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Original Article

Diagnostic and prognostic value of coagulation-related factors in endometriosis

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Abstract: Objective: To investigate the diagnostic and prognostic values of coagulation-related factors in endometriosis and explore the risk factors for the recurrence after treatment. Methods: Forty-eight patients with endometriosis admitted to the Second Hospital of Shandong University from June 2018 to March 2020 were enrolled in this retrospective analysis, along with 45 of subjects who underwent physical examination as healthy controls. Endometriosis patients were seen as research group (RG) and healthy subjects were considered as control group (CG). Patients in RG were treated with mifepristone. The coagulation-related factors, including activated partial thromboplastin time (APTT), prothrombin time (PT), thrombin time (TT) and fibrinogen (FIB), were measured, and their diagnostic value in endometriosis as well as their correlation with the mean cyst diameter and the clinical outcomes of patients were analyzed. Results: APTT, PT, and TT were shortened whereas FIB was increased in the RG as compared with CG (all $P<0.05$). ROC analysis revealed that coagulation-related factors had an excellent diagnostic value in endometriosis ($P<0.05$). Subsequently, we found that patients in the RG had higher APTT, PT, and TT and lower FIB after treatment ($P<0.05$). Moreover, APTT, PT, and TT were prolonged while FIB was decreased in patients with good prognosis as compare with those with poor prognosis (all $P<0.05$). Correlation analysis denoted that APTT, PT, and TT were negatively correlated with mean cyst diameter, whereas FIB was positively correlated with it ($P<0.05$). The abortion times and post-treatment TT were independent risk factors for recurrence within 2 years. Conclusion: Coagulation-related factors are associated with endometriosis. They can effectively assess the treatment efficacy of mifepristone, which has great prospects for future clinical application.

Keywords: Coagulation-related factors, endometriosis, mifepristone, cyst size

Introduction

Endometriosis is a chronic disease that affects approximately 10% of women of reproductive age, with clinical features of dysmenorrhea, dysuria, dyspareunia, and infertility etc. [1, 2]. Although endometriosis is a benign disease, its lesions are widespread, diverse, highly aggressive and recurrent, similar to malignancies in nature; Even after surgical treatment, it still has a high recurrence rate, which seriously affects the physical and mental health of patients [3-5]. The current clinical diagnosis of endometriosis is often based on patients' symptoms, gynecological examination, and ultrasonography, but most patients in early stage do not have specific and typical symptoms. So, early diagnosis is difficult [6]. The gold standard for diagnosis, however, is laparoscopic exploratory surgery, which is invasive [7]. Therefore, non-invasive hematological, biological, or genetic markers have been actively sought to assist in the preoperative diagnosis of endometriosis.

As a kind of infertility, endometriosis is relevant to blood coagulation system. Ding et al. [8] mentioned that endometriosis patients have higher platelet activation and aggregation rates than normal women, indicating that endometriosis, as a hypercoagulable disease, is associated with platelets, and anticoagulation therapy can improve patients' symptoms. There have been few studies on the value of coagulation-related factors in the diagnosis and prognosis of endometriosis. Clinically, the treatment of endometriosis is primarily surgery combined with postoperative medication, which is based on sex hormone drugs [9, 10]. Mifepristone

treatment of endometriosis not only improves the clinical symptoms of patients, but also improves sex hormone levels, which promotes recovery [11].

Thus, in this study, we investigated the diagnostic value of coagulation-related factors in endometriosis and explored their relationship with the prognosis of patients to provide new ideas for early non-invasive diagnosis.

Data and methods

Patient data

In this retrospective study, the clinical data of 48 endometriosis patients and 45 healthy subjects who underwent physical examination in the Second Hospital of Shandong University from June 2018 to March 2020 were collected and analyzed. The endometriosis patients with an average age of (31.00±6.92) were seen as the research group (RG), while the healthy subjects with an average age of (29.56±6.38) were seen as the control group (CG). The RG received mifepristone treatment in the Second Hospital of Shandong University. This research was carried out after being approved by the ethics committee of the Second Hospital of Shandong University (IRB20180542). All patients had given their informed consent on the treatment regimen.

