Review

Evaluating the Prognostic Potential of CA-125 and miRNA Levels in Endometriosis: A Narrative Review

Taha Shokrnejad-Namin *, Morteza Amoozgar, Amirali Hariri, Elnaz Amini

Posted Date: 19 April 2024

doi: 10.20944/preprints202404.1261.v1

Keywords: endometriosis; CA-125; miRNA; gynecology; prognosis; diagnosis; management

 doi:10.20944/preprints202404.1261.v1

Review

Evaluating the Prognostic Potential of CA-125 and miRNA Levels in Endometriosis: A Narrative Review

Taha Shokrnejad-Namin [4]*, Elnaz Amini [1], Morteza Amoozgar [2], and Amirali Hariri [3]

[1] MD Researcher, Medical Advisor, March Health Company, Concord, CA 94521, USA.
[2] CEO, March Health Company, Concord, CA 94521, USA.
[3] Research and Development, March Health Company, Concord, CA 94521, USA.
[4] MD Researcher, Research and Development, March Health Company, Concord, CA 94521, USA.
* Correspondence: Taha Shokrnejad-namin (Shokrnejadtaha@gmail.com)

Abstract: Objective: This study aims to examine CA-125 and miRNAs as biomarkers in endometriosis pathophysiology and their potential roles in clinical diagnostics and prognostics. **Methods:**We reviewed a variety of studies, including cross-sectional, case-control, and prospective designs, examining a broad spectrum of patient demographics, clinical features, and biomarker (CA-125 and miRNA) expression levels. **Results:** CA-125 has been reaffirmed as a valuable biomarker in diagnosing and monitoring endometriosis, with its utility enhanced when used alongside other markers. MiRNAs have emerged as promising molecular regulators, offering new avenues for non-invasive diagnostics and a deeper understanding of the disease's pathophysiology. The integration of these biomarkers presents a potential shift towards more personalized and effective diagnostic and therapeutic strategies. **Conclusion:** Endometriosis presents a significant challenge in women's health, requiring innovative approaches for its management. The convergence of CA-125 and miRNA research represents a promising advance, potentially leading to more accurate diagnostics and personalized treatment. Future research should focus on standardizing methodologies, expanding study cohorts, and integrating findings into clinical practice to fully harness the potential of these biomarkers.

Keywords: endometriosis; CA-125; miRNA; gynecology; prognosis; diagnosis; management

1.1. Introduction

Endometriosis, a common, and complex gynecological disorder, continues to pose a challenge to clinicians and researchers due to its detrimental effects on women's health and quality of life. It is estimated that up to 10% of people assigned female at birth suffer from endometriosis [1,2]. For diagnosing endometriosis accurately, laparoscopy remains the preferred method, followed by transvaginal ultrasound and pelvic magnetic resonance imaging. Up to 50% of women with endometriosis diagnosed during laparoscopy do not have any symptoms at all, emphasizing the condition's challenge. Symptoms of endometriosis can be cyclical or chronic, non-specific, and can mimic those of other gynecological, gastrointestinal, and musculoskeletal conditions[3]. It can be difficult to diagnose endometriosis, given its variety of symptoms. Many studies have documented a time lag between symptom development and diagnosis[4,5]. Menstrual symptoms may be dismissed by both health professionals and patients, causing diagnostic delays and preventing therapy. In the absence of treatment, endometriosis symptoms can result in reduced physical and psychological performance, loss of quality sleep, diminished sexual function, and perceived stress[6]. All of these factors can contribute to a significant drop in quality of life[7,8]. Early detection and treatment of endometriosis can prevent disease progression, adhesion formation, related infertility, and central pain sensitivity, which can all lead to chronic pelvic pain[9]. An accurate diagnosis offers many benefits to people, according to qualitative research, including the validation of their symptoms, the opportunity to discuss their medical condition, the assurance that their symptoms are not associated with cancer, as well as the opportunity for management techniques[10].

The disease is still poorly understood, and its diagnosis and treatment remain difficult for physicians, despite its prevalence and significance for women's health. Biomarkers that are reliable,

 doi:10.20944/preprints202404.1261.v1

minimally invasive, and can aid in early diagnosis, prognosis, and personalized treatment strategies are therefore needed[11]. Hormones, cytokines, glycoproteins, and angiogenic factors are involved in the chronic inflammatory process of endometriosis. Some of these components may serve as biomarkers for endometriosis, and some are connected to the disease's pathogenesis[11,12]. Many blood markers have been studied over the past decades, including those related to apoptosis, cell adhesion molecules, matrix-related proteins, cytoskeleton molecules, nerve growth factors, oxidative stress indicators, tumor markers, and several peptides/proteins that contribute to endometriosis[13]. CA-125 and miRNA are emerging as promising biomarkers due to their roles in cellular processes[13,14].

Due to its possible association with disease severity and recurrence, CA-125 has gained interest in endometriosis research. A higher CA-125 level has been reported in women with endometriosis[15]. In addition to CA-125, microRNAs (miRNAs) play a role in gene regulation. Studies have found that miRNAs derived from damaged tissues and bodily fluids can detect a variety of diseases. Recent advances in sequencing and microarray technologies have allowed researchers to examine the amounts of miRNAs and long non-coding RNAs in the system. Numerous studies have shown altered miRNA expression patterns in endometriosis tissues and blood[16–20].

CA-125 and miRNA's prognostic value in endometriosis remains scattered and heterogeneous. Clinical translation of these potential biomarkers is hindered by variable methodologies, divergent findings, and limited consensus[15]. It is imperative to conduct a comprehensive literature review and critical analysis to uncover the true prognostic significance of CA-125 and miRNA in predicting disease severity, progression, and recurrence.

The aim of this narrative review is to bridge the gap by assessing and consolidating the available evidence regarding the prognostic value of CA-125 and miRNA in endometriosis.

2.1. Results

Research has shown that miRNA, small RNA molecules, regulate crucial factors within the endometriotic microenvironment[21]. These studies display the molecular complexity underlying disease progression and severity. In addition, the traditional biomarker CA-125 has emerged as a potential diagnostic tool for endometriosis[22]. The correlation between CA-125 levels and clinicopathological characteristics offers a new approach to categorizing and managing the disease[23]. By integrating miRNA expression profiles with CA-125 insights, we tried to offer a deeper understanding of endometriosis and its clinical spectrum. In this review, we examine the potential synergistic relationship between miRNAs and CA-125, providing valuable implications for patient diagnosis and treatment.

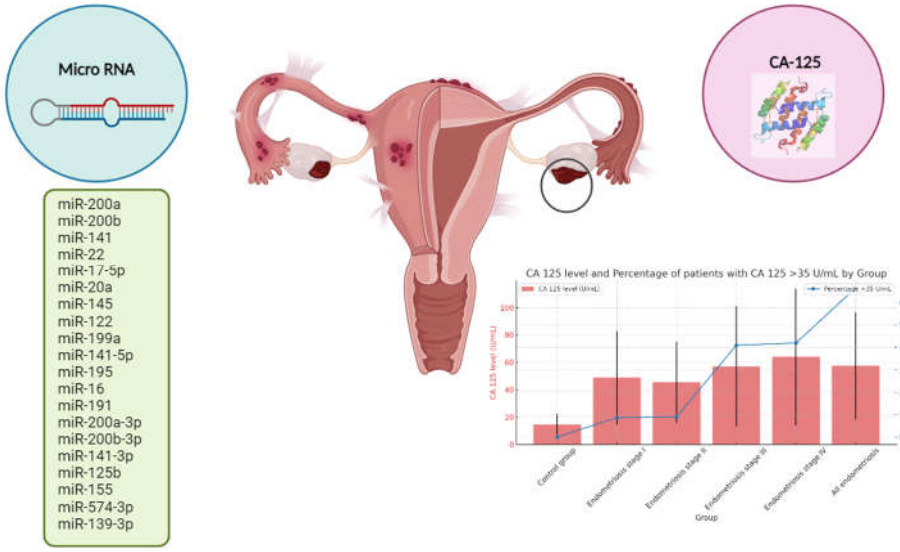

Figure 1. Graphical Abstract.

 doi:10.20944/preprints202404.1261.v1

2.2. CA-125 in Endometriosis

In the following document, we will categorize the different potentials of CA-125 in endometriosis by dividing the studies into four categories: Diagnosis, Cutoff Value, Staging, and Management.

2.2.1. Pathogenesis and Diagnosis

CA-125's role in endometriosis pathogenesis and diagnosis has been widely studied, leading to a detailed understanding of its capabilities and limits.

In 1996, Colacurci et al. investigated CA-125's diagnostic potential for endometriosis, comparing serum and peritoneal tests. They found peritoneal fluid CA-125 levels were consistently elevated in endometriosis cases, whereas serum CA-125 increased mainly in advanced stages. They deduced that peritoneal CA-125 was superior in detecting early-stage endometriosis than its serum counterpart[24].

Kafali et al. 2004 examined the variations in serum CA-125 levels throughout the menstrual cycle to diagnose endometriosis. Notably, endometriosis patients saw a 198.3% surge in CA-125 during menstruation, compared to a 22% rise in healthy women. With a threshold of an 83% CA-125 increment during menstruation, they achieved a 93% sensitivity and 92% specificity for the diagnosis[25].

Agic et al.'s 2008, research evaluated a combined test for endometriosis using CCR1 miRNA in blood leukocytes, and serum MCP-1 and CA-125 measurements. Elevated CCR1/HPRT miRNA ratios, MCP-1, and CA-125 levels were observed in endometriosis patients. The combined test had high sensitivity (92.2%) and specificity (81.6%), outperforming CA-125 used alone (sensitivity: 27.5%, specificity: 100%), suggesting a more comprehensive diagnostic approach for endometriosis[26].

Mihalyi et al.2010 investigated the diagnostic value of CA-125 in 294 patients with infertility (203 with endometriosis). They showed elevated levels of IL-6, IL-8, and CA-125 in all women with endometriosis, particularly those with moderate-severe forms. Further analyses, demonstrated high sensitivity and specificity in diagnosing both minimal-mild and moderate-severe endometriosis. Notably, during the secretory phase, the panel of biomarkers achieved 100% sensitivity and 84% specificity for moderate-severe endometriosis. The study underscores the potential of this biomarker panel, including CA-125, for non-invasive diagnosis[27].

The study by Socolov et al. 2011 aimed to assess selected serum markers' diagnostic and severity-assessment potential for endometriosis. CA-125 levels were elevated in 54% of endometriosis patients, showing diagnostic potential with 54% sensitivity and 91% specificity. IL-6 levels did not exhibit significant discrimination (71% sensitivity, 12% specificity). CA-125 correlated with endometriosis severity based on revised American Fertility Society (rAFS) scores ($p = 0.03$), but IL-6 did not. Other tested markers lacked discriminatory value[28].

In a 2012 study, Vodolazkaia and colleagues aimed to devise a non-invasive diagnostic method for symptomatic endometriosis patients without ultrasound (US) indications. Two models emerged from 353 EDTA plasma samples: Model 1 included annexin V, VEGF, CA-125, and glycodelin; Model 2 included annexin V, VEGF, CA-125, and sICAM-1. Both models were highly sensitive and specific in a training dataset, with 81-90% sensitivity and 68-81% specificity. When tested independently, they maintained an 82% sensitivity and 63-75% specificity. It appears that the models are most effective during menstruation. In symptomatic patients lacking US evidence, these models could be a noninvasive diagnostic option[29].

Zhu et al. 2016 examined the predictive potential of serum markers CA-125 and CA 19-9 alongside pain scores for pelvic endometriosis in infertile women. Notable differences were found in serum levels of both markers. Combining CA-125, CA 19-9, and pain scores improved prediction accuracy for pelvic endometriosis[30].

Oliveira et al. 2017 studied CA-125's diagnostic utility for deep infiltrating endometriosis (DIE) during menstrual and mid-cycle phases. Best diagnostic performance was seen in CA-125 levels during the menstrual phase and the difference between menstrual and mid-cycle phases (AUC of

 doi:10.20944/preprints202404.1261.v1

0.96). The study highlighted CA-125 as a promising diagnostic tool for DIE when measured during both phases[22].

Irungu et al. 2019 conducted a study to identify biomarkers for endometriosis diagnosis. They analyzed proteomics profiles of eutopic and ectopic endometrial tissues from confirmed cases and controls, aiming to validate previously reported biomarkers in serum samples. CA-125 emerged as the top single marker (AUC = 0.63), but combining markers improved accuracy (AUC = 0.71-0.81), especially in different menstrual cycle phases and control groups. Results unveiled menstrual cycle-linked protein changes in endometriosis, enhancing understanding of the condition. While individual markers had limited utility, combined markers showed promise for patient stratification[31].

Tang et al. 2021 aimed to develop a noninvasive diagnostic method for ovarian endometriosis using blood indicators. Study showed tumor markers including AFP, CA-125, CA199, and HE4 contributed to diagnosis, while CA-125, HE4, and CYFRA 21-1 differentiated disease stages. The study highlighted significant differences in serological indicators between patients and healthy controls, underscoring their diagnostic and staging potential[32].

Guralp et al. 2021 explored a quadruple panel of serum markers (CA-125, endocan, YKL-40, and copeptin) for diagnosing endometriosis severity. These markers were significantly elevated in endometriosis cases. The combined panel score had a sensitivity of 96.5% and specificity of 84.6% for moderate-severe endometriosis. The study also proposed a point algorithm for potentially excluding endometriosis[33].

Chen et al. 2021 examined differences in blood cells and tumor biomarkers between endometriosis patients and controls. They found notable discrepancies in blood cell counts and tumor markers, with endometriosis patients having altered levels of eosinophil, neutrophil count, and others. A diagnostic model using HGB, CA199, CA-125, and HE4 showed a sensitivity of 85.4%, specificity of 78.83%, and an AUC of 0.900, suggesting enhanced diagnostic accuracy for early endometriosis detection[34].

Szubert et al. 2023 aimed to create a non-invasive endometriosis diagnostic algorithm. They pinpointed 7 key features, including painful periods, CA-125 levels, and BMI, as strong predictors. Their algorithm boasted a sensitivity of 0.88 and specificity of 0.80, emphasizing the potential of using accessible features for diagnosis[35].

In a 2023 study by Micu et al., the diagnostic potential of serum biomarkers CA-125, HE4, and CA72-4 in ovarian endometriosis (OvEndo) was explored. OvEndo patients exhibited elevated CA-125 (9.02 U/mL) and CA72-4 (6.1 U/mL) levels compared to controls, while HE4 levels were surprisingly lower in OvEndo (7.6 ng/mL). A strong correlation between CA72-4 and HE4, but not CA-125, was found. The findings suggest CA-125 and HE4, especially when combined, as important diagnostic markers for ovarian endometriosis, with the CA72-4 and HE4 relationship warranting further investigation[36].

