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The Effect of Laparoscopic Radical Surgery for Endometriosis on Serum Levels of Lipid Profile and Vitamin D

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ABSTRACT

Background & Objective: Endometriosis is associated with the increased risk of coronary heart disease and immune alterations, which may be attributed to the altered lipid profile and decreased serum level of 25–hydroxyl vitamin D (25(OH)D). The present study aimed to evaluate the effect of radical laparoscopic surgery of endometriosis on serum lipid profile and 25(OH)D.

Materials & Methods: This cross-sectional study was performed on 47 women aged 15 to 45, with body mass index <30kg/m^2, who were referred to Shohaday-eTajrish Hospital, from May 2018 until Jan 2020, for surgical treatment of endometriosis and did not have a systemic disease and did not use oral contraceptives and/or other hormonal therapies three months before surgery were enrolled into the study. The serum lipid profile and 25(OH)D levels of patients two months after surgery were compared with presurgical levels. The results were analyzed by using paired t-test.

Results: A total of 47 patients completed the study (mean age: 32.8±7.5years). About half had no pregnancies (49.1%). Comparing the serum lipid profile of patients before and two months after surgery showed a significant decrease in triglyceride from 108.4±46.2 to 86.4±51.1 mg/dL (P=0.001),], total cholesterol from 172.5±26.5 to 160.0±28.3mg/dl (P=0.002), and low-density lipoprotein levels from 97±28.3 to 89.8±26.1 mg/dL (P=.003); however, high-density lipoprotein and 25(OH)D levels did not show a significant difference (P>0.05).

Conclusion: the results of the current study showed that laparoscopic resection of endometriotic lesions resulted in a significant reduction of the unfavorable lipid profile after two months, but not 25(OH)D levels.

Keywords: Cholesterol, Endometriosis, Laparoscopy, Triglycerides, Vitamin D

Introduction

Endometriosis is generally considered a benign gynecologic disorder, while it may be accompanied by malignant ovarian tumors**(1)** or may cause severe complications, such as pelvic pain, dysmenorrhea, and infertility **(2)**. Endometriosis has been reported in about 10% of women of reproductive age; however, several cases may remain undiagnosed, as it does not have any pathognomonic sign/symptom **(3),** and clinical characteristics can only help predict endometriosis **(4)**.

However, the exact etiology of endometriosis is still unclear. A combination of genetic, familial, and environmental factors has been suggested as the pathophysiology of endometriosis **(5)**. Retrograde menstruation during endometriosis was suggested as the most commonly accepted etiology that can cause inflammatory and immune responses in the peritoneum and other sites **(6)**. A study on animal models suggested altered lipid metabolism as one of the reactions to peritoneal inflammation **(7)**. Clinical studies have also confirmed unfavorable lipid profiles, especially higher levels of triglycerides (TG), total cholesterol (TC), low-density lipoprotein (LDL), and high-density lipoprotein (HDL) levels in women with endometriosis **(8, 9)**, associated with the increased risk of coronary heart disease (CHD) in women with endometriosis **(10)**.

Furthermore, statins have been suggested as novel endometriosis treatments, which can reduce patients' pain and other symptoms, possibly by reducing inflammation and apoptotic activities **(11, 12)**. Accordingly, it seems that endometriosis has a shared mechanism with an unfavorable lipid profile; but some

other researchers have rejected such an association **(9, 13)**. Another important finding in women with endometriosis is the lower serum level of 25–hydroxyl vitamin D (25(OH)D), suggested to be associated with the dysregulated immunity during endometriosis **(14)**. Furthermore, supplementation with 25(OH)D modulates inflammation and proliferation in endometriotic cells and serves as a novel treatment for endometriosis **(15, 16)**. As far as we are concerned, although the effect of laparoscopic resection of endometriosis has been confirmed on pain and clinical symptoms of the patients with endometriosis **(17)**, its impact on the serum levels of lipids and 25(OH)D has not been studied previously. According to the importance of serum lipid profiles in CHD and 25(OH)D in immunity, it is worth studying whether the laparoscopic resection of endometriosis can correct these disorders in women with endometriosis or not. Therefore, the present study aimed to evaluate the effect of radical laparoscopic surgery of endometriosis on serum levels of lipid profile and 25(OH)D.

