Endometriosis: Identification by Carbonic Anhydrase Autoantibodies and Clinical Features*

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ABSTRACT

Reliably diagnosing endometriosis traditionally requires surgery. To evaluate a possible non-surgical method, a case-control series of unexplained infertility patients undergoing diagnostic laparoscopy were scored by clinical criteria and reactivity to human carbonic anhydrase II by Western blotting. The CA II autoantibodies were found in none of the fertile controls, 38 percent of infertile controls, 55 percent of stage 1, 50 percent of stage 2, 73 percent of stage 3, and 85 percent of stage 4 endometriosis patients. Advanced endometriosis was associated with more intense reactivity. Combining clinical and antibody scores for infertile groups showed a positive association with disease stage with positive predictive values of 76 to 95 percent, negative predictive values of 90 to 60 percent, and a likelihood ratio of 18.3.

It is concluded by us that CA II immunoreactivity, clinical, and combined scores all identified stages 2 to 4 endometriosis patients. However, based on predictive values and likelihood ratios, the combined score is best at identifying endometriosis non-surgically.

Introduction

The prevalence of endometriosis has been estimated to vary between 71 and 20 percent of asymptomatic women undergoing tubal sterilization and between 21 and 47 percent of infertile women.3,4 The classical symptoms of dysmenorrhea, dyspareunia, and dyschezia have failed to identify many of the patients afflicted with the disease, particularly those with milder forms, as many patients are largely asymptomatic except for the impairment of fertility.5,6 Hence, it is

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often difficult to decide when surgical investigation for endometriosis is justified in spite of the fact that it is frequently associated with infertility.

Since endometriosis was first proposed as an autoimmune disorder by Weed et al., various clinical assays have been described as evidence of the local and systemic activation of the autoimmune process. These assays include circulating complement levels, prostanoid concentrations, peritoneal fluid volume and constituents, and macrophage type, activation and concentration. CA-125 levels in peripheral blood, or peritoneal fluid and unique proteins seen on Western blots. Alterations in all these assays have been associated with the presence of endometriosis. No marker has yet proven to be significantly predictive to sway the clinical decision of whether or not to perform laparoscopy on a patient complaining of infertility. Since polyclonal B cell activation has been proposed as part of the pathophysiology of endometriosis and of the associated infertility, Gleicher (personal communication) has proposed omitting surgical investigation altogether in cases where tubal status appears normal on hysterosalpingogram and selective salpingography. He reasoned that the visible lesions seen at surgery are only a visible manifestation of a much more extensive disease process which is not curable by surgical excision.

Carbonic anhydrase (carbonate dehydratase, EC 4.2.1.1) is a widely distributed enzyme that catalyzes the reversible reaction of \( \text{H}_2\text{CO}_3 \) to \( \text{CO}_2 \) and \( \text{H}_2\text{O} \). This reaction is important in maintaining local acid/base requirements throughout the body. It has been shown in recent years that carbonic anhydrase exists in mammals as seven isoenzymes, defined by their tissue of origin and enzymatic properties. Carbonic anhydrases I and II are cytosolic, with carbonic anhydrase II the most widely distributed in secretory and absorbing epithelia and red cells. In the human reproductive tract, carbonic anhydrase has been located in the ovary, fallopian tube, and uterine endometrium, by the metal salt capture method which does not differentiate isoenzymes of carbonic anhydrase. A specific non-erythrocyte carbonic anhydrase (U) has been reported in human endometrium, where its concentration has been shown to be modulated by ovarian steroid hormones. Carbonic anhydrase I, II, and III have recently been found in the ovary, fallopian tubes, and uterus of rodents by immunocytochemistry. Others have shown a relationship between serum autoantibodies to bovine and human carbonic anhydrase I and II and autoimmune diseases including systemic lupus erythematosus, systemic sclerosis, polymyositis, and Sjögren's syndrome, suggesting that autoantibodies to carbonic anhydrase are part of the pathophysiology of these diseases. Autoantibodies to carbonic anhydrase I and II have been reported in patients with endometriosis.