Kovalak et al. 2023 investigated novel biomarkers for diagnosing and treating stage III-IV endometriosis using a case-control design. ANXA5, sICAM-1, IL-6, TNF-α, VCAM-1, VEGF, and Ca-125 were evaluated. ANXA5, sICAM-1, IL-6, TNF-α, and VCAM-1 had nonsignificant AUCs for individual diagnosis ($p > 0.05$). However, Ca-125 had a significant AUC (73% sensitivity, 98% specificity, $p < 0.001$). When combined with ANXA5, diagnosis sensitivity remained at 73%, with improved specificity of 100%. The study concluded that the combined use of Ca-125 and ANXA5 enhanced diagnostic accuracy for stage III-IV endometriosis compared to Ca-125 alone[37].

In sum, Using CA-125 in combination with other biomarkers could provide a diagnostic tool for endometriosis. Peritoneal fluid levels show promise for early detection, even when serum levels indicate advanced stages. Endometriosis and healthy individuals can be differentiated by the fluctuations of the marker during the menstrual cycle. The diagnostic accuracy of CA-125 can be enhanced by combining it with other markers such as CCR1 miRNA, MCP-1, and IL-6. It is intended to improve non-invasive diagnostic methods by exploring additional biomarkers and examining the role that CA-125 plays in these biomarkers.

 doi:10.20944/preprints202404.1261.v1

Table 1. Studies related to diagnosis of endometriosis by CA-125.

Year	Main objective	Type of study Size of sample Diagnosis method	Biomarkers	Main result	Main outcome	Ref
1996	To evaluate the clinical utility of CA-125 in the diagnosis of endometriosis and to compare the sensitivity of the serum and the peritoneal test as an indicator of disease.	Case control 26 Laparoscopy	Serum and peritoneal CA-125	CA-125 levels in peritoneal fluid were higher than those found in serum and were significantly elevated ($P < 0.05$)	Levels of CA-125 in peritoneal fluid seem to be a more sensitive indicator of disease than serum levels (0.86 vs. 0.36), especially in early stage endometriosis (0.80 vs. 0.20) which tends to be overlooked by the CA-125 serum test.	[24]
2004	To elucidate whether endometriosis can be diagnosed clinically by assessing the differences between serum CA-125 levels during menstruation and during the rest of the menstrual cycle.	Case control 28 Laparoscopy	CA-125	The mean CA-125 concentrations of healthy women during menstruation and during the rest of the menstrual cycle were 12.2 and 10 U ml.	Assessment of changes in serum CA-125 levels during the menstrual cycle may be useful in the diagnosis of endometriosis.	[25]
2008	Investigated the possible use of CCR1 miRNA measurement in peripheral blood leukocytes with monocyte chemotactic protein-1 (MCP-1) and CA-125 protein in serum as a diagnostic test for endometriosis.	Case control (Retrospective) 151 Laparoscopy	CCR1, HRPT, MCP1, CA-125	The ratio of CCR1/HPRT miRNA in peripheral blood of patients with endometriosis and adenomyosis was significantly elevated compared with women without endometriosis. Additionally, serum levels of MCP-1 and CA-125 were significantly higher in patients with endometriosis.	Found increased levels of CA-125 in peripheral blood of patients with endometriosis compared with healthy controls. Nevertheless, CA-125 alone was above the threshold in only 28 out of 102 endometriosis patients (sensitivity 27.5%). Considering only CA-125, none of the controls showed false positive results (specificity 100%).	[26]
2010	To evaluate the combined performance of six potential plasma biomarkers in the diagnosis of endometriosis.	Case control 294 Laparoscopy	CA-125, IL-6, IL-8, TNF-α, CA-19-9, hsCRP	Increased plasma levels: IL-6, IL-8, CA-125, Decreased plasma level: TNF-α	Results show that multivariate methods such as logistic regression and LSSVMs in general perform better than single protein models, suggesting that more than one protein is necessary to predict the	[27]

 doi:10.20944/preprints202404.1261.v1

					presence of endometriosis.	
2011	Analyzed selected well-known and less well-known serum markers that have been proposed for diagnosis and severity assessment of endometriosis.	Case control 48 Laparoscopy	CA-125, IL6	CA-125 levels were over the cut-off of 35 IU/l in 54% of patients (versus 8% of controls), averaging 67.5 (CI95: ±17.5). The sensitivity and specificity were 54% and 91%, respectively, with a p value of <0.001 (statistically significant).	CA-125 correlated with endometriosis screening and severity, indicating its superiority as a marker for further, larger studies.	[28]
2012	To develop and validate a non-invasive diagnostic test with a high sensitivity (80% or more).	Case control 353 Laparoscopy	CA-125, CA 19–9, IL-1beta, IL-6, IL-8, IL-17, IL-21, RANTES, TNF-alpha, IFN-gamma, MCP-1, MIF, CRP, OPN, IL-4, IL-10, annexin V, sICAM-1, VCAM-1, VEGF, NGF, FGF-2, Leptin, IGFBP-3, glycodelin (PP-14), M-CSF, HGF	Increased plasma levels: IGFBP-3, CA-125, CA 19-9 and glycodelin, Decreased plasma level: IL-1beta, IFN-γ, TNF-alpha, Leptin and sICAM-1	In plasma samples obtained during menstruation, multivariate analysis of four biomarkers (annexin V, VEGF, CA-125 and sICAM-1/or glycodelin) enabled the diagnosis of endometriosis undetectable by US with a sensitivity of 81–90% and a specificity of 63–81% in independent training- and test data set.	[29]
2016	To define the utility of serum carcinogenic antigen CA-125 and CA 19-9 combining pain score in the prediction of pelvic endometriosis in infertile women.	Case control 294 Laparoscopy	CA-125, CA 19-9	Preoperative serum CA-125 and CA 19-9 levels were significantly different between the two groups.	Preoperative CA-125 and CA 19-9 levels combining pain score can be useful for the prediction of pelvic endometriosis and may be included in the evaluation of unexplained infertile women.	[30]
2017	To evaluate the performance of CA-125 measurement in the menstrual and mid-cycle phases of the cycle, as well as the difference in its levels between the two phases, for the early diagnosis of DIE.	Case control (perspective) 54 Laparoscopy	CA-125	Area Under the Curve (AUC) of CA-125 in menstrual phase and of the difference between menstrual and midcycle phases had the best performance (both with AUC = 0.96), followed by CA-125 in the midcycle (AUC = 0.89).	CA-125 may be useful for the diagnosis of deep endometriosis, especially when both are collected during menstruation and in mid cycle.	[22]
2019	To identify potential biomarkers through	Case control (Retrospective) 46	CA-125, sICAM1, FST, VEGF, MCP1,	The best single marker for discriminating endometriosis from	Data indicate that the markers tested, whilst not useful alone, have	[31]

 doi:10.20944/preprints202404.1261.v1

	proteomics profiling of eutopic and ectopic endometrial tissue specimens.	Laparoscopy	MIF, IL1R2, LUM, CPM, TNC, TPM2, PAEP	controls remained CA-125 (AUC = 0.63)	improved diagnostic accuracy when used in combination and demonstrate menstrual cycle specificity.	
2021	To develop a noninvasive diagnostic method for endometriosis.	Case control 293 Laparoscopy	CA-125, CA199, HE4, CYFRA 21-1, TNF-α, IL6, sflt-1, MCP-1	In tumor markers, alpha fetoprotein (AFP), carcinoembryonic antigen (CA) 125, CA199 and human epididymis protein 4 (HE4) helped to diagnose endometriosis; CA-125, HE4, and cytokeratin 19 fragment (CYFRA 21-1) could differentiate stages.	Serological indicators in ovarian endometriosis patients were different from healthy women, which were of certain differential values in diagnosis and disease staging.	[32]
2021	To evaluate the diagnostic value of a quadruple panel of serum markers CA-125, endocan, YKL-40 and copeptin, for the prediction of endometriosis.	Case control 140 Laparoscopy	CA-125, endocan, YKL-40, copeptin	Serum CA-125, endocan, copeptin and YKL-40 levels were significantly increased.	YKL-40, endocan and copeptin levels were significantly increased in the moderate-severe endometriosis group compared to the mild-moderate endometriosis group and CA-125 levels remained nonsignificant. A quadruple panel score (CA-125, endocan, YKL-4 and copeptin) had an AUC of 0.954, a sensitivity of 96.5% and specificity of 84.6% for prediction of moderate to severe endometriosis.	[33]
2021	To analyze the differences in the peripheral blood cells and tumor biomarkers between the patients with endometriosis and healthy people.	Case control (Retrospective) 274 Laparoscopy	CA-125, CA199, HE4, HGB	The ROC curve showed that the combined diagnostic model reached a sensitivity of 85.4%, a specificity of 78.83%, and an area under the curve of 0.900, which was significantly higher than that of the individual index in endometriosis diagnosis.	The combined diagnostic model of HGB, CA199, CA-125, and HE4 may provide a new approach for the early non-invasive diagnosis of endometriosis.	[34]
2023	To find an algorithm based on symptoms and laboratory tests that could diagnose	Retrospective analysis 101 Laparoscopy	CA-125, VEGF	The strongest impact on endometriosis prediction had information about painful periods, CA-125 over 15 u/mL, and the lowest BMI, with a	An algorithm based on three easy features, including painful menses, BMI level, and CA-125 concentration could have an	[35]

 doi:10.20944/preprints202404.1261.v1

	endometriosis in a non-invasive way.			sensitivity of 0.8800 and a specificity of 0.8000, respectively.	important place in the non-invasive diagnosis of endometriosis. If confirmed in a prospective study, implementing such an algorithm in populations with a high risk of endometriosis will allow us to cover patients suspected of endometriosis with proper treatment.	
2023	To evaluate the prognostic value as diagnosis makers of cancer antigen (CA)125, human epididymis 4 (HE4), and CA72-4 serum levels in ovarian endometriosis.	Case control 55 Laparoscopy	CA-125, CA72-4, HE4	(i) For CA-125, a statistically significant difference in-between the mean serum levels of the two groups: 9.02 U/mL in the OvEndo group versus 7.1 U/mL in the CTR group (p=0.0158). (ii) For CA72-4 levels in the OvEndo group, where the mean serum level was 6.1 U/mL compared to 3.5 U/mL in the CTR group, (p=0.0185). (iii) The mean serum level of HE4 in the OvEndo group was 7.6 ng/mL versus 7.8 ng/mL in the CTR group, found highly significant (p=0.0001). HE4 levels were highly correlated with CA72-4 levels (p<0.0001), while CA-125 levels were not correlated with HE4 and CA72-4.	Measurements of CA-125 can be used in the diagnosis of OvEndo mainly in association with HE4 serum levels, which are lower in endometriosis patients.	[36]
2023	To investigate the efficacy of new endometriosis biomarkers in diagnosis and treatment.	Case control (Cohort Retrospective) 79 Laparoscopy	Annexin A5 (ANXA5), soluble intercellular adhesion molecule-1 (sICAM-1), interleukin-6 (IL-6), tumor necrosis factor-α (TNF-α), soluble vascular cell adhesion molecule-1 (sVCAM-1),	Only the AUC of the Ca-125 biomarker values were found to be significant with 73% sensitivity and 98% specificity ($p < 0.001$). However, when Ca-125 and ANXA5 were evaluated together, it was concluded that the diagnosis of endometriosis could be made with 73% sensitivity and 100% specificity.	When Ca-125 and ANXA5 are evaluated together, it seems to be more valuable than Ca-125 alone in diagnosing endometriosis.	[37]

 doi:10.20944/preprints202404.1261.v1

				vascular endothelial growth factors (VEGF) and CA-125		

2.2.2. Cut-off Value for Diagnosis

A specific cutoff value must be established before CA-125 can be used as a useful diagnosis tool for endometriosis.

A specific cutoff value must be established before CA-125 can be used as a useful diagnosis tool for endometriosis. Endometriosis cutoff values are discussed in this section, evaluating their relevancy and effectiveness in clinical diagnosis.

In 1995, Hornstein et al. conducted a study comparing a new CA 125 assay with an older one for diagnosing endometriosis. The new assay showed slight improvements in sensitivity, specificity, and predictive values across stages, notably in stages III and IV. Sensitivities ranged from 0.06 to 0.14 for all stages and 0.20 to 0.54 for stages III-IV, with specificities of 0.91 to 0.97 using a cut-off of 35 U/ml[38].

CA-125 has been evaluated as a diagnostic tool for endometriosis without associated ovarian endometriomas by Kitawaki et al. 2005. The study found that the AUC for endometriosis without endometriomas was 0.788, significantly lower than that for cases with endometriomas (0.935, $P < 0.05$). The optimal diagnostic accuracy (78.8%) was achieved at a cutoff value of 20 U/mL for CA-125, with a negative predictive value (NPV) of 78.0% and a positive predictive value (PPV) of 92.9% at a cutoff value of 30 U/mL. The combined use of these two cutoff values provided improved diagnostic performance over the single cutoff of 35 U/mL[23].

Rosa E Silva et al. 2007 conducted a study to determine serum CA-125 values that could indicate the presence and stage of endometriosis. Using different cut-off values, the study found that a cut-off value of 10 IU/mL had a sensitivity of 64.2% and specificity of 81.1% for diagnosing endometriosis. A cut-off value of 20 IU/mL had lower sensitivity (30.4%) but higher specificity (98%). This value was able to differentiate between stages I and II and stages III and IV of endometriosis. The study concluded that using serum CA-125 levels alone as a diagnostic tool for endometriosis might not be advisable due to the observed low sensitivities[39].

Szubert et al. 2012 conducted a study to assess CA-125 levels in serum and peritoneal fluid as indicators of endometriosis. The mean CA-125 concentration was significantly higher in the endometriosis group in both serum (33.98 U/ml vs. 9.3 U/ml) and peritoneal fluid (2640.23 U/ml vs. 1241.88 U/ml). CA-125 levels showed a significant correlation with the stage of endometriosis ($R = 0.5993$, $p < 0.001$). Serum CA-125 exhibited moderate predictive value for distinguishing between endometriosis and non-endometriosis patients (AUC = 0.794). The study suggested a serum cut-off value of 11 U/ml with 68% sensitivity[40].

In 2016, Karimi-Zarchi et al. conducted a cross-sectional analysis on 87 women with suspected endometriosis. They found a significant correlation between preoperative serum CA-125 levels and various clinicopathological characteristics of endometriosis, including stage, lesion size, and adhesion score. Optimal cut-off levels for pre- and postmenopausal patients were suggested: 37 U/mL and 35 U/mL, respectively[41].