Methods

Study Design

This cross-sectional study included all women aged 15 to 45 who were referred to the Gynecology Department of General and Referral Shohaday-e Tajrish Hospital affiliated with the Shahid Beheshti University of Medical Sciences, from May 2018 until Jan 2020, for the surgical treatment of endometriosis. Exclusion criteria were patients with thyroid disorders, current pregnancy, women with a history of systemic diseases (diabetes and hypertension), women with body mass index (BMI)>30 kg/m^2, women who used alcohol or smoked, and those who used oral contraceptives and/or other hormonal therapies three months before surgery. The eligible patients were enrolled in the study voluntarily and by the census method. This study was approved by the Medical Ethics Committee of the Shahid Beheshti University of Medical Sciences (code: IR.SBMU.RETCH.REC.1398.104). The patients were recommended not to change their diet after surgery.

The following information was collected from medical records, interviewing the patients, and physical examination and recorded in the study checklist: patients' age, weight, age at marriage, number of pregnancies, abortions, live births, and children numbers, endometriosis symptoms, and family history of endometriosis and pelvic infection. The severity of the participants' pelvic pain, dyspareunia, and dyschezia was evaluated using a 10–point visual analog scale (VAS), and the participant was asked to indicate the pain severity from 0 (fewer) to 10 (the highest) on this scale.

All participants had regular and similar diets before and after surgery. On the day of admission and after 12 hours of fasting, the patient was sent to the hospital laboratory. One blood sample (5 mL) was taken from the participant's left cubital vein in the sitting position, collected in two clots activator tubes, and kept at room temperature. The first tube was used for the analysis of the lipid profile. For this purpose, after about 15 minutes, the samples were centrifuged at 4000 RPM to separate serum. The sera were transferred to an Auto Analyzer (Kitachi, Model No. 917), which works by photometry. The Pars Azmoon kits (Iran) were used for the analysis, with the following normal ranges: TG 50–250 mg/dL, TC<120–130 mg/dL, HDL≥35 mg/dL, and LDL≤ 100 mg/dL. For the measurement of 25(OH)D levels, blood samples were centrifuged at 3500 RPM for 20–30 minutes for the separation of serum. The sera were analyzed right after centrifugation or kept at -20°C freezer until the next day. After the samples reached the room temperature, Statfax device Model No. 2000 was used for performing enzyme-linked immunosorbent assay (ELISA), using an Ideal kit (Iran) with the following range: deficient <20 mg/dL, insufficient 20–29 mg/dL, sufficient 30–100 mg/dL, and potential toxicity >100 mg/dL.

After bowel preparation and prophylaxis of thromboembolism, and insertion of ureteral stent (where needed), the patients underwent radical laparoscopic treatment of endometriosis by one gynecologic surgeon using Karl Storz Laparoscopy device (Tuttlingen, Germany). For the surgery, the patient was laid in the lithotomic position. After entering the abdomen, an exploration of the abdomen and pelvis was performed to detect endometriotic lesions and their characteristics (penetration). All the pelvic organs, such as uterosacral, round ligament, peritoneum, and sigmoid colon, were evaluated. Appendicitis and extrapelvic sections were evaluated for the presence of endometriotic lesions. Based on the patient's stage and positive findings, the ovarian adhesions were released, and the endometriotic lesions were resected or ablated. Cystectomy was performed in case of the presence of endometrioma, and the ovaries and uterus were suspended. After releasing the ureters and detection of the hypogastric nerve, the deep lesions below the ovarian fossa and uterosacral area and the deep endometriotic lesions of the rectum and bowel were excised using the shave method or discoid/segmental resection and anastomosis, based on the penetration, size, and location detected before surgery using physical examination, magnetic resonance imaging, and transvaginal/transanal Doppler ultrasound. After cystostomy, the lesions were excised and repaired in cases with the deep involvement of the bladder. Omentum, appendix, and diaphragm were also evaluated, and in case of involvement, the lesions were excised by omentectomy, appendectomy, and resection/ablation of lesions. In cases with an indication of hysterectomy, oophorectomy, or salpingectomy, these procedures were simultaneously performed. Deep lesions involving large pelvic vessels or nerves were shaved or resected.

Any participant who performed the postsurgical serum test earlier than two months was excluded from the study. The study's sample size was calculated at 47, considering α error of 0.05 and β of 0.85, but larger sample size was included, considering the risk of loss to follow–up.

$$n_1 = n_2 = \frac{\left(z_{\alpha/2} + z_{\beta}\right)^2 \left(s_1^2 + s_2^2\right)}{\varepsilon^2}$$

Finally, 73 patients were evaluated, ten patients did not refer for the postsurgical follow–up, and 16 performed the second blood sample examination earlier than two months after surgery and were thus excluded from the study, resulting in analyzing the data of 47 patients. The mean ± SD of the participants' age was 32.8 ± 7.5 (20–45) years and the age at marriage was 22.8 ± 3.7 (14–45) years

Statistical Analysis

For the statistical analysis, SPSS Statistical Software version 21.0 (SPSS Inc., Chicago, IL., USA) was used. The results were described by mean ± standard deviation (SD) and frequency (percentage), and compared between the two intervals (before and after) using paired t-test. The normal distribution of the numeric variables was previously confirmed by the results of the Kolmogorov Smirnov test. P-values<0.05 were considered statistically significant.