The present authors wished to test the hypothesis that immunoreactivity to carbonic anhydrase II is related to the existence of endometriosis, a putative autoimmune disease, in an infertility practice. Human carbonic anhydrase II was chosen as the specific antigen for this investigation based on a pilot study showing more reactivity to carbonic anhydrase II than to carbonic anhydrase I in the serum of endometriosis patients. A simple scoring system was used for the most common pelvic symptoms (dysmenorrhea and dyspareunia) and pelvic findings. Using this scale, examinations were made to ascertain whether or not antibody to carbonic anhydrase II would more accurately predict endometriosis than our clinical score. The last examinations were made to ascertain whether combining clinical criteria with seropositivity to carbonic anhydrase II antibody
would be a better predictor of disease than either clinical or laboratory findings alone.

Materials and Methods

**Patient Selection**

Patients studied comprised a total of 113 subjects including 103 attending the Fertility center at William Beaumont Hospital and 10 drawn from the rheumatology service. This protocol was approved by the Human Investigation Committee of the Beaumont Research Institute, including informed consent. Patients studied were divided into four major groups with no statistical difference in mean age. Weight, socioeconomic, marital and smoking status are not known to affect the risk of endometriosis. The first patient group was 17 known fertile controls who had no symptoms suggesting endometriosis who presented for elective tubal sterilization or consideration of tubal reversal. No patient had visible disease at the time of the procedure. The second group was comprised of 29 patients whose diagnostic workup excluded the presence of endometriosis, i.e., had a negative diagnostic laparoscopy. These patients suffered from male (five), ovulatory (four), tubal factor (seven), or unexplained infertility (13). Unexplained infertility patients by definition had a normal semen analysis, basal body temperature graph, post coital test, hysterosalpingogram, and endocrine studies in addition to a negative diagnostic laparoscopy within 18 months of assay.

The third group consisted of 10 patients from the rheumatology service who exhibited a variety of collagen-vascular diseases, all of whom had high antinuclear antibody titers (>1/40). These patients did not undergo diagnostic laparoscopy. The fourth group included 57 infertile patients with confirmed endometriosis by visual (operative) criteria. Pathologic confirmation (peritoneal biopsy) was performed when classic powder burn type lesions were lacking and when peritoneal excision was deemed more appropriate than vaporization. It was performed in 35 of the 57 endometriosis cases, but not in the fertile or infertile control groups. The patients in this group were further subdivided into groups based on their revised American Fertility Society stage (I to IV).²⁷

Stage assignment was done by a single author; in the few cases where he was not the surgeon of record, review of video prints or tape was utilized. Patients with large masses (>6cm) mandating surgical investigation were excluded from this study as they would require surgical investigation regardless of results. Patients were not disqualified if they had previously been treated medically for endometriosis, but the particular drug regimen was noted along with any other possible clinical confounding variables, i.e., thyroid, liver, or rheumatologic disease. These conditions have previously been shown to affect immunoreactivity to carbonic anhydrase.²⁴,²⁵ Patients who were currently on medical suppressive therapy with either danazol or a GnRH agonist or who had been on suppressive therapy in the previous year were excluded from the study. A total of 13 patients had prior medical therapy; seven with danazol (3,3,5,5,5,5, and 9 years prior to assay), four with GnRHa (1,1,1, and 2 years prior to assay), and two with oral contraceptives (both 1 year prior to assay). No patients were on oral contraceptives or progestins at the time of assay.

In an effort to determine the strength of clinical diagnostic criteria, fertile control and infertile control patients were asked to fill out a questionnaire inquiring whether or not they had symptoms of dysmenorrhea and/or dyspareunia and underwent a standard pelvic exam to identify pelvic tenderness, mass, or fixa-
tion. Symptoms and pelvic findings were scored numerically based on the patients' own subjective perception of severity and the examining physician's impression of pelvic findings. Dysmenorrhea was scored as none (=0, absence of any pain), mild (=1, pain tolerable without medical therapy), moderate (=2, pain tolerable with medical therapy), or severe (=3, inability to perform routine tasks even with therapy). Dyspareunia was scored as 0 or 1 based on absence or presence of any coital discomfort. Pelvic findings were scored as normal (=0), pelvic tenderness only (=1), or palpably abnormal with mass, nodularity, or fixation (=2). The maximum clinical score was 6.

Patients were given numerical scores based on the degree of each individual symptom, a sum of scores for dysmenorrhea, dyspareunia, and pelvic findings (clinical score), and a sum of the clinical score and the carbonic anhydrase antibody assay result (combined score). The maximum combined score was equal to 9.