Table 2. Studies related to cutoff value of CA-125 in endometriosis diagnosis.

Year	Main objective	Type of study Size of sample Diagnosis method	Biomarkers	Main result	Main outcome	Ref
1995	To compare the serum CA-125 concentrations	Cohort 123 Laparoscopy	CA-125	The CA-125 concentrations determined by the new	This study indicates that while new CA-125 assay concentrations are	[38]

 doi:10.20944/preprints202404.1261.v1

	determined by both assays in women with and without endometriosis.			assay were highly correlated with concentrations determined by the older assay in patients with and without endometriosis (r = 0.96).	closely correlated to those of the older CA-125 assay, the improvement in the ability to differentiate between patients with and without endometriosis is small.	
2005	To evaluate the diagnostic significance of CA-125 for endometriosis without ovarian endometriomas.	Cohort 775 Laparoscopy	CA-125	Receiver operating characteristic curve analysis revealed that the area under the curve for endometriosis without endometriomas was 0.788, significantly smaller than that for endometriosis with endometriomas (0.935, $P < 0.05$). In diagnosis of endometriosis without endometriomas, both the maximal accuracy of 78.8% and the maximal diagnostic value of 61.2% were obtained at the cutoff value of 20 U/mL. Negative predictive value was 78.0% at the cutoff value of 20 U/mL, whereas positive predictive value was 92.9% at the cutoff value of 30 U/mL. This range is clearly superior to the empirical single cutoff of 35 U/mL.	In the diagnosis of endometriosis without endometriomas, combined use of two cutoff values for CA-125, 20 and 30 U/mL, provides improved diagnostic performance. However, the accuracy of using only CA-125 testing for diagnosis is still limited. Serum CA-125 testing can be done during initial screenings of women with possible endometriosis.	[23]
2007	To define the serum CA-125 values that best indicate the presence and stage of endometriosis.	Retrospective, cross-sectional 201 Laparoscopy	CA-125	Using a CA-125 serum concentration higher than 10 IU/mL as the cut-off value for the diagnosis of endometriosis yielded a sensitivity of 64.2% and a specificity of 81.1% (with a positive predictive value of 91.3% and a negative predictive value of 45.3%).	It is not advisable to use serum levels of CA-125 as a diagnostic tool. It should be borne in mind that the cut-off values proposed herein present low sensitivity, underscoring the role of laparoscopy in the definitive diagnosis. Combining serum CA-125 measurement with noninvasive methods such as transvaginal ultrasonography and magnetic resonance imaging might increase the sensitivity of this marker.	[39]

 doi:10.20944/preprints202404.1261.v1

2012	To evaluate CA-125 in serum and peritoneal fluid (PF) as an indicator of endometriosis.	Case control 56 Laparoscopy	CA-125	The mean value of CA-125 concentration in the endometriosis group was 33.98 U/ml, vs. 9.3 U/ml in the control group.	Cancer antigen 125 is a well-known biomarker for endometriosis and helpful in daily clinical practice when endometriosis is suspected. The cut-off value in serum suggesting endometriosis with 68% sensitivity is 11 U/ml. This value is the normal range for Ca-125 concentration.	[40]
2016	To evaluate the association between preoperative serum CA-125 levels and clinic pathological characteristic in women with endometriosis and find out the best serum CA-125 levels cut-off in pre- and post-menopause women.	Cross-sectional analysis 87 Laparoscopy	CA-125	The mean serum CA-125 level was 49.93±4.30 U/mL. There was a significant correlation between the endometriosis stage, lesion size, adhesion score and preoperative CA-125 plasma concentration.	Preoperative serum CA-125 is an important predictor for patients with endometriosis and should be considered when surgical management is suspected, especially if stage of disease, lesion size and adhesion score are undertaken.	[41]

2.2.3. Staging (Severity)

The severity and extent of endometriosis often guide therapeutic approaches. If CA-125 levels can be correlated with specific stages of the disease, it might serve as a valuable tool in patient evaluation.

Early work by Moloney et al. [42] established a significant positive correlation between endometriosis severity and CA-125 levels, suggesting the biomarker's potential as an early indicator of the disease. This foundational study opened the door to a series of investigations exploring CA-125's utility in disease staging and assessment.

O'Shaughnessy et al. [43] built on this foundation by examining CA-125 level variations across menstrual phases in women with and without endometriosis. Significant CA-125 level differences were observed between menstrual phases in endometriosis groups, particularly in severe cases (stages III and IV). A screening test was developed using the menses-to-follicular phase CA-125 ratio, demonstrating enhanced sensitivity (62.5%) and specificity (75%) compared to single-level tests.

In 2006, Amaral et al. analyzed CA-125 levels in women with pelvic endometriosis. They found higher serum and peritoneal fluid CA-125 levels in advanced cases compared to controls. A positive correlation between the serum and peritoneal fluid levels was noted ($r = 0.4880$). Increased CA-125 levels were associated with greater endometriosis severity[44].

Maiorana et al. 2007 conducted a study to assess serum CA-125 levels' diagnostic and severity-determining value in endometriosis. Serum CA-125 levels were significantly higher in endometriosis cases than controls ($p < 0.050$) and correlated with R-AFS scores[45].

 doi:10.20944/preprints202404.1261.v1

Ozhan et al. 2014 studied nine serum biomarkers for endometriosis diagnosis. Of these, only CA-125, syntaxin-5, and laminin-1 showed significant differences between endometriosis and control groups and among disease stages ($p < 0.01$). CA-125 levels particularly differed in stages III and IV compared to controls[46].

In a hypothetical study Santulli et al. 2014 could investigate the link between CA-125 levels and endometriosis progression across various factors like age, BMI, and clinical conditions. The results might indicate CA-125 as predictive of disease severity, with potential variations based on ethnicity. Interactions with age, BMI, and hormonal status might offer deeper insights. The study might suggest CA-125's utility in tracking disease progression, while emphasizing the need to consider ethnicity and other factors[47].

Knific et al. (2018) conducted a prospective case-control study to evaluate CA-125 and HE4 levels in patients with endometriosis-like symptoms and construct diagnostic models for endometriosis. CA-125 levels were significantly higher in endometriosis patients ($p<0.05$), with increased levels correlating with endometriosis severity. CA-125 showed significance across different stages and types of endometriosis compared to control [48].

In 2020, Sasamoto et al. conducted a cross-sectional analysis to evaluate CA-125's correlation with pain types and severity in young women with and without endometriosis. Their findings indicated that CA-125 levels didn't significantly differ based on pain type, severity, or frequency in both cases and controls. CA-125's ability to discriminate between endometriosis cases and controls, especially in those with dysmenorrhea, was limited (AUC = 0.51). The study suggests that while CA-125 is associated with endometriosis, variations in disease presentation and mechanisms could impact its effectiveness as a biomarker[49].

Zhao and Qu 2021 evaluated biomarkers for severe ovarian endometriosis (OEM) diagnosis, including CA-125, CA 19-9, PT, aPTT, TT, FIB, and D-dimer. Significant differences in CA-125, aPTT, FIB, and D-dimer levels were observed between stages I-II and III-IV OEM. CA-125 had an AUC of 0.953 for stage III-IV diagnosis. Combining CA-125 with D-dimer and aPTT gave an AUC of 0.961, suggesting an effective noninvasive method for diagnosing moderate to severe OEM[50].

Herranz-Blanco et al. 2023 ran a two-phase study on a diagnostic test for endometriosis. They confirmed CA-125, Brain-Derived Neurotrophic Factor (BDNF), and clinical variables' potential to distinguish endometriosis cases. The in vitro diagnostic test, which combined CA-125 and BDNF, showed superior accuracy over individual biomarkers. CA-125 was more effective for high-stage cases, while BDNF was consistent across stages. Elevated levels of CA-125 and BDNF were highlighted in endometriosis cases[51].

Overall, CA-125 proves to be a crucial biomarker for endometriosis staging, especially valuable in advanced stages and when used with other markers. Despite its variability, it remains a key tool in understanding disease severity and guiding treatment strategies.

Table 3. Studies related to staging value of CA-125 in endometriosis.

Year	Main objective	Type of study Size of sample Diagnosis method	Biomarkers	Main result	Main outcome	Ref
1989	To test whether there was a relationship between CA-125 levels and the severity of endometriosis that would allow CA-125 to be used to monitor disease progression.	Cohort 60 Laparoscopy	CA-125	There was a positive correlation (r = 0.63; P less than .0001) between disease severity and CA-125 levels. CA-125 was also elevated, compared with women with a normal pelvis, in patients with mild and moderate disease.	These results suggest that after malignancy has been excluded, CA-125 levels may offer a useful method of monitoring disease progress.	[42]

 doi:10.20944/preprints202404.1261.v1

				There was no relationship between CA-125 levels and the day of the menstrual cycle.		
1993	To examine variations in CA-125 levels during the three phases of the menstrual cycle in women with and without endometriosis.	Case control 100 Laparoscopy	CA-125	In the endometriosis groups, there was a significant difference in the mean CA-125 levels drawn at menses and those drawn in the follicular phase. In patients with severe endometriosis, there was also a difference in the mean CA-125 levels drawn at menses and in the luteal phase.	CA-125 levels during menses are elevated compared with those during the follicular phase in patients with endometriosis. Screening tests based on the relationship of multiple CA-125 levels taken throughout the menstrual cycle were more sensitive for detection of endometriosis than tests based on a single CA-125 level.	[43]
2006	To correlate CA-125 levels in serum and peritoneal fluid from women with and without pelvic endometriosis.	Prospective, cross-sectional, controlled study 52 Laparoscopy	CA-125	CA-125 levels in serum and peritoneal fluid were higher in patients with advanced pelvic endometriosis (means of 39.1 ± 45.8 U/ml versus 10.5 ± 5.9 U/ml in serum, $p < 0.005$; 1,469.4 ± 1,350.4 U/ml versus 888.7 ± 784.3 U/ml in peritoneal fluid, $p < 0.05$), and showed a positive correlation between each other (correlation coefficient (r) = 0.4880).	There is a positive correlation between serum and peritoneal fluid values of CA-125 in women with and without endometriosis, and their levels are higher in peritoneal fluid. Advanced endometriosis is related to higher levels in both serum and peritoneal fluid.	[44]
2007	To investigate the clinical value of the serum CA-125 level for diagnosing and determining the severity of endometriosis and pelvic pain associated with endometriosis.	Case control 86 Laparoscopy	CA-125	The mean serum CA-125 levels of women with endometriosis were higher than those of the control group ($p<0.050$).	CA-125 serum levels were related to endometriosis and R-AFS score in the evaluated patient series. No correlation was found between serum levels of CA-125 and pelvic pain in patients with endometriosis.	[45]
2014	To investigate the diagnostic potentials	Case control 80	α-enolase, macrophage	The serum levels of α-enolase, macrophage	The concurrent measurement of the	[46]

 doi:10.20944/preprints202404.1261.v1

	of the serum levels of nine different biomarkers in endometriosis.	Laparoscopy	migration inhibitory factor, leptin, interleukin-8, anti-endometrial antibody, phosphoinositide dependent protein kinase 1, CA-125, syntaxin-5, and laminin-1	migration inhibitory factor, leptin, interleukin-8 and anti-endometrial antibodies showed a statistically significant difference neither between control and endometriosis groups nor among control group and endometriosis subgroups. The serum levels of CA-125, syntaxin-5 and laminin-1 showed a statistically significant difference both between the control and endometriosis groups ($p<0.01$) and among control group and endometriosis subgroups	three biomarkers including CA-125, STX-5 and LN-1 might be a useful non-invasive test in strengthening the diagnosis of endometriosis and in predicting its severity.	
2014	To determine whether cancer antigen-125 (CA-125) levels are increased in women with endometriosis, especially in those with endometriomas (OMAs), deep infiltrating lesions (DIE), and superficial endometriosis (SUP).	Cross-sectional 679 Laparoscopy	CA-125	Women with endometriosis displayed higher mean serum CA-125 levels compared with disease-free controls (50.1 ± 62.4 U/mL vs 22.5 ± 25.2 U/mL; $p \leq .001$).	Serum CA-125 levels were significantly increased in women with severe forms of endometriosis, OMA, and DIE lesions. In addition, elevated serum Ca-125 levels were associated with more severe and extended DIE lesions. In women with superficial peritoneal lesions, CA-125 levels were not different from women without endometriosis.	[47]
2018	To evaluate preoperative levels of CA-125 and HE4 in patients with endometriosis-like symptoms.	Case control (Prospective) 221 Laparoscopy	CA-125, HE4	CA-125 serum levels were significantly elevated in the endometriosis patients compared with the control patients ($p = 1.3 \times 10\text{-}10$), while the difference in the HE4 serum levels did not reach significance ($p > 0.05$).	CA-125 levels were significantly elevated in endometriosis patients compared with control patients ($p<0.001$).	[48]

 doi:10.20944/preprints202404.1261.v1

2020	To examine whether CA-125 correlates with different types and severity of pain among adolescents and young women with and without endometriosis and assess its performance as an endometriosis biomarker among those presenting with dysmenorrhea in this young population.	Cross-sectional analysis 575 Laparoscopy	CA-125	Average CA-125 values were 12.5 U/mL in controls and 12.1 U/mL in cases adjusted for age. CA-125 did not differ by pain type, its severity, or frequency in endometriosis cases or controls.	CA-125 did not efficiently discriminate endometriosis cases from controls, even when accounting for pain symptomatology. Average CA-125 values were low in adolescents and young women in both endometriosis cases and controls, suggesting cautious interpretation may be needed when measuring CA-125 in this population.	[49]
2021	To investigate the relationships of cancer antigen (CA) 125, CA 19-9, prothrombin time (PT), activated partial thromboplastin time (aPTT), thrombin time (TT), fibrinogen (FIB), and D-dimer values with ovarian endometriosis (OEM).	Case control (Retrospective) 571 Laparoscopy	CA-125, CA 19-9, aPTT, TT, PT, D-dimer, and FIB	The serum CA-125, aPTT, FIB and D-dimer levels were statistically different between OEM patients in the stages I to (and) II group and those in the stages III and IV group (P<0.05). However, a statistical difference in CA 19-9 levels and TT was only found between patients with stages III and IV OEM.	The combined index of CA-125, aPTT, and D-dimer is a valid noninvasive preoperative method for the evaluation of moderate and severe OEM, and may help to decrease the interval between the first complaint and a definitive diagnosis.	[50]
2023	To develop a diagnostic test based on the combination of serum biomarkers and clinical variables.	Case control 204 Laparoscopy	CA-125, BDNF	CA-125 and BDNF, can distinguish endometriosis cases from controls with statistical significance.	Although no individual cut-off values were set, CA-125 and BDNF levels were demonstrated to be elevated in patients with endometriosis, with CA-125 mostly able to identify high-stage endometriosis and BDNF performing well for both low- and high-stage disease.	[51]

2.2.4. Management (Treatment and Prognosis)

In the realm of endometriosis management, CA-125 has shown promise as a tool for guiding therapeutic decisions, monitoring disease progression, and assessing treatment efficacy.

 doi:10.20944/preprints202404.1261.v1

Pittaway and Fayez 1986 found CA-125 levels linked to endometriosis severity and other conditions. With a 53% sensitivity and 93% specificity at 16 U/ml, CA-125 correlated with endometriosis in 84% of cases and showed potential in monitoring treatment response[52].