Inclusion and exclusion criteria

Inclusion criteria: Patients with complete clinical data; Patients who were diagnosed as endometriosis by laparoscopic surgery [12].

Exclusion criteria: Patients complicated with adenomyosis, uterine leiomyoma or pelvic inflammation; Patients allergic to therapeutic drugs; Patients in pregnancy or menopause; Patients with a birth plan during the diagnosis and treatment; Patients complicated with autoimmune diseases; Patients complicated with hematological diseases or blood coagulation disorders; Patients who had used corticosteroids and contraceptives within three months before treatment.

Mode of treatment

On the first day of the first menstruation after laparoscopic surgery, the RG was treated with mifepristone tablets (manufacturer: Beijing Zizhu Pharmaceutical Co., Ltd., approval number: H10950003, specification: 25 mg/tablets) 12.5 mg orally, once a day for 6 months.

Efficacy evaluation

The treatment efficacy was classified in to four grades, namely cure, marked effective, effective and ineffective. Cured: After treatment, patients' clinical symptoms disappeared, and the physical signs returned to normal. Marked effective: After treatment, the clinical symptoms of patients were improved, and the rate of reduction in pelvic cyst size was more than 50%. Effective: After treatment, the clinical symptoms of patients were improved, and the rate was 30% to 49%. Ineffective: After treatment, there was no improvement in clinical symptoms or even worsened symptoms, and the rate was less than 30%.

Outcome measures

(1) The four indexes of prothrombin time (PT), thrombin time (TT), activated partial thromboplastin time (APTT) and fibrinogen (FIB) at admission were compared between the two groups. Venous blood (6 mL) was collected in the anticoagulant tube before and after treatment in the RG and during the physical examination in the CG. APTT, PT, TT and FIB were measured by Coatron3000 automatic hemagglutination analyzer (Micro Medical Devices, Germany). (2) The diagnostic value of coagulation factors APTT, PT, TT and FIB in endometriosis was assessed by ROC curve. (3) The relationship between coagulation-related factors APTT, PT, TT, FIB, and mean cyst diameter in the RG was assessed by Pearson correlation, and the cyst diameter was examined by B-ultrasound. (4) The changes of APTT, PT, TT and FIB in the RG were observed before and after mifepristone treatment. (5) The treatment efficacy in the RG was counted, and the changes of APTT, PT, TT and FIB were observed. (6) All patients were followed up for 2 years by telephone and outpatient reexamination, and the recurrence of patients within 2 years was counted. Multivariate analysis was performed to explore the independent risk factors for recurrence of patients.

Statistical methods

SPSS 20.0 software (Chicago SPSS Co., Ltd.) was used for statistical analysis. Continuous variables were expressed by mean ± standard

Table 1. Baseline data

	Research group (n=48)	Control group (n=45)	X^2/t	P
Age (year)	31.00±6.92	29.56±6.38	1.041	0.301
BMI (kg/m^2)	21.19±2.04	20.87±2.35	0.703	0.484
Gravida (n)			4.452	0.108
0	24	13		
1	19	24		
2	5	8		
Parity (n)			1.230	0.267
0	36 (75.00)	29 (64.44)		
1	12 (25.00)	16 (35.56)		
Abortion (n)			0.505	0.477
0	27 (56.25)	22 (48.89)		
1	21 (43.75)	23 (51.11)		
Marriage			0.796	0.672
Married	33	32		
Unmarried	8	9		
Divorce	7	4		
Stages				
Stage I	7			
Stage II	16			
Stage III	19			
Stage IV	6			
Mean cyst diameter (cm)	3.55±0.44			

BMI: Body Mass Index.

