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Prognostic Assessment of Fertility Potential in Women with Endometriosis

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Abstract

Aim. To identify and evaluate clinical, hormonal, biochemical, and molecular markers that can predict fertility potential and reproductive outcomes in women with endometriosis.

Materials and Methods. The study included 58 women aged 25–48 years with external genital endometriosis and 20 healthy fertile controls. Diagnosis was based on clinical assessment, ultrasound, and, when available, intraoperative findings. Clinical evaluation included reproductive history, infertility duration, and pain severity assessed by VAS. Hormonal analysis included AMH, FSH, LH, and estradiol levels. Oxidative stress markers (MDA, SOD, TAC), cytokines (IL-6, TNF- α , IL-10), apoptosis-related proteins (Bax, Bcl-2, caspase-3), and microRNA expression (miR-21, miR-200, miR-135a/b) were assessed using ELISA and qRT-PCR. Disease severity was classified using rASRM, Enzian, AAGL systems, and reproductive prognosis was evaluated using the Endometriosis Fertility Index (EFI). Statistical analysis included comparative tests, correlation analysis, logistic regression, and ROC analysis.

Results. Infertility was observed in 79.3% of women with endometriosis, with a mean duration of 4.3 ± 1.7 years. AMH levels were significantly lower compared with controls (1.9 ± 0.7 vs 2.8 ± 0.9 ng/mL, $p < 0.01$), indicating reduced ovarian reserve. Increased oxidative stress was confirmed by elevated MDA and decreased antioxidant parameters (SOD, TAC).

Pro-inflammatory cytokines (IL-6, TNF- α) were significantly elevated, while IL-10 levels were reduced. Molecular analysis showed increased miR-21 and miR-135a/b expression and decreased miR-200 levels, associated with disease severity and infertility. Moderate and severe endometriosis accounted for over 60% of cases. EFI scores indicated low reproductive prognosis in 19.0%, moderate in 43.1%, and favorable in 37.9% of patients. Logistic regression identified reduced AMH, elevated IL-6, increased MDA, and altered microRNA expression as key predictors of unfavorable reproductive outcomes. ROC analysis demonstrated good predictive performance of the integrated model.

Conclusion. Endometriosis-associated infertility is determined not only by anatomical disease extent but also by systemic inflammation, oxidative stress, reduced ovarian reserve, and dysregulated microRNA expression. Integration of clinical, hormonal, and molecular markers significantly improves prognostic assessment and supports personalized reproductive management in women with endometriosis.

Key words: endometriosis; infertility; ovarian reserve; microRNA; oxidative stress

Endometriosis is a chronic, estrogen-dependent inflammatory disease characterized by the presence of endometrial-like tissue outside the uterine cavity. It affects approximately 10–15% of women of reproductive age and is strongly associated with chronic pelvic pain, dysmenorrhea, and infertility [1, 2]. According to current epidemiological data, up to 30–50% of women diagnosed with endometriosis experience difficulties conceiving [1, 3]. Despite significant progress in the understanding of the pathophysiology of the disease, predicting reproductive outcomes in women with endometriosis remains a complex clinical challenge.

The mechanisms through which endometriosis impairs fertility are multifactorial and include anatomical distortion of pelvic organs, inflammatory alterations within the peritoneal environment, impaired oocyte quality, reduced ovarian reserve, and disruptions in endometrial receptivity [1, 4]. In addition, surgical treatment of endometriotic lesions, particularly ovarian endometriomas, may further influence reproductive potential by affecting ovarian tissue and follicular reserve [1, 5]. These factors make individualized prognostic assessment particularly important for optimizing reproductive management strategies.

In clinical practice, various diagnostic and prognostic tools are used to evaluate fertility potential in women with endometriosis. These include assessment of ovarian reserve markers such as anti-Müllerian hormone (AMH), antral follicle count (AFC), imaging

methods for evaluating pelvic anatomy, and classification systems such as the revised American Society for Reproductive Medicine (rASRM) staging and the Endometriosis Fertility Index (EFI) [6-8]. However, the predictive value of these indicators varies, and their integration into a comprehensive prognostic framework remains an area of ongoing research [6, 9]. Therefore, a better understanding of factors influencing reproductive outcomes in endometriosis is essential for improving fertility counseling, selecting appropriate treatment strategies, and optimizing assisted reproductive technologies.