Results

Ten patients were excluded from this study; because they did not perform lab tests on time.

The findings of the present study showed that TG, TC, and LDL levels reduced significantly two months after surgery, but HDL and 25(OH)D did not change. The mean ± SD of the participants' weight was 60.6 ± 6.3 kg (50 to 74 kg), and that of BMI was 25.0 ± 2.8 kg/m^2 (17 to 29 kg/m^2). About half had no pregnancies (49.1%); the frequency of the demographic and clinical characteristics of the patients are shown in Table 1.

Table 1. Demographic and clinical characteristics of the participants

Variable	Category	Number	Frequency(%)
Number of pregnancies	0	28	49.1%
	1	15	26.3%
	2	8	14%
	3	3	5.3%
	4	3	5.3%
			100%
Number of live births	0	29	50.9%
	1	18	31.6%
	2	7	12.3%
	3	2	3.5%
	4	1	1.8%
			100%
Number of abortions	0	50	87.7%
	1	5	8.8%
	2	1	1.8%
	3	1	1.8%
			100%
Number of children	0	29	50.9%
	1	18	31.6%
	2	7	12.3%
	3	2	3.5%

Variable	Category	Number	Frequency(%)
	4	1	1.8%
			100%
Type of delivery	Normal vaginal delivery	6	21.4%
	Cesarean section	17	60.7%
	Both	5	17.9%
			100%
Positive family history of endometriosis		6	10.5
Positive family history of pelvic infection		5	8.8
Endometriosis symptoms	Abnormal uterine bleeding	23	40.4
	Pelvic pain	18	38.3
	Dyspareunia	24	51.1
	Dyschezia	10	21.3
	Dysmenorrhea	39	83

The mean ± SD of the patients' pain severity were 3.3 ± 3.5 in dyspareunia, 1.2 ± 2.5 in dyschezia, 6.5 ± 3.4 in dysmenorrhea, and 1.4 ± 2.7 in pelvic pain (all with a range of 0 to 10). As shown in Figure 1 and Table 3, comparing the serum lipid profile of the patients before and two months after surgery showed a significant decrease in TG from 108.4 ± 46.2 to 86.4 ± 51.1 mg/dL (*P*=0.001), in TC from 172.5 ± 26.5 to 160.0 ± 28.3 mg/dL (*P*=0.002), and in LDL levels from 97 ± 28.3 to 89.8 ± 26.1 mg/dL (*P*=0.003); however, HDL levels did not show any significant difference (from 51.6 ± 21.1 to 50.3 ± 15.9 mg/dL; *P*=0.711). The serum levels of 25(OH)D also did not show any significant difference before and after surgery; the serum level of 25(OH)D was 19.5 ± 8.4 mg/dL before surgery, and the postsurgical value was 20.2 ± 8.9 mg/dL (*P*=0.487) (Table 2).