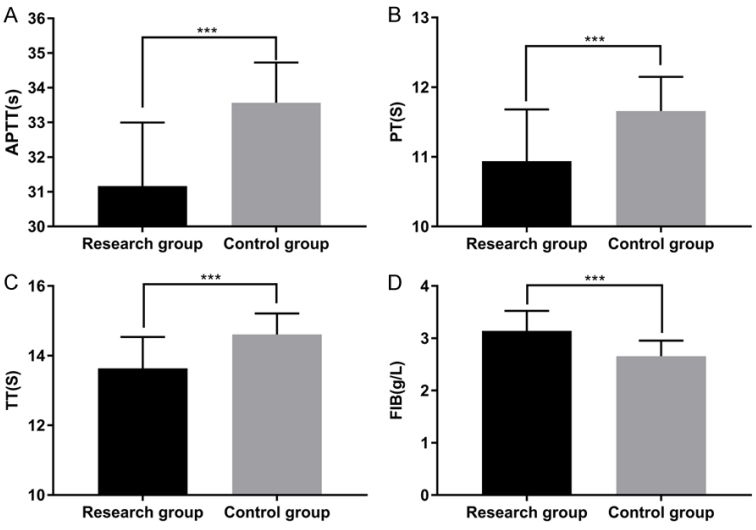

Figure 1. Comparison of coagulation-related factors between groups. A. Activated partial thromboplastin time (APTT) in the research group was lower than that in the control group (P<0.001). B. Prothrombin time (PT) in the research group was lower than that in the control group (P<0.001). C. Thrombin time (TT) in the research group was lower than that in the control group (P<0.001). D. Fibrinogen (FIB) in the research group was higher than that in the control group (P<0.001). Note: *** means P<0.001.

deviation. Independent t-test test was used for comparison between both groups, while paired t-test was used for comparison at different times in the same group, and the results were expressed by t. Classification variables were represented as the number of classification cases or percentages and detected using chi-square analysis. The diagnostic value of coagulation factors in endometriosis was tested by ROC curve. The independent risk factors for recurrence were analyzed by logistic regression. P<0.05 indicated statistical difference.

Results

Baseline data

It was found that there were no statistical differences in age, BMI, gravida, parity, abortion, and marriage status between groups (all P>0.05) (**Table 1**).

Comparison of coagulation-related factors between two groups

We collected the coagulation-related factors APTT, PT, TT and FIB, and found that APTT, PT and TT of the RG were significantly shortened while FIB was notably increased as compared with those of the CG (all P<0.05) (**Figure 1**).

Diagnostic value of coagulation factors in endometriosis

We assessed the diagnostic value of coagulation factors in endometriosis by drawing ROC curves. It turned out that the areas under the curve (AUCs) of APTT, PT, TT and FIB were 0.865 (95% CI: 0.792-0.937), 0.793 (95% CI: 0.704-

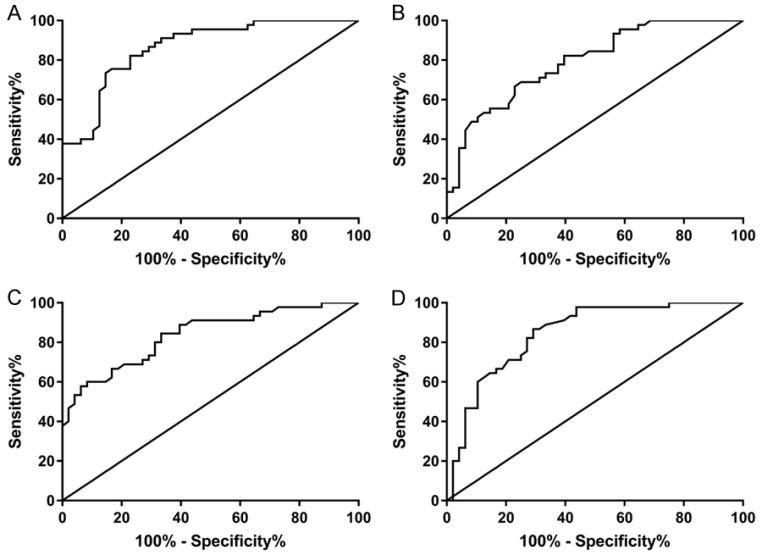

Figure 2. Diagnostic value of coagulation factors in endometriosis. A. The AUC of APTT in diagnosing endometriosis was 0.865 at the cut off value of 32.66, with the specificity and sensitivity of 77.08% and 82.22%, respectively. B. The AUC of PT in diagnosing endometriosis was 0.793 at the cut off value of 11.49, with the specificity and sensitivity of 77.08% and 66.67%, respectively. C. The AUC of TT in diagnosing endometriosis was 0.836 at the cut off value of 14.57, with the specificity and sensitivity of 91.67% and 60.00%, respectively. D. The AUC of FIB in diagnosing endometriosis was 0.846 at the cut off value of 3.01, with the specificity and sensitivity of 70.83% and 86.67%, respectively. Note: APTT: Activated Partial Thromboplastin Time; PT: Prothrombin Time; TT: Thrombin Time; FIB: Fibrinogen; AUC: Area Under Curve.