ASSAY PROCEDURES

Assays for carbonic anhydrase II antibody were performed on blood samples taken either at laparoscopy, or during subsequent cycles where patients were undergoing superovulation cycles and having phlebotomy for baseline follicular phase steroid determinations (infertile controls only). All serum samples were stored at −20°C until assayed. Samples with visible hemolysis were discarded to prevent potential erythrocyte carbonic anhydrase I or II contamination.

Sodium dodecyl sulfate-polyacrylamide gel electrophoresis of carbonic anhydrase isoenzyme II, electrophoretic transfer to nitrocellulose, and incubation of the immobilized isoenzyme with serum from patients to screen for autoantibodies for analysis by Western blot were done as described previously. The staining intensity of the colored reaction product resulting from immunoblotting was visually graded. The reaction was considered positive if the staining intensity was greater or equal to that of a positive control of pooled sera made from previously tested endometriosis patients. Strongly positive reactions (=3) had a staining intensity that was markedly stronger than the positive control (=2). Weakly positive reactions (=1) had a staining intensity that was less than the positive control but more than the negative controls (=0).

STATISTICAL METHODS

To test for group differences in identification of endometriosis, Pearson’s chi-square was used on collapsed binary versions of each clinical variable, carbonic anhydrase II immunoreactivity, clinical score, and combined score. For dysmenorrhea, a score of 2 or greater was considered positive; for dyspareunia, a score of 1 was positive; and for pelvic exam findings, a score of 1 or greater was considered positive. For immunoreactivity to human carbonic anhydrase II, any reactivity (1 to 3) was considered positive. For the clinical score, a total of 3 or more points from dysmenorrhea, dyspareunia, and pelvic exam scores was considered positive. For the combined score, a total of 4 or more points from the sum of the immunoreactivity score and the clinical score was considered positive. This value was chosen to represent the assay result added to an equivocal clinical assessment.

Because rheumatology service patients were not subject to either pelvic or laparoscopic exams, they were excluded from this portion of the analysis but used to compare the antibody assay to results obtained during prior observations with similar groups of patients where reactivity was observed. P-values for Pearson’s chi-square were obtained using.
either an exact method when possible or a Monte Carlo method in StatXact. P-values based on the chi-square distribution were inappropriate given the presence of some small expected frequencies. These exact and Monte Carlo p-values assess the strength of evidence against the null hypothesis for tables with the same set of row and column totals as the observed table. All p-values reported are two-sided. Confidence intervals for sensitivity for each of these test procedures were obtained for each stage of endometriosis; confidence intervals for specificity were obtained for the infertile controls without endometriosis. These intervals were obtained from the Blyth and Still confidence interval table for binomial proportions appropriate for small samples.

Linear-by-linear association tests were used to assess whether there was an increasing trend in a test score with endometriosis severity; for severity, consecutive integer scores were used with a score of 1 for infertile controls without endometriosis to 5 for patients with stage IV endometriosis. Positive predictive values were calculated using a weighted average of stage-specific sensitivities to reflect unequal sampling rates among the stages. In the absence of published data for endometriosis prevalence by each of the four stages, data from our practice was used. Likelihood ratios were also calculated comparing infertile controls with all stages and separately for stages II to IV since many clinicians discount the significance of minimal (stage I) disease.

McNemar's test was used to compare pairs of test procedures on the proportion of women identified as having endometriosis; these comparisons were done separately for each of the five groups of infertile women. P-values for McNemar's test were based on the binomial distribution. Since the effective sample size for McNemar's test is the number of women for whom two test procedures disagree, these tests have small statistical power. As a consequence, large p-values do not indicate the absence of a difference. McNemar's test addressed the hypothesis of equal sensitivity when applied to endometriosis stages and the hypothesis of equal specificity when applied to infertile controls.

Results

There was no statistically significant difference in age among the patient groups. Total clinical scores were associated with endometriosis when a clinical discriminatory score of 3 was used (data not shown). This value was chosen to represent the most frequently encountered clinical dilemma (i.e., a history of moderate or severe dysmenorrhea with mild tenderness on pelvic exam) to see if clinical impressions alone are accurate enough to justify the decision to perform laparoscopy.

The immunoreactivity of all of the patient groups to human carbonic anhydrase II antigen are shown in table I. The frequency results show that the intensity of reaction to the antigen tended to increase with the stage of disease. When differences between infertile groups and carbonic anhydrase antibody titers were examined, significant linear by linear association between the intensity of immunoreactivity and stage of disease was established with more severe endometriosis having more intense immunoreactivity to carbonic anhydrase II (P < 0.001). Revised American Fertility Society staging of endometriosis was also strongly associated in a linear-by-linear fashion with a combined score ≥4 (P < 0.0001).