Aim: To identify and evaluate clinical, hormonal, and imaging markers that can predict fertility potential and reproductive outcomes in women with endometriosis

Materials and Methods. The study included 58 women aged 25–48 years diagnosed with external genital endometriosis who were examined and treated at the gynecological department of the University Clinic of Odesa National Medical University during 2024–2025. The diagnosis of endometriosis was established based on clinical manifestations, pelvic ultrasound findings, and, when available, intraoperative confirmation. The control group consisted of 20 healthy fertile women of comparable age without clinical or ultrasound signs of gynecological pathology.

Clinical evaluation included assessment of reproductive history, duration of infertility, and presence of pelvic pain. The severity of pain was evaluated using a visual analog scale (VAS) [9]. Ovarian reserve was assessed by measuring serum anti-Müllerian hormone (AMH), follicle-stimulating hormone (FSH), luteinizing hormone (LH), and estradiol (E2) levels using chemiluminescent immunoassay [10].

Molecular and biochemical analyses included evaluation of oxidative stress markers (malondialdehyde (MDA), superoxide dismutase (SOD), and total antioxidant capacity, TAC) [11], cytokine profile (IL-6, TNF- α , IL-10) [12], and apoptosis-related proteins (Bax, Bcl-2, caspase-3) [13] determined by enzyme-linked immunosorbent assay (ELISA). Expression of microRNAs (miR-21, miR-200, and miR-135a/b) in peripheral blood samples was analyzed using quantitative real-time polymerase chain reaction (qRT-PCR) [14].

Disease severity and reproductive prognosis were assessed using the revised American Society for Reproductive Medicine (rASRM) classification [6], the Enzian [15] and AAGL [16] staging systems, and the Endometriosis Fertility Index (EFI) [8].

The study was conducted in accordance with the ethical principles for medical research involving human subjects as outlined in the World Medical Association Declaration of Helsinki (2024 revision) and complied with applicable national regulations and institutional policies on biomedical research, data protection, and patient privacy. The study

protocol, including procedures for data collection and handling of clinical and reproductive health information, was reviewed and approved by the local Institutional Review Board / Independent Ethics Committee prior to initiation. Written informed consent was obtained from all participants before enrollment; for retrospective components involving previously collected, de-identified clinical data or biological samples, a waiver of consent was granted where permitted by law and justified by minimal risk and impracticability of re-contact [17].

Statistical analysis was performed using Statistica 13.0 software [18]. Normality of distribution was assessed using the Shapiro–Wilk test. Depending on data distribution, results were presented as mean $\pm$ standard error or median with interquartile range. Comparative analysis was performed using Student's t-test or the Mann–Whitney U test, while correlations were evaluated using Pearson or Spearman coefficients. Logistic regression and ROC analysis were applied to assess the predictive value of clinical and molecular markers for reproductive outcomes.

Results.

The mean age of the examined women with endometriosis was 36.9 ± 4.8 years and did not significantly differ from that of the control group (36.2 ± 5.1 years, $p > 0.05$). Infertility was observed in 46 of the 58 patients (79.3%), while 12 women (20.7%) had preserved fertility. Among patients with infertility, primary infertility was diagnosed in 26 cases (56.5%) and secondary infertility in 20 cases (43.5%). The mean duration of infertility was 4.3 ± 1.7 years. Clinical manifestations of endometriosis included dysmenorrhea in 72.4% of patients, chronic pelvic pain in 51.7%, and dyspareunia in 36.2%. The average intensity of pelvic pain according to the visual analog scale was 6.7 ± 1.3 points.

Assessment of ovarian reserve demonstrated significantly lower AMH levels in women with endometriosis compared with the control group (1.9 ± 0.7 ng/mL vs 2.8 ± 0.9 ng/mL, $p < 0.01$). In a subgroup of patients with infertility, AMH values were even lower (1.6 ± 0.6 ng/mL), indicating a reduction in ovarian reserve. Mild elevation of FSH levels was observed in some patients with long-standing infertility, while LH and estradiol concentrations remained within age-related reference ranges.