Koninckx et al. 1996 observed higher CA-125 levels during menstruation associated with endometriosis. Combining clinical exams and CA-125 assessment proved reliable for diagnosing deep endometriosis, suggesting a non-invasive diagnostic method[53].

Chen et al. 1998 reported CA-125's 61.1% sensitivity and 87.5% specificity for diagnosing endometriosis in women with dysmenorrhea. While it had limited screening use, CA-125 was useful in monitoring therapy and spotting recurrence in advanced cases[54].

These studies underscore the potential of CA-125 as a tool not only for assessing the effectiveness of various interventions and treatments but also as a means of monitoring long-term disease progression and recurrence. This will contribute to more tailored and effective management strategies for endometriosis.

Table 4. Studies related to role of CA-125 in managing endometriosis.

Year	Objective	Type of study Size of sample Diagnosis method	Biomarkers	Main result	Main outcome	Ref
1986	To determine CA-125 potential usefulness in the diagnosis and management of endometriosis.	Cohort 392 Laparoscopy	CA-125	In women with minimal, mild, moderate, and severe endometriosis, the mean CA-125 levels (+/- standard deviation) were 13.6 +/- 6.8, 22.8 +/- 15.5, 27 +/- 17, and 50 +/- 28 U/ml, respectively, and were significantly higher than mean levels (7.8 +/- 4.1) in 46 women with a normal laparoscopic examination.	Changes in the CA-125 levels correlated with the clinical course of endometriosis in 37 of 44 (84%) women (P less than 0.001). The determination of CA-125 levels may assist in the evaluation and treatment of women with endometriosis.	[52]
1996	To evaluate, a clinical examination during menstruation and plasma CA-125 concentrations to diagnose deep endometriosis.	Prospective, Retrospective, Clinical validation 217 Laparoscopy	CA-125	CA-125 concentrations were higher during menstruation and correlated with deep endometriosis and with deep and cystic ovarian endometriosis. Nodularities at clinical examination or follicular phase CA-125 concentrations > 35 U/mL are useful to decide that a bowel preparation should be given, achieving a sensitivity of 87% and a specificity of 83%.	Clinical examination during menstruation can diagnose reliably deep endometriosis, cystic ovarian endometriosis, or cul-de-sac adhesions. This test, preferentially combined with a follicular phase CA-125 assay, should be used to decide whether a preparation for bowel surgery should be given.	[53]
1998	To estimate the value of CA-125 for the diagnosis of endometriosis in	Prospective 157 Laparoscopy	CA-125	The sensitivity and specificity of serum CA-125 for the diagnosis of	For endometriosis, CA-125 is a valuable adjuvant in the follow-	[54]

 doi:10.20944/preprints202404.1261.v1

women with dysmenorrhea, as well as its significance in monitoring therapy and follow-up.		endometriosis were 61.1% and 87.5% respectively. Elevated CA-125 (±35 U/ml) was noted in 65/75 cases (86.7%) with advanced endometriosis, but in only 15/56 patients (26.8%) with minimal and mild endometriosis.	up of recurrence in patients with advanced endometriosis and initially elevated CA-125 levels. It is not an effective screening tool for patients with dysmenorrhea, or for monitoring therapy.

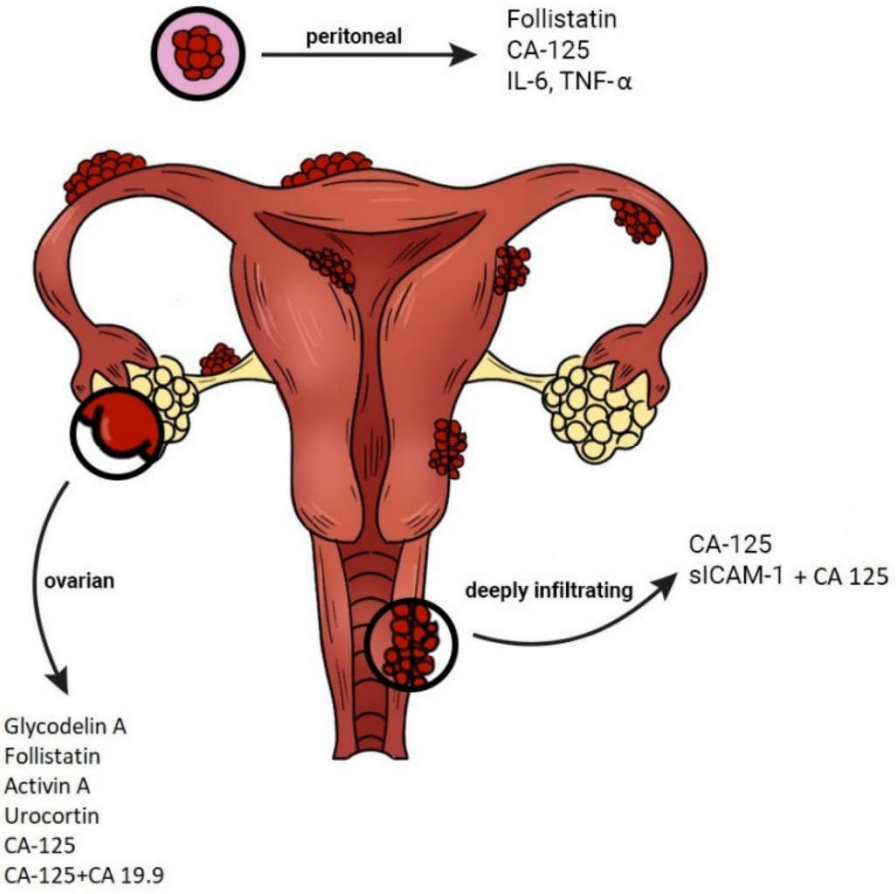

Figure 2. CA-125 in endometriosis [55].

2.3. MiRNAs in Endometriosis

In this part of the review, we'll organize the diverse roles of miRNA in endometriosis into three main sections: Diagnosis, Staging, and Management. We'll look at key studies in each area to better understand how miRNA contributes to each aspect of the disease.

2.3.1. Pathogenesis and Diagnosis

MiRNAs have emerged as critical molecular players in the pathogenesis and diagnosis of endometriosis, offering insights into the disease's complex mechanisms and potential diagnostic markers.

The narrative begins with Fassbender et al. and Jia et al., whose pioneering work on miRNAs as non-invasive diagnostic tools opened the door to a new era of molecular diagnostics. Fassbender et al. highlighted the role of proteomics, noting a 91% sensitivity and 80% specificity for diagnosing endometriosis during the luteal phase, despite no miRNA differences by stage or cycle phase [56].

 doi:10.20944/preprints202404.1261.v1

In a 2013, study by Jia et al. investigated plasma microRNAs as non-invasive tools for endometriosis diagnosis. They found 27 differentially expressed microRNAs, with miR-17-5p, miR-20a, and miR-22 significantly down-regulated in endometriosis cases. These microRNAs exhibited promising diagnostic potential, as validated by AUC values of 0.74, 0.79, and 0.85, respectively. The study highlights the viability of these microRNAs as potential diagnostic markers for endometriosis[57].

In 2016, Cosar et al. examined serum miRNAs for endometriosis diagnosis. They found miR-125b-5p, among others, significantly up-regulated in patients. MiR-125b-5p, combined with miR-451a and miR-3613-5p, achieved a perfect AUC score of 1.000 in differentiation. The study emphasizes miR-125b-5p's potential as a diagnostic biomarker for endometriosis[58].

In 2016, Wang et al. studied serum miRNA profiles in minimal-mild endometriosis patients. They found 98 downregulated and 10 upregulated miRNAs. qPCR highlighted 21 new miRNAs among the downregulated. These miRNAs play roles in endometriosis' pathophysiology. The research emphasizes miRNAs as early detection biomarkers and underscores their complex role in the disease[59].

In a 2018 study, Xu et al. explored circRNA differences in eutopic endometrium. They found 11 upregulated and 77 downregulated circRNAs in ovarian endometriosis. qRT-PCR confirmed high levels of circ_0004712 and circ_0002198, with circ_0002198 having an AUC of 0.846 and circ_0004712 an AUC of 0.704 in ROC analysis. This highlights circRNAs' role in endometriosis and potential biomarkers, including miRNA, for diagnosis[60].

In 2018, Haikalis ME et al. studied miRNA differences in endometriosis. They found miR-204 down-regulated in the eutopic endometrium of women with endometriosis. Various ectopic implants (peritoneal lesions, ovarian endometriomas, deep-infiltrating endometriosis) showed distinct miRNA expressions. miRNA profiles differed among women with different lesion types and controls. Comparing ectopic lesions to eutopic endometrium revealed significant miRNA expression differences. This highlights complex miRNA patterns in endometriosis, suggesting unique biochemistry in various lesion types. Understanding these miRNA variations can aid personalized care, improving diagnostics and treatments[61].

In the study by Pateisky et al. 2018, researchers aimed to find non-invasive biomarkers for endometriosis. Specific miRNAs were identified that differed significantly between the groups. Notably, hsa-miR-154-5p showed promise as a potential diagnostic marker for endometriosis, both alone and in combination with other miRNAs. Additionally, these miRNAs' expressions varied during different menstrual cycle phases. The study suggests that specific plasma miRNAs, possibly in combination with clinical factors, could aid in diagnosing endometriosis non-invasively[62].

In a 2019 study, Nisenblat et al. aimed to identify plasma microRNAs (miRNAs) for diagnosing endometriosis. They found three miRNAs (miR-155, miR574-3p, miR139-3p) consistently dysregulated in endometriosis patients. The combined miRNA panel showed a sensitivity of 83% and specificity of 51%, with AUC values ranging from 0.619 to 0.674. The researchers concluded that while specific miRNAs have diagnostic relevance, using them as independent diagnostic tests requires further research[63].

Vanhie et al.'s 2019 study evaluated plasma miRNAs as markers for endometriosis. 42 miRNAs differed between patients with and without the condition. Model 2, including miRNAs hsa-miR-125b-5p, hsa-miR-28-5p, and hsa-miR-29a-3p, had a 60% AUC with 78% sensitivity and 37% specificity, suggesting miRNAs' diagnostic potential for endometriosis[64].

In the 2020 study by Zhang et al., exosomal miRNA profiles were investigated to find potential biomarkers for endometriosis. They identified 24 significantly differentially enriched miRNAs. Notably, miR-22-3p and miR-320a were upregulated in serum exosomes from endometriosis patients. ROC analysis revealed AUC values of 0.855 for miR-22-3p and 0.827 for miR-320a, suggesting their potential as diagnostic biomarkers for endometriosis[65].

In Razi et al. 's 2020 study, they examined miR-185-5p and target genes VEGF and PDGF in endometriosis patients versus controls. miR-185-5p was significantly downregulated in endometriosis patients, with an AUC of 0.919 in ROC analysis. However, VEGF and PDGF miRNA

 doi:10.20944/preprints202404.1261.v1

expression didn't significantly differ between groups. The study suggests plasma miR-185-5p could be a non-invasive biomarker for early endometriosis detection[66].

In Papari et al.'s 2020 study, eight miRNAs were found at lower levels in women with endometriosis. These miRNAs had sensitivities and specificities between 0.36 and 1.00. A combination improved sensitivity and specificity to 0.92 and 0.86. Notably, a panel of five miRNAs (miR-17-5p, miR-20a-5p, miR-199a-3p, miR-143-3p, and let-7b-5p) achieved 0.96 sensitivity and 0.79 specificity. This miRNA panel could be a non-invasive diagnostic tool for endometriosis, potentially replacing laparoscopy[19].

Borisov et al. 2020 aimed to diagnose adenomyosis (AM) non-invasively by analyzing dysregulated miRNAs in eutopic endometrium. Four miRNAs were upregulated (miR-181a, miR-191, miR-195, miR-200b) and five were downregulated (miR-10b, miR-200c, miR-10a, miR-221, miR-31) in AM patients' endometrium. The miRNA expression ratios could diagnose AM with sensitivities of 65% to 74% and specificities of 72% to 86%. The study suggests these miRNAs and ratios could be useful markers for non-invasive adenomyosis diagnosis[67].

In Misir et al.'s 2021 study, they assessed serum miR-34a-5p and miR-200c levels in women to identify potential endometriosis biomarkers. MiR-34a-5p was lower, and miR-200c was higher in endometriosis patients than controls. ROC analysis showed both miRNAs' diagnostic potential, with miR-200c having 100% sensitivity and specificity. The research suggests these miRNAs as possible endometriosis diagnostic tools[68].

The study by Zafari et al. 2021 aimed to find a diagnostic panel for endometriosis using miRNAs. They studied miR-224-5p, miR-199-3p, and let-7d-3p in blood samples. They found upregulation of miR-199b-3p and downregulation of miR-224-5p and let-7d-3p in endometriosis. Individually, these miRNAs showed moderate diagnostic accuracy. However, when combined, they had high accuracy (AUC 0.992) in distinguishing endometriosis patients from controls. This suggests that these miRNAs could serve as a non-invasive diagnostic biomarker panel for endometriosis[69].

A pivotal study by Bendifallah et al. 2022 has established a significant breakthrough in the diagnosis of endometriosis through a blood-based miRNA signature. In this prospective trial, involving analysis of 200 plasma samples from women with symptoms suggestive of endometriosis, the research team leveraged the synergy of advanced miRNA profiling and artificial intelligence (AI) technologies. The developed diagnostic model demonstrated exceptional efficacy, exhibiting a sensitivity of 96.8%, specificity of 100%, and an Area Under the Curve (AUC) of 98.4%. The study identified a specific miRNA panel, comprising miR-124-3p, miR-6509-5p, miR-548l, miR-26a-2-3p, miR-3622a-3p, miR-3168, miR-29b-1-5p, miR-30e-3p, miR-3124-5p, and miR-4511, as highly effective in diagnosing endometriosis. This panel was notably efficient in capturing the heterogeneous nature of the disease. The findings from Bendifallah et al.'s study highlight the transformative potential of this miRNA signature in diagnosing endometriosis. The accuracy and reproducibility of this non-invasive method suggest it could replace invasive diagnostic surgeries. The implications of such a development are profound, likely influencing future clinical guidelines and recommendations at both national and international levels[18].