The numbers of patients positive for endometriosis identified by anticarbonic anhydrase II antibody assay, clinical score and combined score are shown in table II. P-values for McNemar's tests comparing the ability of the three scoring
methods to identify endometriosis in infertile patients are reported in table III. Positive and negative predictive values for the three scoring methods were calculated for a range of prevalences and are reported in table IV. Of the infertile patients who met the criteria for inclusion in this study, 59 percent had endometriosis diagnosed by laparoscopy. The prevalence of stage I disease was 15.4 percent, stage II was 17.5 percent, stage III was 15.4 percent, and stage IV was 10.5 percent.

**Discussion**

Analysis of the clinical data shows a trend toward higher clinical scores in the endometriosis patient groups, especially as the stage of disease increases (data not
<table>
<thead>
<tr>
<th>Patient Group (number)</th>
<th>Immunoreactivity to HCA II</th>
<th>Clinical Score ≥ 3&lt;sup&gt;g&lt;/sup&gt;</th>
<th>Combined Score ≥ 4&lt;sup&gt;g&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>N&lt;sub&gt;a&lt;/sub&gt;</td>
<td>Pb (%)</td>
<td>Cl Sen&lt;sub&gt;b&lt;/sub&gt; (%)</td>
</tr>
<tr>
<td>Fertile control (n = 17)</td>
<td>0</td>
<td>0</td>
<td>—</td>
</tr>
<tr>
<td>Infertile control (n = 29)</td>
<td>11</td>
<td>38</td>
<td>—</td>
</tr>
<tr>
<td>Stage I (n = 11) Endometriosis</td>
<td>6</td>
<td>55</td>
<td>(26,80)</td>
</tr>
<tr>
<td>Stage II (n = 18) Endometriosis</td>
<td>9</td>
<td>50</td>
<td>(27,73)</td>
</tr>
<tr>
<td>Stage III (n = 15) Endometriosis</td>
<td>11</td>
<td>73</td>
<td>(47,90)</td>
</tr>
<tr>
<td>Stage IV (n = 13) Endometriosis</td>
<td>11</td>
<td>85</td>
<td>(57,97)</td>
</tr>
</tbody>
</table>

HCA II = Human carbonic anhydrase II.
<sup>a</sup>N = Number of patients identified as positive for endometriosis.
<sup>b</sup>Percent positive: estimated sensitivity for endometriosis patients, 100% - estimated sensitivity for patients without endometriosis.
<sup>c</sup>95% confidence interval for sensitivity.
<sup>d</sup>95% confidence interval for specificity.
<sup>e</sup>Any reactivity to HCA II is considered positive.
<sup>f</sup>A total of three or more points from dysmenorrhea, dyspareunia and pelvic exam scores is considered positive (p < 0.0001).
<sup>g</sup>Immunoreactivity score (0-3) + clinical score (0-6); a total of 4 or more points is considered positive (p < 0.0001).
TABLE III
Comparison of Screening Methods for Endometriosis, P-Value for McNemar's Test

<table>
<thead>
<tr>
<th>Patient Groups</th>
<th>Immuno-reactivity to HCAII(a) vs. Clinical Score ≥ 3(b)</th>
<th>Immuno-reactivity to HCAII(a) vs. Combined Score ≥ 4(c)</th>
<th>Clinical Score ≥ 4(c) vs. Combined Score ≥ 4(c)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infertile control (n = 29)</td>
<td>0.04</td>
<td>0.002</td>
<td>0.50</td>
</tr>
<tr>
<td>Stage I Endometriosis (n = 11)</td>
<td>0.06</td>
<td>0.06</td>
<td>d</td>
</tr>
<tr>
<td>Stage II Endometriosis (n = 18)</td>
<td>0.73</td>
<td>0.12</td>
<td>0.62</td>
</tr>
<tr>
<td>Stage III Endometriosis (n = 15)</td>
<td>1.00</td>
<td>1.00</td>
<td>1.00</td>
</tr>
<tr>
<td>Stage IV Endometriosis (n = 13)</td>
<td>1.00</td>
<td>1.00</td>
<td>0.50</td>
</tr>
</tbody>
</table>