Analysis of oxidative stress markers revealed increased lipid peroxidation activity in patients with endometriosis. The mean concentration of malondialdehyde (MDA) reached 4.8 ± 0.9 nmol/mL compared with 2.9 ± 0.7 nmol/mL in the control group ($p < 0.01$). At the same time, antioxidant defense parameters were decreased, with reduced superoxide dismutase activity (1.5 ± 0.4 U/mL vs 2.2 ± 0.5 U/mL) and lower total antioxidant capacity (0.89 ± 0.14 mmol/L vs 1.24 ± 0.18 mmol/L).

Evaluation of the cytokine profile demonstrated elevated levels of pro-inflammatory cytokines. Serum IL-6 and TNF- α concentrations were significantly higher in the endometriosis group (9.2 ± 2.5 pg/mL and 12.4 ± 3.1 pg/mL, respectively) compared with healthy women (3.8 ± 1.2 pg/mL and 6.0 ± 1.7 pg/mL). In contrast, the anti-inflammatory cytokine IL-10 tended to be lower in patients with endometriosis.

Molecular analysis revealed significant differences in microRNA expression. Increased expression of miR-21 was predominantly observed in patients with pronounced pelvic pain and inflammatory activity. Reduced levels of miR-200 were associated with more severe disease forms and decreased reproductive potential. Elevated expression of miR-135a/b was most characteristic of patients with infertility and showed a negative correlation with AMH levels.

According to the rASRM classification, stage I endometriosis was identified in 7 patients (12.1%), stage II in 14 (24.1%), stage III in 22 (37.9%), and stage IV in 15 women (25.9%). Thus, moderate and severe forms of the disease accounted for more than 60% of cases. Evaluation using the Endometriosis Fertility Index demonstrated a mean EFI score of 5.7 ± 1.9 . Low reproductive prognosis (EFI 0–3) was observed in 11 patients (19.0%), moderate prognosis (EFI 4–6) in 25 patients (43.1%), and favorable prognosis (EFI 7–10) in 22 women (37.9%).

Correlation analysis revealed significant relationships between several clinical and molecular parameters. Increased miR-21 expression correlated with IL-6 levels ($r = 0.51$) and pain severity ($r = 0.47$), whereas miR-135a/b showed a positive correlation with infertility duration ($r = 0.44$) and a negative correlation with AMH levels ($r = -0.42$). Reduced miR-200 expression was associated with higher oxidative stress indicators.

Discussion. The mean age of the examined women with endometriosis was 36.9 ± 4.8 years and did not significantly differ from that of the control group (36.2 ± 5.1 years, $p > 0.05$). Infertility was observed in 46 of the 58 patients (79.3%), while 12 women (20.7%) had preserved fertility. Among patients with infertility, primary infertility was diagnosed in 26 cases (56.5%) and secondary infertility in 20 cases (43.5%). The mean duration of infertility was 4.3 ± 1.7 years. Clinical manifestations of endometriosis included dysmenorrhea in 72.4% of patients, chronic pelvic pain in 51.7%, and dyspareunia in 36.2%. The average intensity of pelvic pain according to the visual analog scale was 6.7 ± 1.3 points.

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Logistic regression analysis demonstrated that a combination of reduced AMH, elevated IL-6, increased MDA, and altered expression of miR-21 and miR-135a/b had the

highest predictive value for identifying patients with an unfavorable reproductive prognosis. ROC analysis confirmed good diagnostic performance of the integrated model, indicating its potential usefulness for prognostic assessment of fertility potential in women with endometriosis.

Conclusion. Endometriosis-related infertility is associated not only with the anatomical extent of the disease but also with systemic inflammatory activity, oxidative stress, reduced ovarian reserve, and altered microRNA expression. The study demonstrated that different clinical phenotypes of endometriosis show distinct biological profiles that may influence reproductive potential and disease progression. Integrating clinical parameters with molecular markers such as AMH, inflammatory cytokines, oxidative stress indicators, and microRNA expression may improve prognostic assessment and support more individualized reproductive management in women with endometriosis.

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