Another noteworthy study in our review is by Kumari et al. 2022, which focused on assessing the diagnostic potential of various miRNAs in serum samples from women affected by endometriosis, endometrioid carcinoma of the ovary (ECO), and endometrioid endometrial cancer (EC). Conducted as a cohort study, it involved 40 patients who underwent surgical procedures for various gynecological conditions, including pelvic pain, endometriosis, infertility, and ovarian tumors. The study found distinct patterns of miRNA expression across different conditions. In cases of endometriosis, miR-16 was significantly down regulated, while miR-99b, miR-125a, miR-143, and miR-145 were upregulated ($P<0.05$). For the ECO group, a down regulation of miR-16 and miR-125a and an upregulation of miR-99b, miR-143, and miR-145 were observed. In endometrioid EC, there was a down regulation in miR-16, miR-99b, miR-125, and miR-145, with an upregulation in miR-143. ROC curve analysis conducted revealed that miR-99b, miR-125a, miR-143, and miR-145 could serve as diagnostic markers for endometriosis. Additionally, miR-145 showed diagnostic power for ECO, and a combination of miR-16, miR-99b, miR-125a, and miR-145 was indicative of endometrioid EC.

 doi:10.20944/preprints202404.1261.v1

They concluded that certain circulating miRNAs, notably miR-99b, miR-16, miR-125a, and miR-145, might act as valuable indicators for endometriosis, ECO, and EC. These miRNAs hold promise as potential biomarkers for the early diagnosis and management of these debilitating diseases[70].

Abo et al. 2022 studied circulating miRNAs related to ovarian endometriosis (OMA). They found let-7b-5p and miR-92a-3p overexpressed in OMA patients' plasma. These miRNAs, along with miR-93-5p, potentially target the down regulated gene KIAA1324. Experiments showed let-7b-5p and miR-92a-3p reduce KIAA1324 expression. The study indicates KIAA1324's involvement in endometriosis, regulated by these miRNAs[71].

Lin et al. 2023 evaluated the diagnostic value of serum miR-17-5p and miR-424-5p for endometriosis. Both miRNAs were down regulated in patients. Individually, miR-17-5p had an AUC of 0.865 and miR-424-5p had 0.737. Together, they achieved an AUC of 0.938. These miRNAs were inversely related to factors like VEGF, IL-4, IL-6, and CA-125. The study suggested that their combined expression could be a key diagnostic marker for endometriosis[72].

In sum, miRNA profiles present a promising avenue for the non-invasive diagnosis of endometriosis, offering high sensitivity and specificity across various studies. While individual miRNAs show potential, panels of miRNAs combined with other diagnostic factors demonstrate enhanced accuracy, suggesting their significant role in early detection, disease differentiation, and potentially replacing more invasive diagnostic methods.

Table 5. Studies related to diagnostic value of miRNAs in endometriosis.

Year	Objective	Type of study Size of sample Diagnosis method	miRNA type	Main result	Main outcome	Ref
2012	To perform a combined miRNA microarray and proteomics analysis.	Case control 49 Laparoscopy	Not mentioned	miRNA analysis of eutopic endometrium did not show any differentially expressed genes in women with endometriosis when compared with controls, regardless of endometriosis stage or cycle phase.	miRNA expression of eutopic endometrium was comparable in women with and without endometriosis, but different in menstrual endometrium when compared with luteal endometrium in women with endometriosis.	[56]
2013	Can plasma microRNAs be used as a non-invasive diagnostic test for the detection of endometriosis?	Prospective study 46 Laparoscopy	miR-15b-5p, miR-17-5p, miR-20a, miR-21, miR-22 and miR-26a	miR-17-5p, miR-20a and miR-22 were significantly down-regulated.	Plasma miR-17-5p, miR-20a and miR-22 are down-regulated in women with endometriosis compared with those without endometriosis.	[57]

 doi:10.20944/preprints202404.1261.v1

2016	To investigate serum microRNAs (miRNAs) in women with endometriosis.	Case control 48 Laparoscopy	miR-3613-5p, miR-125b-5p, miR-150-5p, miR-342-3p, miR-143-3p, miR-145-5p, miR-500a-3p, miR-451a, miR-18a-5 piR-6755-3p,	miR-3613-5p, miR-6755-3p were down-regulated and miR-125b-5p, miR-150-5p, miR-342-3p, miR-143-3p, miR-145-5p, miR-500a-3p, miR-451a, miR-18a-5p were up-regulated more than 10-fold in the microarray.	Identified several miRNAs in serum that distinguished subjects with endometriosis from those without. miR-125b-5p had the greatest potential as a single diagnostic biomarker. A combination of that miRNA with miR-451a and miR-3613-5p further improved diagnostic performance.	[58]
2016	To further analyze the serum miRNAs profile in endometriosis.	Case control 50 Laparoscopy	miR-30c-5p, miR-127-3p, miR-99b-5p, miRNA-15b-5p, miRNA-20a-5p, miR-424-3p, miR-185-5p	Only 21 of 98 significantly downregulated miRNAs, and none of significantly upregulated miRNAs were reported in published literature, which may be due to the differences in samples and analytical methods.	Circulating miRNAs may be useful as detection biomarkers for the early diagnosis of minimal-mild endometriosis.	[59]
2018	To profile the circular RNAs (circRNAs) expressed in eutopic endometrium from patients with ovarian endometriosis and explore potential clues to the pathogenesis of endometriosis, providing evidence for clinical diagnosis and treatment.	Case control 63 Laparoscopy	Two upregulated circRNAs: 1.circ_0004712 2.circ_000219	Among 88 differentially expressed circRNAs, 11 were upregulated and 77 were downregulated in the eutopic endometrium of patients with endometriosis.	Provides evidence that circRNAs are differentially expressed between eutopic and normal endometrium, which suggests that circRNAs are candidate factors in the activation of endometriosis. circ_0002198 and circ_0004712 may be potential novel biomarkers for the diagnosis of ovarian endometriosis.	[60]

 doi:10.20944/preprints202404.1261.v1

2018	To determine differences in miRNA expression between eutopic endometrium.	Case control 47 Laparoscopy	miR-21, miR-424, miR-10b	miRNA expression between ectopic implants Of the six miRNAs quantified, expression of miR-21, miR-424, and miR-10b was differentially regulated between endometriotic lesions from different anatomical sites. miR-21 expression was significantly lower in PE compared to OMA	miRNA expression differs between the eutopic endometrium of women with endometriosis compared to a symptomatic control population without endometriosis.	[61]
2018	This study tested whether they could serve as putative non-invasive biomarkers for endometriosis, and their expression differences between endometriosis patients and controls.	Case control (Prospective) 92 Laparoscopy	hsa-miR-154-5p,hsa-miR-196b-5p, hsa-miR-378a-3p, hsa-miR-33a-5p	Data showed that a specific plasma miRNA signature is associated with endometriosis and that hsa-miR-154-5p, which alone or in combination with hsa-miR-196b-5p, hsa-miR-378a-3p, and hsa-miR-33a-5p and the clinical parameters of body mass index and age, are potentially applicable for non-invasive diagnosis of the disease.	miRNA seem to be promising candidates for the non-invasive diagnosis of endometriosis.	[62]
2019	To identify endometriosis-specific plasma miRNAs and determine their diagnostic test accuracy.	Case control and cohort studies 249 Laparoscopy	miR-155, miR574-3p and miR139-3p	Forty-nine miRNAs were differentially expressed in women with endometriosis. Nine maintained dysregulation in the selection cohort, but only three (miR-155, miR574-3p and miR139-3p) did so in the validation cohort.	Plasma miRNAs demonstrated modest sensitivity and specificity as diagnostic tests or triage tools for endometriosis.	[63]
2019	Retrieving data. Wait a few seconds	Case control and Cohort 210	hsa-miR-125b-5p, hsa-miR-28-5p and hsa-miR-29a-3p	hsa-miR-125b-5p, hsa-miR-28-5p and hsa-miR-29a-3p)	A possible biological link between certain	[64]

 doi:10.20944/preprints202404.1261.v1

	and try to cut or copy again.	Laparoscopy		had diagnostic power above chance performance in the independent validation (AUC = 60%) with an acceptable sensitivity (78%) but poor specificity (37%).	miRNAs and endometriosis, but the potential of these miRNAs as clinically useful biomarkers is questionable in women with infertility.	
2020	Exploring aberrant exosomal miRNA profiles by using miRNA microarray and at providing more accurate molecular biomarkers of endometriosis.	Case control 50 Laparoscopy	miR-22-3p, miR-320a	miR-22-3p and miR-320a were significantly upregulated in serum exosomes from patients with endometriosis compared with negative individuals. ROC curve revealed that the serum exosomal miR-22-3p and miR-320a yielded the area under the curve values of 0.855 and 0.827.	Results demonstrated that exosomal miR-22-3p and miR-320a were significantly increased in the sera of patients with endometriosis. The two miRNAs may be useful potential biomarkers for endometriosis diagnosis.	[65]
2020	Evaluated the differential expression of circulating miRNA-185-5p (miR-185-5p), vascular endothelial growth factor (VEGF), and platelet-derived growth factor (PDGF) target genes between endometriosis and healthy women.	Case control 50 Laparoscopy	miRNA-185-5p, VEGF, PDGF	miR-185-5p was significantly down-regulated in the case group compared with the controls.	The low expression of miR-185-5p in the plasma of women with endometriosis could be employed as an important non-invasive biomarker for early detection and screening of endometriosis by blood samples.	[66]
2020	To identify novel candidate diagnostic microRNA (miRNA) markers of endometriosis.	Retrospective cohort 20 Laparoscopy	24 candidate miRNAs and 3 reference miRNA: hsa-miR-150-5p, hsa-miR-199a-3p, hsa-miR-143-3p, hsa-miR-199a-5p, hsa-miR-335-3p, hsa-miR-381-3p, hsa-miR-224-5p, hsa-miR-340-5p, hsa-let-7d-3p, hsa-miR-92a-3p, hsa-miR-221-3p, hsa-miR-486-5p, hsa-let-7b-	Combination of five miRNAs (miR-17-5p, miR-20a-5p, miR-199a- 3p, miR-143-3p, and let-7b-5p) produced sensitivity and specificity of 0.96 and 0.79 with PPV and NPV of 0.80	A panel of candidate miRNAs was comparable to laparoscopy in distinguishing between women with endometriosis and control women.	[19]

 doi:10.20944/preprints202404.1261.v1

			5p, hsa-miR-122-5p, hsa-miR-21-5p, hsa-miR-133a-3p, hsa-miR-148a-5p, hsa-let-7a-3p, put-miR-5, put-miR-27, hsa-miR-125b-5p, hsa-miR-17-5p, hsa-miR-20a-5p, hsa-miR-3613-5p, hsa-miR-103a-3pa, hsa-miR-30e-5p, hsa-miR-148b-3p	and 0.96, respectively.		
2020	To explore the diagnostic potency of this approach[67].	Case control 66 Laparoscopy	Up-regulated: miR-181a, miR-191, miR-195 and miR-200b Down-regulated: miR-10b, miR-200c, miR-10a, miR-221 and miR-31	mir-10b, miR-200c and miR-191 were significantly dysregulated in the eutopic endometrium of AM patients. The expression ratio of reciprocally dysregulated microRNAs allowed us to diagnose AM with a range of sensitivity from 65% to 74%, and of specificity from 72% to 86%.	The analysis of microRNAs from the eutopic endometrium might present a promising low-invasive method of AM diagnostics.	[67]
2021	To evaluate the level of serum miR-34a-5p and miR-200c from women with and without endometriosis[68].	Case control 136 Laparoscopy	CA-125, miR-34a-5p, miR-200c	miR-34a-5p expression levels were decreased and miR-200c expression levels were increased in the endometriosis patients compared to the control group.	Serum miRNAs may provide a promising opportunity for diagnosis of endometriosis. Understanding the role of circulating miRNAs will serve a better comprehension of the systemic effects of endometriosis and offer options for new treatments.	[68]
2021	To investigate whether the combination of miR-224-5p, miR-199-3p, and let-7d-3p is a suitable diagnostic panel for endometriosis[69].	Case control 50 Laparoscopy	miRNAs 199b-3p, 224-5p,Let-7d-3p	Upregulation of miRNAs 199b-3p (P value < 0.001) and down-regulation of 224-5p (P value < 0.001) and miRNA let-7d-3p (P value < 0.05) in women	The levels of miRNAs 199b-3p, 224- 5p, and Let-7d-3p in plasma are potential diagnostic biomarkers for	[69]

 doi:10.20944/preprints202404.1261.v1

				with endometriosis compared to non-endometriosis women.	endometriosis patients.	
2022	To analyze the current human miRNAome to differentiate between patients with and without endometriosis, and to develop a blood-based miRNA diagnostic signature for endometriosis with internal cross-validation[70].	Prospective trial 200 Laparoscopy	miR-3622a-,miR-504-3p,miR-526a-3p,miR-124-3p,miR-3923, miR-5004-3p,miR-520h, miR-5700, miR-6502-5p, miR-6799-3p,miR-6826-5p,miR-6837-5p,miR-7108-3p,miR-1180-5p,miR-3064-3p,miR-3168, miR-3185, miR-4674, miR-4764-5p, miR-516a-3p, miR-542-5p, miR-889-5p, miR-1253, miR-1292-5p, miR-138-1-3p,miR-1910-5p,miR-216b-3p,miR-26a-2-3p, miR-29b-1-5p, miR-30e-3p, miR-3117-5p, miR-3122, miR-3137, miR-4696, miR-4703-5p, miR-4715-5p, miR-4740-5p, miR-4749-5p, miR-4797-3p, miR-4804-5p, miR-4999-5p, miR-5681a, miR-6075, miR-6509-5p, miR-6824-3p, miR-6875-3p, miR-1278, miR-1343-5p, miR-1973, miR-203a-5p, miR-208a-3p, miR-208a-5p, miR-3124-5p,miR-3176, miR-3683, miR-3691-5p, miR-375-5p, miR-3939, miR-3975, miR-4260, miR-4295, miR-4296, miR-433-3p, miR-4445-3p, miR-4455, miR-4511, miR-4536-3p, miR-4655-5p, miR-4725-5p, miR-4738-5p, miR-4750-3p, miR-514b-5p, miR-548aw, miR-548w, miR-5572, miR-5702, miR-573, miR-6788-3p, miR-6811-3p, miR-6813-5p, miR-6830-5p, miR-6872-3p, miR-6888-5p,miR-7109-5p, miR-7150, miR-7152-5p	The most accurate signature provides a sensitivity, specificity, and Area Under the Curve (AUC) of 96.8%, 100%, and 98.4%, respectively, and is sufficiently robust and reproducible to replace the gold standard of diagnostic surgery.	The present study supports the use of a blood-based miRNA signature of endometriosis.	[18]