HCAII = human carbonic anhydrase.
\(a\) Any reactivity to HCAII is considered positive.
\(b\) A total of 3 or more points from dysmenorrhea, dyspareunia, and pelvic examination is considered positive.
\(c\) Immunoreactivity score (0-3) + clinical score (0-6); a total of 4 or more points is considered positive.
\(d\) Test always agrees; cannot compute.

shown). However, there were several patients with laparoscopically diagnosed endometriosis who would not have been identified as having endometriosis based on clinical symptoms highlighting the pitfalls of diagnosis on clinical criteria alone. The combined score identified a greater number of mild to severe (stages II to IV) endometriosis patients as positive for endometriosis than did either the carbonic anhydrase II score or the clinical score (table I). However, the differences were small, and the number of patients in each group small; these differences were not statistically significant. McNemar's tests comparing the clinical scores with the combined scores gave no indication of differences in positive rates for the two tests for either infertile controls or infertile endometriosis patients (table III). A larger group of patients would be needed to evaluate the comparative performance of clinical scores alone relative to the combined score. However, the sample positive predictive values for the combined score are better than for the carbonic anhydrase II antibody score or the clinical score alone (table IV).

The predictive value of the combined score in diagnosing endometriosis varies by prevalence of disease as illustrated in table IV. Since our practice is a referral center, the prevalence of endometriosis was 59 percent. A positive predictive value of 95 percent for the combined score at this prevalence represents a valuable diagnostic tool. Even in a general gynecology practice where prevalence rates approximate 20 percent, the combined score still has a positive predictive value of 76 percent, which compares favorably to previously published values for CA-125 assays.\(^{14,15,32,33}\) Since minimal disease is felt by many clinicians to be of little significance,\(^{34}\) positive predictive values were also calculated for mild to severe (stages II to IV) endometriosis only. The prevalence of these endometriosis stages in our practice is 43 percent. The positive predictive values for mild to severe endometriosis only are similar to those for all endometriosis stages, and the negative predictive values are improved.
### TABLE IV

Predictive Values of Screening Methods for Endometriosis

<table>
<thead>
<tr>
<th>Patient Groups</th>
<th>Prevalence (%)</th>
<th>Immunoreactivity to HCAII Predictive Value</th>
<th>Clinical Score ≥ 3&lt;sup&gt;e&lt;/sup&gt; Predictive Value</th>
<th>Clinical Score ≥ 4&lt;sup&gt;e&lt;/sup&gt; Predictive Value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Positive (%)</td>
<td>Negative (%)</td>
<td>Positive (%)</td>
</tr>
<tr>
<td>A.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Stage I–IV</td>
<td>20</td>
<td>28</td>
<td>87</td>
<td>54</td>
</tr>
<tr>
<td>Endometriosis</td>
<td>30</td>
<td>40</td>
<td>79</td>
<td>67</td>
</tr>
<tr>
<td></td>
<td>40</td>
<td>51</td>
<td>71</td>
<td>76</td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>61</td>
<td>62</td>
<td>82</td>
</tr>
<tr>
<td></td>
<td>60</td>
<td>70</td>
<td>52</td>
<td>88</td>
</tr>
<tr>
<td>B.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Stage II–IV</td>
<td>20</td>
<td>27</td>
<td>89</td>
<td>62</td>
</tr>
<tr>
<td>Endometriosis</td>
<td>30</td>
<td>39</td>
<td>79</td>
<td>73</td>
</tr>
<tr>
<td></td>
<td>40</td>
<td>50</td>
<td>71</td>
<td>81</td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>60</td>
<td>62</td>
<td>87</td>
</tr>
</tbody>
</table>

HCAII = human carbonic anhydrase.

A = prevalence if all stages of endometriosis considered to be diseased = 59%.

B = Prevalence if only mild to severe (II, III, IV) endometriosis is considered to be diseased = 43%.

b Any reactivity to HCAII is considered positive.

c A total of 3 or more points from dysmenorrhea, dyspareunia, and pelvic examination scores is considered positive.

d Immunoreactivity score (0 – 3) + clinical score (0 – 6); a total of 4 or more points is considered positive.