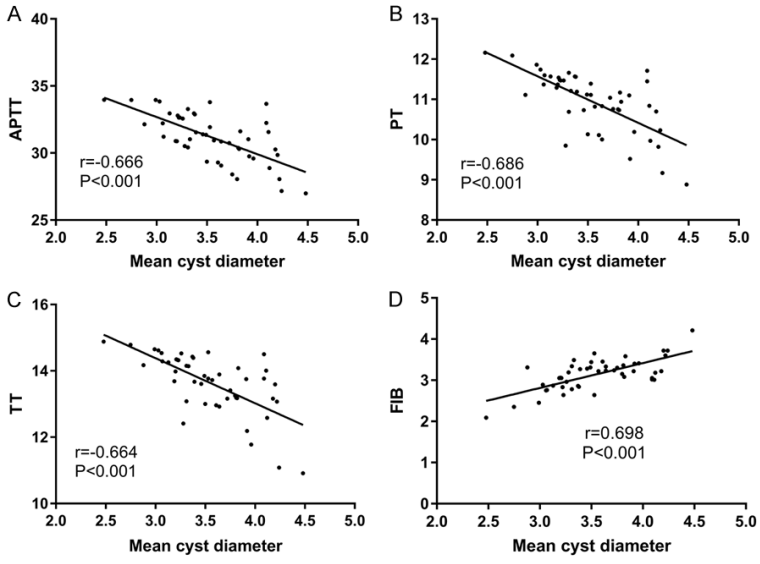

Figure 3. Relationship between coagulation-related factors and size of cysts in patients. A. APTT was negatively correlated with the mean cyst diameter (r=-0.666, P<0.001). B. PT was negatively correlated with the mean cyst diameter (r=-0.686, P<0.001). C. TT was negatively correlated with the mean cyst diameter (r=-0.664, P<0.001). D. FIB was positively correlated with the mean cyst diameter (r=0.698, P<0.001). APTT: Activated Partial Thromboplastin Time; PT: Prothrombin Time; TT: Thrombin Time; FIB: Fibrinogen.

0.882), 0.836 (95% CI: 0.756-0.917), and 0.846 (95% CI: 0.766-0.926), respectively (**Figure 2**).

Relationship between coagulation-related factors and size of cysts in patients

We assessed the relationship between the coagulation-related factors APTT, PT, TT, FIB, and the mean cyst diameter by Pearson correlation. We found that APTT, PT and TT were negatively correlated with the mean cyst diameter, while FIB was positively correlated with it (**Figure 3**).

Mifepristone's effects on coagulation-related factors

We observed the changes of coagulation-related factors in the RG before and after treatment with mifepristone, and the results showed that APTT, PT and TT in the RG were prolonged than those before treatment, while FIB was decreased (all P<0.05) (**Figure 4**).

Changes of coagulation-related factors in patients with different therapeutic effects

According to the efficacy of patients in the RG, 16 cases were cured, 14 were markedly effective, 9 were effective and 9 were ineffective. We discovered that APTT, PT and TT were prolonged (P<0.05) and FIB was decreased (P<0.05) in patients with better clinical efficacy (**Table 2**).

Univariate analysis of prognosis

The recurrence of patients in 2 years was counted, and the patients were divided into a