 doi:10.20944/preprints202404.1261.v1

2022	To evaluate the diagnostic potential of differentially expressed miRNAs in serum samples of women with endometriosis, ECO and EC to establish them as diagnostic biomarkers[71].	Cohort 40 Laparoscopy	miR-16, miR-20a, miR-99b, miR-125a, miR-143, miR-145	miR-16 was downregulated (P<0.05) whereas miR-99b, miR-125a, miR-143 and miR-145 were upregulated (P<0.05).	Certain circulating miRNAs (miB99b, miR-16, miR-125a, miR-145) might act as indicators and discriminators of endometriosis and endometrioid subtypes of EC and ovarian cancer and might serve as potential biomarkers for early diagnosis and management of these debilitating diseases.	[70]
2022	To identify circulating miRNAs associated with ovarian endometriosis (OMA)[72[.	Case control 188 Laparoscopy	miR-484, miR-192-5p, miR-16-5p, miR-215-5p, let-7b-5p, miR-92a-3p, miR-93-5p, miR-30a-5p, U6	let-7b and miR-92a-3p, and miR-93-5p	Results suggested that KIAA1324 might be involved in endometriosis through the downregulating action of two circulating miRNAs. As these miRNAs were found to be overexpressed, their quantification in plasma could provide a tool for an early diagnosis of endometriosis.	[71]
2023	Investigated the diagnostic value of serum miR-17-5p, miR-424-5p, and their combined expressions for EMT[73].	Case control 160 Laparoscopy	miR-424-5p, miR-17-5p	miR-17-5p and miR-424-5p were down regulated in EMT patients. For diagnosing EMT, the AUC of miR-17-5p was 0.865 and cutoff value was 0.890 (91.3% sensitivity and 85% specificity).	miR-424-5p combined with miR-17-5p has high diagnostic efficacy for EMT.	[72]

2.3.2. Staging (Severity)

The progression of endometriosis is categorized into stages, which influence therapeutic strategies and provide insights into the disease's severity. Understanding whether miRNA profiles correlate with these stages can be invaluable for clinicians.

 doi:10.20944/preprints202404.1261.v1

The study by Bashti et al. 2018 aimed to investigate the potential of miR-31 and miR-145 expression in plasma as non-invasive biomarkers for endometriosis. The study found that miR-31 expression was greatly reduced in patients with moderate/severe and minimal/mild endometriosis, while miR-145 expression was significantly increased in patients with minimal/mild endometriosis. These findings suggest that these miRNAs may have diagnostic implications, potentially aiding in early diagnosis and patient monitoring for endometriosis[73].

In a 2020 study by Moustafa et al., six miRNAs were identified with strong diagnostic potential for endometriosis. Their combined use resulted in a robust AUC of 0.94. These miRNAs could differentiate between endometriosis stages and weren't affected by menstrual cycle or hormonal medications. This suggests their potential for early diagnosis and better patient outcomes[10].

In a 2022 study by Perricos et al., they examined miRNA expression in saliva and plasma of women with endometriosis, focusing on 28 miRNAs. They found hsa-mir-135a significantly upregulated in both samples from affected women, especially during the secretory menstrual phase. This suggests hsa-mir-135a could be a noninvasive biomarker for endometriosis diagnosis[74].

Table 6. Studies related to staging value of miRNAs in endometriosis.

Year	Objective	Type of study Size of sample Diagnosis method	miRNA type	Main result	Main outcome	Ref
2018	To examine the association of miR-31 and miR-145 expression in plasma with the presence of endometriosis[74].	Case control 88 Laparoscopy	miR-145 and miR-31	miR-31 expression levels in stage 3 or 4 and stage 1 or 2 were significantly down-regulated (less than 0.01-fold, P<0.05), while the expression level of miR-145 was significantly up-regulated in women with endometriosis in stage 1 or 2.	miR- 31 was under-expressed in patients with endometriosis, while miR-145 was over-expressed in stage 1 or 2, indicating that they were relatively down-regulated in the more severe forms.	[73]
2020	To validate the use of a microRNA panel as a noninvasive diagnostic method for detecting endometriosis[10].	Prospective study 100 Laparoscopy	miR-125b-5p, miR-150-5p, miR-342-3p, and miR-451a, miR-3613-5p and let-7b	Upregulated: miR-125b-5p, miR-150-5p, miR-342-3p, and miR-451a. Down regulated: miR-3613-5p and let-7b.	This is the first report showing that microRNA biomarkers can reliably differentiate between endometriosis and other gynecological pathologies, with an area under the curve >0.9 across 2 independent studies.	[10]
2022	Analyzed miRNA expression in saliva of women with and without endometriosis using a FireFly custom multiplex	Case control 34 Laparoscopy	hsa-mir-135a	A significant upregulation of plasma hsa-mir-135a was only observed in the secretory phase, although the proliferative phase showed a similar trend.	hsa-mir-135a was expressed significantly higher in the saliva of women with endometriosis, independent of disease stage and menstrual cycle phase. Also confirmed that hsa-mir-135a showed	[74]

 doi:10.20944/preprints202404.1261.v1

circulating miRNA assay[75].	significantly elevated expression in the plasma of endometriosis patients.

2.3.3. Management (Treatment and Prognosis)

Endometriosis management is multifaceted, encompassing pain relief, halting disease progression, and improving fertility outcomes where applicable. The potential role of miRNA profiles in guiding these management strategies cannot be underestimated.

Braza-Boïls et al. 2014 studied miRNA expression's effect on endometriotic lesions. They found reduced miRNAs (miR-202-3p, miR-424-5p, miR-449b-3p, miR-556-3p) in endometrium and increased VEGF-A and uPA proteins. Ovarian endometrioma tissues showed higher PAI-1 and TSP-1. Altered miRNA expression might influence endometriosis pathogenesis due to changes in angiogenic and fibrinolytic factors[21].

In the study by Wu et al. 2020, researchers investigated exosomal RNA profiles in endometriosis using primary endometrial stromal cells from different groups. They identified differentially expressed exosomal lncRNAs, miRNAs, and miRNAs associated with endometriosis. Co-expression and competing endogenous RNA (ceRNA) networks were constructed to understand their interactions. This study provides insights into the molecular mechanisms underlying endometriosis and offers potential for identifying biomarkers and therapeutic targets[75].

Dong et al. (2020) studied circ_0007331's role in endometriosis. They observed higher circ_0007331 levels in endometrial cells from endometriosis patients. Its inhibition reduced cell growth and invasion. Circ_0007331 influenced cells by interacting with miR-200c-3p, affecting HIF-1α. This interaction slowed endometriosis progression, suggesting circ_0007331 as a potential treatment target[76].

The study by Gu et al. (2020) aimed to investigate miRNA expressions in patients with ovarian endometriosis and healthy controls using microarray analysis. The diagnostic potential of these downregulated miRNAs was assessed using ROC curve analysis, and hsa-let-7i-5p showed the highest AUC (0.900) for diagnosing ovarian endometriosis. The study suggested that these differentially expressed miRNAs could serve as biomarkers and potential therapeutic targets for ovarian endometriosis[77].

In a 2021 case-control study, Pokrovenko et al. investigated microRNA let-7 and mir-9 as potential non-invasive biomarkers for diagnosing and treating external genital endometriosis. The study found significant differences in let-7 expression between groups, while mir-9 did not show significant differences. In comparison to CA-125, let-7 exhibited better specificity for distinguishing patients with and without endometriosis. Let-7 emerged as a promising diagnostic marker, prompting the researchers to plan an endometriosis diagnostic and treatment algorithm[78].

In the study by Bendifallah et al. (2022), the aim was to establish a saliva-based diagnostic miRNA signature for endometriosis. The diagnostic performance of this miRNA signature was assessed, yielding a sensitivity of 96.7%, specificity of 100%, and an impressive AUC of 98.3%. The study emphasized the potential of the saliva-based diagnostic approach. The non-invasive nature and the possibility of repeated sampling make it promising for early identification and improved management of endometriosis[79].

In a 2022 study, Wu et al. explored the link between serum exosomal miRNA expression and endometriosis severity. They found 26 up-regulated and 19 down-regulated miRNAs in endometriosis patients. These miRNAs, validated with qRT-PCR, were associated with cellular processes and signaling pathways. The results highlight miRNA's role in understanding and potentially diagnosing and treating endometriosis[80].

In a 2022 study, Dabi et al. analyzed miRNA expression in endometriosis patients. They focused on 57 miRNAs with AUC values ≥ 0.6, including up-regulated ones like miR-6502-5p and down-regulated ones such as miR-3137. Interestingly, 20 miRNAs, including some from the miRNA-548

 doi:10.20944/preprints202404.1261.v1

family, were newly linked to endometriosis. The research connected these miRNAs to key endometriosis-related pathways[81].

In a 2022 study, Neuhausser et al. examined serum miRNA levels in women with endometriosis during the early follicular phase. They found 18 up-regulated miRNAs and one down-regulated, hsa-miR-34c-3p. This down-regulated miRNA, when modified in vitro, reduced endometrial cell growth. The findings suggest these miRNAs, especially hsa-miR-34c-3p, could be diagnostic biomarkers or treatment targets for endometriosis[82].

Table 7. Studies related to role of miRNAs in managing endometriosis.

Year	Objective	Type of study Size of sample Diagnosis method	miRNA type	Main result	Main outcome	Ref
2014	Could an aberrant microRNA (miRNA) expression profile be responsible for the changes in the angiogenic and fibrinolytic states observed in endometriotic lesions?	Case control 83 Laparoscopy	miR-202-3p, miR-424-5p, miR-449b-3p and miR-556-3p	Patient endometrial tissue showed significantly lower levels of miR-202-3p, miR-424-5p, miR-449b-3p and miR-556-3p, and higher levels of VEGF-A and uPA than healthy (control) endometrium.	Differences in miRNA levels could modulate the expression of VEGF-A and TSP-1, which may play an important role in the pathogenesis of endometriosis. The higher angiogenic and proteolytic activities observed in eutopic endometrium from patients might facilitate the implantation of endometrial cells at ectopic sites.	[21]
2020	To investigate exosomal RNAs (long noncoding RNAs (lncRNAs), microRNAs (miRNAs) and messenger RNAs (miRNAs)) profiling and their related networks in endometriosis (EMs).	Case control 30 Laparoscopy	KRAS, RAB5B, BIRC2, TRAK1, PSMC5, MIB2, ATP6V1A, ADCY3, HMGCR, SMO, ATF2. RBM19 HERC3 MIB2 DMXL1 TRAK1 QKI PSMC5 DDX55 AGFG1 UBR4 PUS7L BIRC2 PSMD8 SKP1 WDR75 RAB38 DDX47 KRAS PSMD14 RAB12 PAPSS1 RAB5B OXR1	Overlapped differentially expressed 938 lncRNAs, 39 miRNAs and 1449 miRNAs were identified. 13 co-expression modules and 61 ceRNA networks were constructed.	Revealed exosomal lncRNA, miRNA and miRNA expression profiles in EMs, and identified EMs-associated exosomal co-expression networks and ceRNA networks. It is a novel and comprehensive research of EMs related RNAs.	[75]

 doi:10.20944/preprints202404.1261.v1

			ATP6V1A SMO ADCY3 ATF2 PRKACB MZT2A HMGCR MDC1 NAPB			
2020	Identified aberrant high expression of circ_0007331 in ectopic endometrial cells by comparing the endometrial samples from patients with and without endometriosis.	Case control 50 Laparoscopy	circ_0007331, MiR-200c-3p, HIF-1α	1. The expression of circ_0007331 is abnormally elevated in endometriosis. 2. Circ_0007331 knock-down results in decreased cell proliferation and invasion of the EE cells in endometriosis. 3. Circ_0007331 knock-down suppresses the proliferation of EE cells by down-regulating the expression of HIF-1α. 4. MiR-200c-3p is predicted to mediate the regulation of HIF-1α by circ_0007331. 5. Circ_0007331 affects the proliferation and invasion of EE cells by sponging miR-200c-3p to regulate HIF-1α. 6. Circ_0007331 knock-down suppresses the progression of endometriosis via miR-200c-3p/HIF-1α axis in vivo	Results show that circ_0007331 is abnormally highly expressed in patients with endometriosis. Both in vitro or in vivo, circ_0007331 knock-down suppresses the progression of endometriosis by inhibiting the proliferation and invasion of ectopic endometrial cells. Further mechanism studies confirmed that the molecular basis of this function is the circ_0007331/miR-200c-3p/HIF-1α axis.	[76]
2020	Analyzed the miRNA expressions in patients with ovarian endometriosis and healthy controls with microarray analysis to identify differentially expressed miRNAs.	Case control 60 Laparoscopy	hsa-let-7i-5p, hsa-let-7a-5p, hsa-let- 7b-5p, hsa-let-7d-5p, hsa-let-7f-5p, hsa-let-7g-5p, hsa-miR-199a-3p, hsa-miR-320a, hsa-miR-320b, hsa-miR-320c, hsa-miR-320d, hsa-miR-328-3p, hsa-miR-331-3p, hsa-miR-320e	hsa-let-7i-5p showed the highest area under the ROC curve (AUC) with a value of 0.900.	The identified 14 differentially expressed miRNAs could be potential biomarkers and therapeutic targets for the diagnosis and treatment of endometriosis.	[77]

 doi:10.20944/preprints202404.1261.v1

2021	To evaluate the possibility of using microrna let-7 and mir-9 as non-invasive biomarkers for the diagnosis and treatment of external genital endometriosis.	Case control 86 Laparoscopy	mir-9, let-7 microRNA	Showed that the difference in mir-9 miRNA between the groups with and without endometriosis, as well as between the groups with more clinically and histologically severe and mild endometriosis, was statistically insignificant. In addition, a significant difference was noted regarding let-7 microRNA between the groups with and without endometriosis, as well as between the groups with more clinically and histologically severe and mild endometriosis. Comparison with cancer antigen-125 (CA-125) showed that let-7 microRNA was a more specific test than CA-125.	Believed that the measurement of microRNA let-7 is promising for routine use in patients with endometriosis.	[78]
2022	To analyze the human miRNAome to define a saliva-based diagnostic miRNA signature for endometriosis.	Case control 200 Laparoscopy	Not mentioned	The respective sensitivity, specificity, and AUC for the diagnostic miRNA signature were 96.7%, 100%, and 98.3%.	Data support the use of a saliva-based diagnostic miRNA signature for endometriosis in the diagnosis care pathways after an external validation to confirm these results.	[79]
2022	Focused on examining the relationship between serum exosomal miRNA expression and the severity of endometriosis.	Case control 66 Laparoscopy	miR-26b-5p, miR-215-5p, and miR-6795-3p	qRT-PCR analysis verified the differential expression of three miRNAs, miR-26b-5p, miR-215-5p, and miR-6795-3p.	Further analysis indicated that these differentially expressed miRNAs in serum exosomes may be involved in the pathogenesis of endometriosis and are related to the severity and certain symptoms of endometriosis.	[80]
2022	To investigate functions and pathways associated with the various miRNAs differentially	Case control 200 Laparoscopy	miR-124-3p, miR-6502-5p; miR-515-5p; miR-548j-5p; miR-29b-1-5p;	Up-regulated: miR-6502-5p; miR-515-5p; miR-548j-5p; miR-29b-1-5p; miR-4748	Results provide evidence of the relation between the miRNA profiles of patients with	[81]

 doi:10.20944/preprints202404.1261.v1

	expressed in patients with endometriosis.		miR-4748, miR-3137 and miR-3168. miRNA–548 family	Down regulated: miR-3137 and miR-3168.	endometriosis and various signaling pathways implicated in its pathophysiology.	
2022	To identify early follicular phase micro ribonucleic acids (miRNAs) that are altered in serum of women with endometriosis.	Case control 45 Laparoscopy	hsa-miR-34c-3p	hsa-miR-34c-3p was significantly down-regulated in the follicular phase of patients with endometriosis.	These results support hsa-miR-34-3p as a potential therapeutic target in endometriosis.	[82]