Clinical decisions regarding endometriosis are further complicated by the lack of reliable data. The medical literature can be difficult to interpret based on recent findings that the classic visual appearance of endometriosis is only one of several possible types. This variation in appearance leads to frequent under-diagnosis of the disease and raises major questions about the validity of studies which have failed to include pathologic criteria in determining prevalence, stage, and response to therapy. Indeed, some studies have shown that visually normal peritoneum may have microscopic disease when routine biopsies are taken from visually normal uterosacral ligaments or random peritoneal biopsies are subjected to electron microscopy. Prior attempts to find a serum marker for endometriosis have generally been centered around measurements of CA-125. Both supportive and critical citations can be found in the literature. This protein has been shown to have statistically significant positive predictive values, but these are difficult to interpret since prevalence data are often not reported. When prevalence information is lacking or unreliable, likelihood ratios provide valuable information. Ratios of <0.1 or >10 are recognized as having clinical utility. Adding HCA-II results to the clinical score improved the likelihood ratio from 5.6 to 18.3 for all stages and from 6.7 to 22.1 for stages II to IV, a significant improvement in clinical utility.
Given the complexity of the disease and variability of clinical presentation, it is unlikely that a single diagnostic test will prove to be a binary indicator of disease. Certainly any biochemical finding is subject to the ultimate test of clinical significance, particularly in a syndrome such as infertility where the observed effect may be the result of any one of multiple contributing factors or any combination thereof. Clinical acumen will logically always factor into the equation that results in decisions about patient care. The use of a simplified, subjective evaluation of pain symptoms for a clinical score is a potential source of bias owing to variable individual pain tolerance and could have conceivably affected the results, but was included because patient perception of pain severity is, in fact, still a frequent indication for surgical investigation. Others have used similar subjective scoring mechanisms in reference to endometriosis patients for similar purposes. It has been shown that the clinical decision of whether and when to consider a diagnostic laparoscopy in an infertility patient to detect the presence of endometriosis may be facilitated by the addition of carbonic anhydrase II antibody to the clinical findings. Moreover, the antibody assay itself, the clinical scores, and the combined scores all show a strong linear by linear relationship with the severity of disease. Inagaki and colleagues reported that anticarbonic anhydrase antibody titers in their systemic lupus erythematosus patients tended to parallel disease activity. They also observed reduced reactivity of the antibody in patients who were being treated with oral corticosteroids. Of interest in this regard are the presumed false negative cases of stage II to IV disease in our study. Nine of the 13 in this category had prior treatment with either surgery or medical suppression. There is evidence that suppression and/or surgical excision/vaporization may reduce the measurement of anti-endometrial antibodies in other assay systems for periods of time up to 9 months. This reduction may account for some of the endometriosis patients failing to be captured by the carbonic anhydrase II assay. In general, patients with significant disease tend to have higher clinical scores and would be identified by using the combined scoring approach. The use of more quantitative assays, such as flow cytometry, would be more useful in longitudinal studies. Such evaluations are currently underway in our laboratory.

Certainly, there are known conditions under which carbonic anhydrase II antibody testing could be misleading if used alone. Prior publications have shown 20 to 40 percent seropositivity in various autoimmune diseases, but by combining the clinical parameters with anticarbonic anhydrase II immunoreactivity, it is our belief that such “false” positivity would have minimal clinical impact (i.e., no patient should have surgery on the basis of a positive test alone without at least some coexisting supportive symptoms). Of the patients in this study who could be presumed to be “falsely” positive by the antibody test alone, i.e., the infertile control patients, 4 of the 11 had significant peritoneal adhesions found at surgery that were unsuspected by hysterosalpingogram. One additional patient was highly suspicious for adenomyosis. Another reason for the high rate of reactivity in what seem to be infertility patients without endometriosis may be the fact that some patients may have developed a mild form of the disease in the interval between surgery and assay; alternatively, they may have been underdiagnosed at the time of their surgery owing to lack of recognition of atypical forms of the disease by a less experienced laparoscopist. Equally plausible is the possibility of microscopic or minimal disease which could go undetected even in experienced hands.
By virtue of its presence in preovulatory follicles, fallopian tubes, and endometrium, carbonic anhydrase may impact reproductive processes from the moment of ovulation through nidation. Why endometriosis patients form autoantibodies to carbonic anhydrase is, at this point, unknown, although peritoneal exposure to subcellular endometrial fragments or blood breakdown products may be hypothesized. Whether or not the subsequent autoimmune derangement alters fertility through effects on sperm function, embryonic development, or implantation awaits future investigation.

Acknowledgement

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References