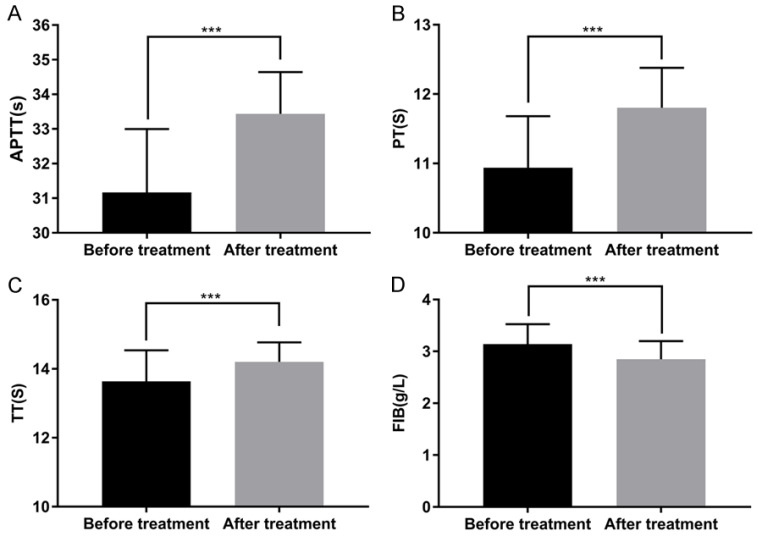

Figure 4. Mifepristone's effects on coagulation-related factors. A. APTT increased after the treatment with mifepristone (P<0.001). B. PT increased after the treatment with mifepristone (P<0.001). C. TT increased after the treatment with mifepristone (P<0.001). D. FIB decreased after the treatment with mifepristone (P<0.001). Note: *** means P<0.001. APTT: Activated Partial Thromboplastin Time; PT: Prothrombin Time; TT: Thrombin Time; FIB: Fibrinogen.

Table 2. Changes of coagulation-related factors in different therapeutic effects

	Cured (n=16)	Markedly effective (n=14)	Effective (n=9)	Ineffective (n=9)
APTT	34.51±1.06	33.37±0.54[a]	32.97±0.61[a]	32.11±1.03[a,b,c]
PT	12.25±0.45	11.97±0.34	11.49±0.39[a,b]	11.06±0.24[a,b,c]
TT	14.54±0.56	14.42±0.41	13.94±0.33[a,b]	13.54±0.20[a,b,c]
FIB	3.11±0.25	2.97±0.22	2.65±0.14[a,b]	2.39±0.25[a,b,c]

Note: APTT: Activated Partial Thromboplastin Time; PT: Prothrombin Time; TT: Thrombin Time; FIB: Fibrinogen; [a]denotes P<0.05 compared with cured cases; [b]denotes P<0.05 compared with markedly effective cases; [c]denotes P<0.05 compared with effective cases.

good prognosis group and a poor prognosis group according to whether there was a recurrence. It was found that a total of 12 patients had recurrence. There were differences in post-treatment APTT, PT and TT, stage, mean cyst diameter and abortion times between the good and poor prognosis groups (all P<0.05) (**Table 3**).

Multivariate analysis of prognosis

A binary logistic regression analysis was further conducted and found that the average cyst diameter, APTT and PT were not independent risk factors for the prognosis, while higher abortion times and lower TT were independent risk factors for recurrence (**Table 4**).

Discussion

With the increase of cesarean section rate, artificial abortion and uterine laparoscopy, the incidence of endometriosis is also increasing year by year [13-15]. About 60-80% of endometriosis patients suffer pelvic pain, and 30-40% develop infertility. So, timely diagnosis and treatment is critically needed [16]. In the process of clinical diagnosis and treatment, the specific treatment measures for endometriosis should proceed from its fundamental purpose, that is, relieving and controlling pain, meeting patients' fertility requirements, reducing or even removing lesions, and improving their prognosis and quality of life. In clinical practice, surgery and drug treatment are still the primary options [17-19].

Research has found that tissue factor (TF) increases in the lesion site and peritoneal fluid of endometriosis patients. TF can activate and combine coagulation factors and induce thrombin formation [20]. Our study compared the coagulation factor indexes APTT, PT, TT and FIB between endometriosis and healthy subjects, and found that APTT, PT and TT were shorter while FIB was higher in endometriosis patients, indicating that endometriosis patients have a higher blood coagulation. Both Wu et al. [21] and Vigand et al. [22] reported higher coagulation in endometriosis, which is similar to our study. Facing with the differences in coagulation indexes between groups, we then drew the ROC curve to test the diagnostic value of APTT, PT, TT, and FIB in endometriosis; The AUCs of the four were all >0.7, indicating that all the four indicators have good diagnostic value.