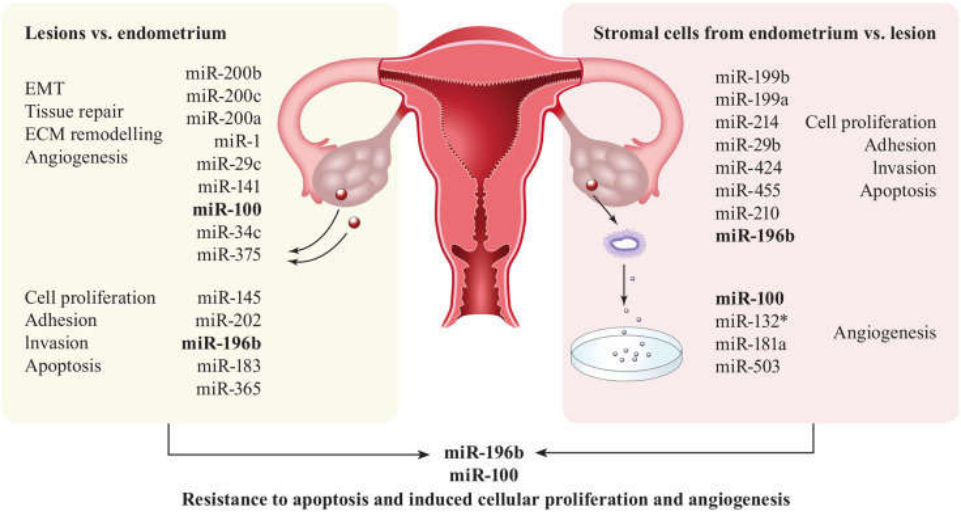

Figure 3. miRNA in endometriosis [83].

3.1. Discussion

Decades of research have significantly advanced our understanding of endometriosis, reflecting a journey marked by persistent innovation. This discussion aims to capture the essence of this evolution, spotlighting key milestones and the promising new directions emerging in the field.

3.2. Historical Context and the Role of CA-125

CA-125 has been crucial in endometriosis research, noted for its link to disease severity. Key studies by Colacurci et al. (1996), Kafali et al. (2004), and Socolov et al. (2011) highlighted its potential as a diagnostic and prognostic tool, contributing to the understanding of CA-125. While it's viewed with cautious optimism for its role in monitoring disease progression and informing treatment, its effectiveness is nuanced, affected by menstrual cycles and limitations in sensitivity and specificity. This underscores the complexity of CA-125 as a biomarker in the medical context.

3.3. MiRNAs: A Molecular Revolution

MiRNAs have become pivotal in understanding endometriosis, as shown by early research from Fassbender et al. (2012) and Jia et al. (2013), and later by Cosar et al. (2016) and Wang et al. (2016). These studies highlight miRNAs' varied roles, suggesting their strong potential as biomarkers. Yet, the complexity increases with findings like those from Haikalis ME et al. (2018), showing that miRNA profiles can vary significantly across different stages and types of endometriosis lesions, adding intricate layers to our knowledge.

 doi:10.20944/preprints202404.1261.v1

3.4. Interplay between CA-125 and miRNAs

The relationship between CA-125 and miRNAs is key in advancing endometriosis diagnosis, suggesting a synergistic approach for enhanced accuracy. CA-125 offers a broad perspective, while miRNAs provide detailed molecular insights. Research from Szubert et al. (2023), Zhao and Qu (2021), and Herranz-Blanco et al. (2023) indicates that combining clinical features with CA-125 and miRNA profiles could lead to more precise diagnoses, marking progress towards a more complete diagnostic method.

Yet, this progress is complex. Nisenblat et al. (2019) caution about miRNAs' varying specificity and sensitivity as standalone diagnostic tools. Additionally, the interaction of CA-125 with menstrual cycles and hormonal medications, highlighted by Moustafa et al. (2020) and Pateisky et al. (2018), adds complexity to the diagnosis. These factors emphasize the need for a deeper, nuanced understanding in the quest to fully exploit the diagnostic potential of these biomarkers.

3.5. A Multidimensional Perspective on Endometriosis

The review illustrates a journey that is both optimistic and complex, advocating for a comprehensive diagnostic approach in endometriosis. It emphasizes merging traditional biomarkers like CA-125 with newer ones like miRNAs. According to recent studies, such as those by Lin et al. (2023), this integration could improve diagnosis, prognosis, and treatment. It highlights the significance of combining molecular data with clinical insights for a holistic approach to patient care.

However, the narrative also acknowledges the limitations of these studies, including varying sample sizes, methods, and the intricate nature of endometriosis. It suggests future research focus on larger, more consistent cohorts, standardized protocols, and extended studies to fully determine the clinical usefulness of these biomarkers in the management of endometriosis.

4.1. Conclusion

The story of understanding endometriosis has unfolded over years of dedicated research, revealing the condition's complex nature. This review has journeyed through the detailed world of molecules and trusted biomarkers, helping us understand the pathophysiology, diagnosis, and potential therapeutic strategies of endometriosis.

4.2. miRNAs: The Molecular Narrators of Endometriosis

In this narrative, miRNAs are like lead characters, guiding the complex gene activity in endometriosis. Studies by Braza-Boïls et al. (2014) and Haikalis ME et al. (2018) reveal different miRNA patterns in the disease, showing us the varied nature of endometriosis. The work of researchers like Bendifallah et al. (2022), Xu et al. (2018), and Nisenblat et al. (2019) further highlights how these tiny molecules could help us diagnose and treat the condition more effectively, signaling a shift towards more customized care in endometriosis.

4.3. CA-125: The Enduring Biomarker

Alongside miRNAs, the story of CA-125 as a trusted biomarker continues. It's been a key tool in diagnosing and understanding endometriosis, with studies like those by Kafali et al. (2004) and Kitawaki et al. (2005) showing its value. But this narrative also notes that CA-125's effectiveness isn't the same for everyone, reminding us to look closer and understand its role better in each unique case of diagnosis.

4.4. Synergistic Perspectives

The review shows that combining miRNA expression profiles with CA-125 levels enhances our understanding of endometriosis. This approach, merging molecular insights with established biomarkers, is crucial for future progress in diagnosis, patient classification, and tailored treatment. It underscores the importance of continued research to validate and develop these diagnostic and therapeutic methods.

 doi:10.20944/preprints202404.1261.v1

In conclusion, the review indicates that advancing endometriosis care will involve validating and systematically integrating these biomarkers into healthcare, ensuring that current research achievements become standard practice in future healthcare.

4.5. Further Investigations

The narrative calls for further research with diverse patient groups and settings to validate findings on endometriosis. It suggests detailed studies on miRNAs and developing combined biomarker panels, including miRNAs and CA-125, for better diagnosis and treatment. Emphasizing clinical collaboration, it envisions personalized treatments based on these biomarkers. It also recommends longitudinal studies and using bioinformatics to deepen understanding of endometriosis progression.

References

1. Zondervan KT, Becker CM, Missmer SA. Endometriosis. N Engl J Med. 2020;382(13):1244-56.
2. Taylor HS, Kotlyar AM, Flores VA. Endometriosis is a chronic systemic disease: clinical challenges and novel innovations. Lancet. 2021;397(10276):839-52.
3. Smolarz B, Szyłło K, Romanowicz H. Endometriosis: Epidemiology, Classification, Pathogenesis, Treatment and Genetics (Review of Literature). International Journal of Molecular Sciences. 2021;22(19):10554.
4. Surrey E, Soliman AM, Trenz H, Blauer-Peterson C, Sluis A. Impact of Endometriosis Diagnostic Delays on Healthcare Resource Utilization and Costs. Adv Ther. 2020;37(3):1087-99.
5. Ghai V, Jan H, Shakir F, Haines P, Kent A. Diagnostic delay for superficial and deep endometriosis in the United Kingdom. J Obstet Gynaecol. 2020;40(1):83-9.
6. Moss KM, Doust J, Homer H, Rowlands IJ, Hockey R, Mishra GD. Delayed diagnosis of endometriosis disadvantages women in ART: a retrospective population linked data study. Human Reproduction. 2021;36(12):3074-82.
7. Warzecha D, Szymusik I, Wielgos M, Pietrzak B. The Impact of Endometriosis on the Quality of Life and the Incidence of Depression-A Cohort Study. Int J Environ Res Public Health. 2020;17(10).
8. Kamath MS, Subramanian V, Antonisamy B, Sunkara SK. Endometriosis and oocyte quality: an analysis of 13614 donor oocyte recipient and autologous IVF cycles. Hum Reprod Open. 2022;2022(3):hoac025.
9. Al Shukri M, Al Riyami AS, Al Ghafri W, Gowri V. Are There Predictors of Early Diagnosis of Endometriosis Based on Clinical Profile? A Retrospective Study. Oman Med J. 2023;38(1):e458.
10. Moustafa S, Burn M, Mamillapalli R, Nematian S, Flores V, Taylor HS. Accurate diagnosis of endometriosis using serum microRNAs. American Journal of Obstetrics and Gynecology. 2020;223(4):557.e1-.e11.
11. Kimber-Trojnar Ż, Pilszyk A, Niebrzydowska M, Pilszyk Z, Ruszała M, Leszczyńska-Gorzelak B. The Potential of Non-Invasive Biomarkers for Early Diagnosis of Asymptomatic Patients with Endometriosis. Journal of Clinical Medicine [Internet]. 2021; 10(13).
12. Nisenblat V, Bossuyt PM, Shaikh R, Farquhar C, Jordan V, Scheffers CS, et al. Blood biomarkers for the non-invasive diagnosis of endometriosis. Cochrane Database Syst Rev. 2016;2016(5):Cd012179.
13. Tian Z, Chang XH, Zhao Y, Zhu HL. Current biomarkers for the detection of endometriosis. Chin Med J (Engl). 2020;133(19):2346-52.
14. Anastasiu CV, Moga MA, Elena Neculau A, Bălan A, Scârneciu I, Dragomir RM, et al. Biomarkers for the Noninvasive Diagnosis of Endometriosis: State of the Art and Future Perspectives. Int J Mol Sci. 2020;21(5).
15. Chen Y, Pan M, Zuo Y, Yang B, Wang S. Research progress of CA125 in endometriosis: Teaching an old dog new tricks. Gynecology and Obstetrics Clinical Medicine. 2022;2(4):191-8.
16. Azam INA, Wahab NA, Mokhtar MH, Shafiee MN, Mokhtar NM. Roles of microRNAs in Regulating Apoptosis in the Pathogenesis of Endometriosis. Life. 2022;12(9):1321.
17. Bjorkman S, Taylor HS. MicroRNAs in endometriosis: biological function and emerging biomarker candidates†. Biol Reprod. 2019;100(5):1135-46.
18. Bendifallah S, Dabi Y, Suisse S, Jornea L, Bouteiller D, Touboul C, et al. MicroRNome analysis generates a blood-based signature for endometriosis. Sci Rep. 2022;12(1):4051.
19. Papari E, Noruzinia M, Kashani L, Foster WG. Identification of candidate microRNA markers of endometriosis with the use of next-generation sequencing and quantitative real-time polymerase chain reaction. Fertility and Sterility. 2020;113(6):1232-41.
20. Bendifallah S, Dabi Y, Suisse S, Delbos L, Spiers A, Poilblanc M, et al. Validation of a Salivary miRNA Signature of Endometriosis — Interim Data. NEJM Evidence. 2023;2(7):EVIDoa2200282.
21. Braza-Boïls A, Marí-Alexandre J, Gilabert J, Sánchez-Izquierdo D, España F, Estellés A, et al. MicroRNA expression profile in endometriosis: its relation to angiogenesis and fibrinolytic factors. Hum Reprod. 2014;29(5):978-88.

