is noteworthy that the exon 4 mutation is located in the domain which has been shown to be of importance in binding to the Mac-1 form of the leukocyte integrin (Diamond et al., 1991). The interaction between Mac-1 and ICAM-1 makes an important contribution to leukocyte adhesion in the execution of immunological and inflammatory functions and may play a role in regulating localization of leukocytes (van der Stolpe and van der Saag, 1996). The mutation of glycine with arginine in the third domain of ICAM-1 might affect the adhesive function of ICAM-1. Indeed, the common allele G241, which encodes for glycine, is maintained in the analogous position in different species thus supporting the importance of this site for the control of ICAM-1-mediated interactions (Yang, 1997). Moreover, it has been shown that single amino acid changes in the third domain of ICAM-1 can strongly influence binding to Mac-1 in vivo and, consequently, polymorphisms in this region may likewise provide both a gain or a loss of function (Diamond et al., 1991). Therefore, there is a substantial possibility that the protein product derived from the rare allele R241 might be responsible for a different recruitment and activation of immune cells that could contribute to the inflammatory response in endometriosis. In keeping with this hypothesis, polymorphism of the ICAM-1 gene at codon 241 has been associated with susceptibility to other immune-mediated diseases that, similarly to endometriosis, are characterized by an important inflammatory process with a strong expression of ICAM-1 in the lesions and elevated serum level of the soluble molecule (Macchioni et al., 2000; Salvarani et al., 2000; Boiardi et al., 2001). However, experimentally proven evidence that this base substitution can modify molecular activity is still lacking.

The fact that the frequency of the codon 241 polymorphism is higher in patients with Stage IV endometriosis may suggest that the mutated ICAM-1 protein is involved in the disease pathogenesis more as a disease-modulating factor rather than in its initial development. In this regard, it has to be pointed out that the presence of the mutated allele resulted in an association with an increased risk of developing severe adhesions, a deep form of the disease and several cysts. On the other hand, in women with a single ovarian cyst the prevalence of the polymorphism was similar between patients and controls. There is a potential explanation for these observations. It has been suggested that there may exist different endometriomas with a different pathogenesis (Nisolle and Donnez, 1997). Therefore, it might be speculated that the codon 241 polymorphism may play a role in some but not all pathogenetic types of the disease.

The prevalence of ICAM-1 gene polymorphisms in our controls was in complete agreement with previously published reports for the Caucasian population (Macchioni et al., 2000). We have extensively evaluated our control group to better understand the potential impact of the sampling design on the reported association. Our controls consisted of women in whom endometriosis was laparoscopically excluded. Other studies enrolled healthy individuals as control subjects (Baranova et al., 1999; Baxter et al., 2001). We believe that our study design is more accurate considering that the disease can be diagnosed only by surgical visualization. A control group consisting of healthy individuals who did not undergo pelvic examination, might include asymptomatic affected women. This could lead to the possibility to overestimate the frequency of the G/R241 polymorphism in the control population with important consequences on statistical analysis especially for a rare polymorphism.

Finally, we have to recognize that, although our data support the possibility that the presence of the R241 allelic variant may represent a predisposing factor for endometriosis, we cannot rule out that these results may be due to linkage disequilibrium with other unknown mutations. In the same area of chromosome 19, in close proximity to the ICAM-1 gene, there are genes which may influence the inflammatory response such as ICAM-3, interleukin-11 and human heat shock protein 40 (Macchioni et al., 2000).

To the best of our knowledge, this is the first evidence of an association between endometriosis and a polymorphic variant of a gene important in the immune system and leads to the hypothesis that specific endometriosis predisposing genes may be the same as those associated with other common inflammatory conditions. However, given the low allelic frequency of this variant in the population studied and the potential effects of ethnic stratification, these data should be
confirmed in more than one ethnic group. In any case, the low frequency of this polymorphism would not obviously justify a population screening to identify potentially affected women. Rather, genotyping of the patients could be useful to select subgroups that could develop a more severe form of the disease.

References


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