Table 3. Univariate analysis of variables affecting patients' prognosis

	Poor prognosis group (n=12)	Good prognosis group (n=36)	X^2/t	P
Age (year)	32.92±6.69	30.36±6.87	1.125	0.267
BMI (kg/m^2)	21.39±1.54	21.13±2.20	0.378	0.707
Gravida (n)			0.837	0.658
0	2 (16.66)	13 (36.11)		
1	7 (58.33)	24 (66.67)		
2	3 (25.00)	8 (22.22)		
Parity (n)			0.152	0.697
0	7 (58.33)	29 (64.44)		
1	5 (41.67)	16 (35.56)		
Abortion (n)			4.035	0.045
0	2 (16.67)	22 (48.89)		
1	10 (83.33)	23 (51.11)		
Marriage			1.105	0.575
Married	8 (66.67)	25 (69.44)		
Unmarried	3 (25.00)	5 (13.89)		
Divorce	1 (8.33)	6 (16.67)		
Stages			9.661	0.022
Stage I	0 (0.00)	7 (19.44)		
Stage II	2 (16.67)	14 (38.89)		
Stage III	6 (50.00)	13 (36.11)		
Stage IV	4 (33.33)	2 (5.56)		
Mean cyst diameter (cm)	3.78±0.37	3.47±0.44	2.262	0.028
APTT after treatment (s)	32.84±1.10	33.63±1.19	2.027	0.049
PT after treatment (s)	11.45±0.44	11.92±0.57	2.603	0.012
TT after treatment (s)	13.87±0.51	14.32±0.54	2.533	0.015
FIB after treatment (g/L)	3.00±0.22	2.80±0.36	1.808	0.077

Note: APTT: Activated Partial Thromboplastin Time; PT: Prothrombin Time; TT: Thrombin Time; FIB: Fibrinogen.

The cystic lesions of endometriosis are usually benign, but their impact on patients' reproductive function is obvious, and the characteristics of the cyst are relevant to the prognosis of the disease [23, 24]. We also found that coagulation-related factors APTT, PT, TT, and FIB had a certain relationship with the cyst size of patients. Pearson correlation test revealed that APTT, PT and TT were negatively correlated with the mean cyst diameter, while FIB was positively correlated with it. APTT, PT, and TT increased in patients treated with mifepristone, while the FIB decreased. Those with good treatment efficacy had longer APTT, PT, and TT but lower FIB as compared with those with poor efficacy. This also shows that better improvement in abnormal blood coagulation system correlates with better mifepristone treatment efficacy. Ding et al. [25] compared the indicators of patients with endometriosis cysts and those with benign ovarian cysts without endometriosis and found that patients with endometriosis cyst had shorter TT and PT, but higher FIB as compared with patients with benign ovarian cysts; In addition, FIB was associated with inflammatory markers C-reactive protein (CRP), neutrophil-to-lymphocyte ratio (NLR), and platelet-to-lymphocyte ratio (PLR). Finally, the two-year follow up data of patients were analyzed; and the multivariate analysis showed higher abortive times and shorter post-treatment TT were independent risk factors for the relapse in patients.

Nevertheless, this research still has some shortcomings. First, this is a mere retrospective study of clinical data, which involved no endometrium and tissue samples of endometriosis patients, so the results might be not convincing enough. In the subsequent study, endometrial tissue samples and ovarian benign cysts will be compared. Second, for different kinds of endometriosis, such as ovarian endometriosis, peritoneal endometriosis, and deep infiltrating endometriosis, it's vague whether these indicators have the same diagnostic values, which need to be explored later [26]. Finally, although we have found some relationship between coagulation factors and patients with cysts, the specific mechanism still needs to be explored through animal experiments.

To sum up, coagulation-related factors are associated with endometriosis. They can effectively evaluate the treatment efficacy of mifepristone, which has a great application prospect.

Disclosure of conflict of interest

None.

Table 4. Multivariate analysis of variables affecting patients' prognosis

	B	S.E	Wals	Sig.	Exp (B)	95% CI of EXP (B)	
						lower limit	Upper limit
Abortion (n)	3.676	1.462	6.324	0.012	39.485	2.260	692.833
TT after treatment (s)	-3.250	1.462	4.940	0.026	0.039	0.002	0.681

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