 doi:10.20944/preprints202404.1261.v1

22. Oliveira MAP, Raymundo TS, Soares LC, Pereira TRD, Demôro AVE. How to Use CA-125 More Effectively in the Diagnosis of Deep Endometriosis. Biomed Res Int. 2017;2017:9857196.
23. Kitawaki J, Ishihara H, Koshiba H, Kiyomizu M, Teramoto M, Kitaoka Y, et al. Usefulness and limits of CA-125 in diagnosis of endometriosis without associated ovarian endometriomas. Hum Reprod. 2005;20(7):1999-2003.
24. Colacurci N, Fortunato N, De Franciscis P, Fratta M, Cioffi M, Zarcone R, et al. Serum and peritoneal CA-125 levels as diagnostic test for endometriosis. Eur J Obstet Gynecol Reprod Biol. 1996;66(1):41-3.
25. Kafali H, Artuc H, Demir N. Use of CA125 fluctuation during the menstrual cycle as a tool in the clinical diagnosis of endometriosis; a preliminary report. Eur J Obstet Gynecol Reprod Biol. 2004;116(1):85-8.
26. Agic A, Djalali S, Wolfler MM, Halis G, Diedrich K, Hornung D. Combination of CCR1 mRNA, MCP1, and CA125 Measurements in Peripheral Blood as a Diagnostic Test for Endometriosis. Reproductive Sciences. 2008;15(9):906-11.
27. Mihalyi A, Gevaert O, Kyama CM, Simsa P, Pochet N, De Smet F, et al. Non-invasive diagnosis of endometriosis based on a combined analysis of six plasma biomarkers. Hum Reprod. 2010;25(3):654-64.
28. Socolov R, Butureanu S, Angioni S, Sindilar A, Boiculese L, Cozma L, et al. The value of serological markers in the diagnosis and prognosis of endometriosis: a prospective case-control study. Eur J Obstet Gynecol Reprod Biol. 2011;154(2):215-7.
29. Vodolazkaia A, El-Aalamat Y, Popovic D, Mihalyi A, Bossuyt X, Kyama CM, et al. Evaluation of a panel of 28 biomarkers for the non-invasive diagnosis of endometriosis. Hum Reprod. 2012;27(9):2698-711.
30. Zhu H, Lei H, Wang Q, Fu J, Song Y, Shen L, et al. Serum carcinogenic antigen (CA)-125 and CA 19-9 combining pain score in the diagnosis of pelvic endometriosis in infertile women. Clin Exp Obstet Gynecol. 2016;43(6):826-9.
31. Irungu S, Mavrelos D, Worthington J, Blyuss O, Saridogan E, Timms JF. Discovery of non-invasive biomarkers for the diagnosis of endometriosis. Clin Proteomics. 2019;16:14.
32. Tang T, Lai H, Huang X, Gu L, Shi H. Application of serum markers in diagnosis and staging of ovarian endometriosis. J Obstet Gynaecol Res. 2021;47(4):1441-50.
33. Guralp O, Kaya B, Tüten N, Kucur M, Malik E, Tüten A. Non-invasive diagnosis of endometriosis and moderate-severe endometriosis with serum CA125, endocan, YKL-40, and copeptin quadruple panel. J Obstet Gynaecol. 2021;41(6):927-32.
34. Chen T, Wei JL, Leng T, Gao F, Hou SY. The diagnostic value of the combination of hemoglobin, CA199, CA125, and HE4 in endometriosis. J Clin Lab Anal. 2021;35(9):e23947.
35. Szubert M, Rycerz A, Wilczyński JR. How to Improve Non-Invasive Diagnosis of Endometriosis with Advanced Statistical Methods. Medicina (Kaunas). 2023;59(3).
36. Micu R, Gaia-Oltean AMI, Budişan L, Braicu C, Irimie A, Berindan-Neagoe I. The added value of CA125, HE4, and CA72-4 as markers for ovarian endometriosis diagnosis. Rom J Morphol Embryol. 2023;64(2):159-64.
37. Kovalak EE, Karacan T, Zengi O, Karabay Akgül Ö, Özyürek Ş E, Güraslan H. Evaluation of new biomarkers in stage III and IV endometriosis. Gynecol Endocrinol. 2023;39(1):2217290.
38. Hornstein MD, Harlow BL, Thomas PP, Check JH. Use of a new CA 125 assay in the diagnosis of endometriosis. Hum Reprod. 1995;10(4):932-4.
39. Rosa ESAC, Rosa ESJC, Ferriani RA. Serum CA-125 in the diagnosis of endometriosis. Int J Gynaecol Obstet. 2007;96(3):206-7.
40. Szubert M, Suzin J, Wierzbowski T, Kowalczyk-Amico K. CA-125 concentration in serum and peritoneal fluid in patients with endometriosis - preliminary results. Arch Med Sci. 2012;8(3):504-8.
41. Karimi-Zarchi M, Dehshiri-Zadeh N, Sekhavat L, Nosouhi F. Correlation of CA-125 serum level and clinico-pathological characteristic of patients with endometriosis. Int J Reprod Biomed. 2016;14(11):713-8.
42. Moloney MD, Thornton JG, Cooper EH. Serum CA 125 antigen levels and disease severity in patients with endometriosis. Obstet Gynecol. 1989;73(5 Pt 1):767-9.
43. O'Shaughnessy A, Check JH, Nowroozi K, Lurie D. CA 125 levels measured in different phases of the menstrual cycle in screening for endometriosis. Obstet Gynecol. 1993;81(1):99-103.
44. Amaral VF, Ferriani RA, Sá MF, Nogueira AA, Rosa e Silva JC, Rosa e Silva AC, et al. Positive correlation between serum and peritoneal fluid CA-125 levels in women with pelvic endometriosis. Sao Paulo Med J. 2006;124(4):223-7.
45. Maiorana A, Cicerone C, Niceta M, Alio L. Evaluation of serum CA 125 levels in patients with pelvic pain related to endometriosis. Int J Biol Markers. 2007;22(3):200-2.
46. Ozhan E, Kokcu A, Yanik K, Gunaydin M. Investigation of diagnostic potentials of nine different biomarkers in endometriosis. Eur J Obstet Gynecol Reprod Biol. 2014;178:128-33.
47. Santulli P, Streuli I, Melonio I, Marcellin L, M'Baye M, Bititi A, et al. Increased serum cancer antigen-125 is a marker for severity of deep endometriosis. J Minim Invasive Gynecol. 2015;22(2):275-84.

48. Knific T, Vouk K, Vogler A, Osredkar J, Gstöttner M, Wenzl R, et al. Models including serum CA-125, BMI, cyst pathology, dysmenorrhea or dyspareunia for diagnosis of endometriosis. Biomark Med. 2018;12(7):737-47.
49. Sasamoto N, DePari M, Vitonis AF, Laufer MR, Missmer SA, Shafrir AL, et al. Evaluation of CA125 in relation to pain symptoms among adolescents and young adult women with and without surgically-confirmed endometriosis. PLoS One. 2020;15(8):e0238043.
50. Zhao K, Qu P. Noninvasive evaluation of ovarian endometriosis: a single-center experience. Ann Palliat Med. 2021;10(4):4728-35.
51. Herranz-Blanco B, Daoud E, Viganò P, García-Velasco JA, Colli E. Development and Validation of an Endometriosis Diagnostic Method Based on Serum Biomarkers and Clinical Variables. Biomolecules. 2023;13(7).
52. Pittaway DE, Fayez JA. The use of CA-125 in the diagnosis and management of endometriosis. Fertil Steril. 1986;46(5):790-5.
53. Koninckx PR, Meuleman C, Oosterlynck D, Cornillie FJ. Diagnosis of deep endometriosis by clinical examination during menstruation and plasma CA-125 concentration. Fertil Steril. 1996;65(2):280-7.
54. Chen FP, Soong YK, Lee N, Lo SK. The use of serum CA-125 as a marker for endometriosis in patients with dysmenorrhea for monitoring therapy and for recurrence of endometriosis. Acta Obstet Gynecol Scand. 1998;77(6):665-70.
55. Kimber-Trojnar, Żaneta et al. "The Potential of Non-Invasive Biomarkers for Early Diagnosis of Asymptomatic Patients with Endometriosis." Journal of clinical medicine vol. 10,13 2762. 23 Jun. 2021, doi:10.3390/jcm10132762
56. Fassbender A, Verbeeck N, Börnigen D, Kyama CM, Bokor A, Vodolazkaia A, et al. Combined mRNA microarray and proteomic analysis of eutopic endometrium of women with and without endometriosis. Hum Reprod. 2012;27(7):2020-9.
57. Jia SZ, Yang Y, Lang J, Sun P, Leng J. Plasma miR-17-5p, miR-20a and miR-22 are down-regulated in women with endometriosis. Hum Reprod. 2013;28(2):322-30.
58. Cosar E, Mamillapalli R, Ersoy GS, Cho S, Seifer B, Taylor HS. Serum microRNAs as diagnostic markers of endometriosis: a comprehensive array-based analysis. Fertil Steril. 2016;106(2):402-9.
59. Wang L, Huang W, Ren C, Zhao M, Jiang X, Fang X, et al. Analysis of Serum microRNA Profile by Solexa Sequencing in Women With Endometriosis. Reprod Sci. 2016;23(10):1359-70.
60. Xu XX, Jia SZ, Dai Y, Zhang JJ, Li XY, Shi JH, et al. Identification of Circular RNAs as a Novel Biomarker for Ovarian Endometriosis. Chin Med J (Engl). 2018;131(5):559-66.
61. Haikalis ME, Wessels JM, Leyland NA, Agarwal SK, Foster WG. MicroRNA expression pattern differs depending on endometriosis lesion type. Biol Reprod. 2018;98(5):623-33.
62. Pateisky P, Pils D, Szabo L, Kuessel L, Husslein H, Schmitz A, et al. hsa-miRNA-154-5p expression in plasma of endometriosis patients is a potential diagnostic marker for the disease. Reprod Biomed Online. 2018;37(4):449-66.
63. Nisenblat V, Sharkey DJ, Wang Z, Evans SF, Healey M, Ohlsson Teague EMC, et al. Plasma miRNAs Display Limited Potential as Diagnostic Tools for Endometriosis. J Clin Endocrinol Metab. 2019;104(6):1999-2022.
64. Vanhie A, O D, Peterse D, Beckers A, Cuéllar A, Fassbender A, et al. Plasma miRNAs as biomarkers for endometriosis. Hum Reprod. 2019;34(9):1650-60.
65. Zhang L, Li H, Yuan M, Li D, Sun C, Wang G. Serum Exosomal MicroRNAs as Potential Circulating Biomarkers for Endometriosis. Dis Markers. 2020;2020:2456340.
66. Hossein Razi M, Eftekhar M, Ghasemi N, Hasan Sheikhha M, Dehghani Firoozabadi A. Expression levels of circulatory mir-185-5p, vascular endothelial growth factor, and platelet-derived growth factor target genes in endometriosis. Int J Reprod Biomed. 2020;18(5):347-58.
67. Borisov E, Knyazeva M, Novak V, Zabegina L, Prisyazhnaya T, Karizkiy A, et al. Analysis of Reciprocally Dysregulated miRNAs in Eutopic Endometrium Is a Promising Approach for Low Invasive Diagnostics of Adenomyosis. Diagnostics (Basel). 2020;10(10).
68. Misir S, Hepokur C, Oksasoglu B, Yildiz C, Yanik A, Aliyazicioglu Y. Circulating serum miR-200c and miR-34a-5p as diagnostic biomarkers for endometriosis. J Gynecol Obstet Hum Reprod. 2021;50(4):102092.
69. Zafari N, Tarafdari AM, Izadi P, Noruzinia M, Yekaninejad MS, Bahramy A, et al. A Panel of Plasma miRNAs 199b-3p, 224-5p and Let-7d-3p as Non-Invasive Diagnostic Biomarkers for Endometriosis. Reprod Sci. 2021;28(4):991-9.
70. Kumari P, Sharma I, Saha SC, Srinivasan R, Bhardwaj P. Role of serum microRNAs as biomarkers for endometriosis, endometrioid carcinoma of ovary & endometrioid endometrial cancer. Indian J Med Res. 2022;156(3):516-23.
71. Abo C, Biquard L, Girardet L, Chouzenoux S, Just PA, Chapron C, et al. Unbiased In Silico Analysis of Gene Expression Pinpoints Circulating miRNAs Targeting KIAA1324, a New Gene Drastically Downregulated in Ovarian Endometriosis. Biomedicines. 2022;10(9).

 doi:10.20944/preprints202404.1261.v1

72. Lin C, Zeng S, Li M. miR-424-5p combined with miR-17-5p has high diagnostic efficacy for endometriosis. Arch Gynecol Obstet. 2023;307(1):169-77.
73. Bashti O, Noruzinia M, Garshasbi M, Abtahi M. miR-31 and miR-145 as Potential Non-Invasive Regulatory Biomarkers in Patients with Endometriosis. Cell J. 2018;20(1):84-9.
74. Perricos A, Proestling K, Husslein H, Kuessel L, Hudson QJ, Wenzl R, et al. Hsa-mir-135a Shows Potential as A Putative Diagnostic Biomarker in Saliva and Plasma for Endometriosis. Biomolecules. 2022;12(8).
75. Wu J, Huang H, Huang W, Wang L, Xia X, Fang X. Analysis of exosomal lncRNA, miRNA and mRNA expression profiles and ceRNA network construction in endometriosis. Epigenomics. 2020;12(14):1193-213.
76. Dong L, Zhang L, Liu H, Xie M, Gao J, Zhou X, et al. Circ_0007331 knock-down suppresses the progression of endometriosis via miR-200c-3p/HiF-1α axis. J Cell Mol Med. 2020;24(21):12656-66.
77. Gu CL, Zhang Z, Fan WS, Li LA, Ye MX, Zhang Q, et al. Identification of MicroRNAs as Potential Biomarkers in Ovarian Endometriosis. Reprod Sci. 2020;27(9):1715-23.
78. Pokrovenko DA, Vozniuk V, Medvediev MV. MicroRNA let-7: A promising non-invasive biomarker for diagnosing and treating external genital endometriosis. Turk J Obstet Gynecol. 2021;18(4):291-7.
79. Bendifallah S, Suisse S, Puchar A, Delbos L, Poilblanc M, Descamps P, et al. Salivary MicroRNA Signature for Diagnosis of Endometriosis. J Clin Med. 2022;11(3).
80. Wu Y, Yuan W, Ding H, Wu X. Serum exosomal miRNA from endometriosis patients correlates with disease severity. Arch Gynecol Obstet. 2022;305(1):117-27.
81. Dabi Y, Suisse S, Jornea L, Bouteiller D, Touboul C, Puchar A, et al. Clues for Improving the Pathophysiology Knowledge for Endometriosis Using Serum Micro-RNA Expression. Diagnostics (Basel). 2022;12(1).
82. Neuhausser WM, Faure-Kumar E, Mahurkar-Joshi S, Iliopoulos D, Sakkas D. Identification of miR-34-3p as a candidate follicular phase serum marker for endometriosis: a pilot study. F S Sci. 2022;3(3):269-78.
83. Saare, Merli et al. “Challenges in endometriosis miRNA studies - From tissue heterogeneity to disease specific miRNAs.” Biochimica et biophysica acta. Molecular basis of disease vol. 1863,9 (2017): 2282-2292. doi:10.1016/j.bbadis.2017.06.018