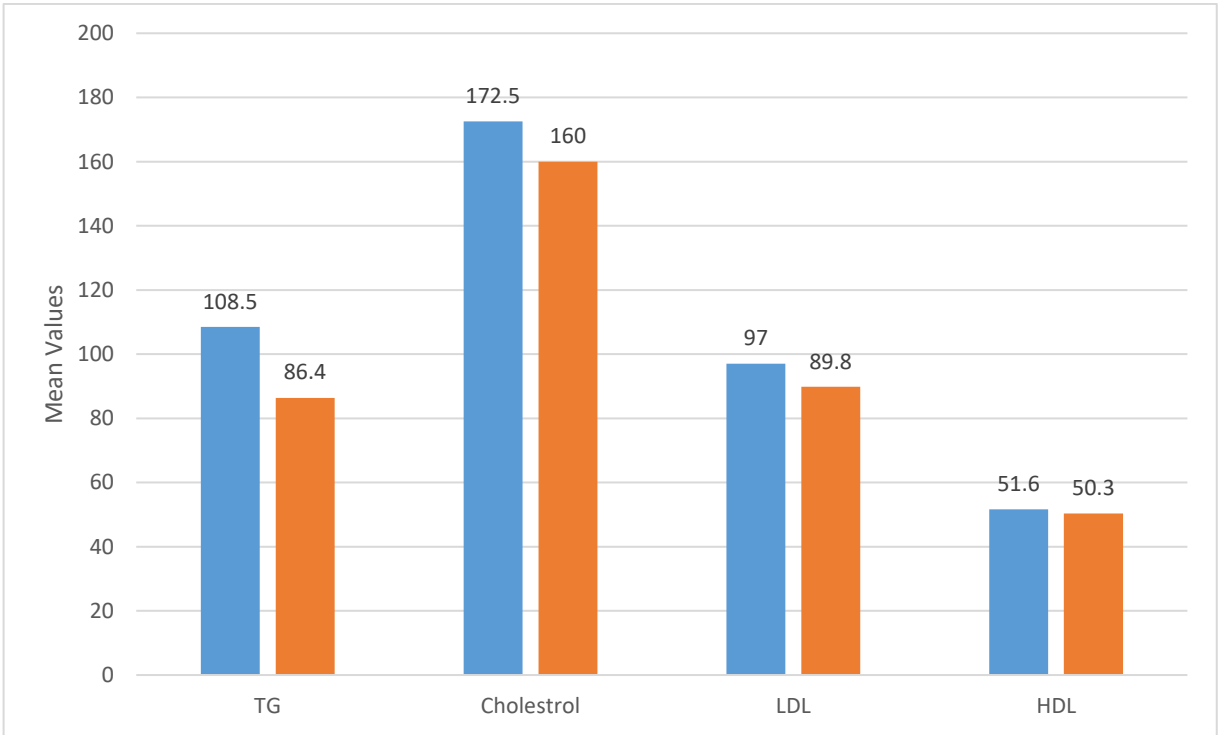

Figure 1. Comparing the serum levels of lipid profile before and after surgery

Table 2. Comparing the serum levels of lipid profile and vitamin D before and after surgery

Before surgery	After surgery	Mean (SD)	t	df	*P*
TG	108.4(+-46.3)	86.4(+-51.1)	3.473	46	0.001
Cholesterol	172.5(-+26.5)	160(+-28.3)	3.336	46	0.002
LDL	97(+-28.3)	89.8(+-26.1)	3.087	46	0.003
HDL	51.6(+-21.1)	50.3(-+15.9)	0.372	46	0.711
Vitamin D	19.5(+-8.4)	20.2(-+8.9)	0.701	46	0.487

Table 3. Comparing symptoms with the mean and SD of VAS Score

	Mean	SD	VAS (MEAN)	VAS (MAX)
Dyspareunia	3.3	3.5	0	10
Dysmenorrhea	6.5	3.4	0	10
Dyschezia	1.2	3.5	0	10
Chronic pelvic pain	1.4	2.7	0	10

Discussion

These results of our study indicated the effect of radical laparoscopic surgery of endometriosis on lipid profile. It has been previously shown that women with endometriosis have an unfavorable lipid profile. In the study by Melo *et al.*, 40 women with endometriosis aged 18–40 years were compared with 80 age-matched controls, and the results showed higher TG (105.3 ± 68.1 vs. 83 ± 33.6 mg/dL), TC (232.2 ± 75.7 vs. 178.4 ± 35.2 mg/dL), and LDL levels (150.2 ± 58.5 vs. 108.3 ± 31.5 mg/dL) in the endometriosis group than the control group **(8)**, which was associated with increased risk of CHD in women with endometriosis **(10)**. The study by Melo *et al.*, had the advantage of considering several inclusion criteria for the participants, such as excluding pregnant women, women with BMI >30 kg/m^2, history of dyslipidemia, and those under hormonal therapy that confirmed the effect of endometriosis on unfavorable lipid profile. Similarly, in the present study, we considered several inclusion/exclusion criteria in order to reduce the effect of confounders. Nevertheless, the mean values of serum lipid profile seemed lower in our study compared to the endometriosis group in the study by Melo et al **(8)**. In another study, Almassinokiani and colleagues compared 25 women with endometriosis with 25 controls, matched in terms of age, parity, and BMI. The results showed that women with endometriosis had a higher level of TC (160.9 ± 29.5 vs. 140.9 ± 20.4 mg/dL), while HDL, LDL, and TG levels were not statistically different **(9)**. In another study on Iranians, a comparison of intima-media thickness (IMT) of 30 women with endometriosis with 30 controls using carotid ultrasonography also showed no difference in IMT of these two groups **(13)**. Similar to these results, in the present study, mean levels of TC, TG, LDL, and HDL were not above the normal range. These discrepancies in the results of the studies could be due to the effect of several factors on the serum lipid profile of the individuals, such as genetics, lifestyle, diet, and exercise **(18)**, which may differ in the study populations.

Considering the efficacy of endometriosis treatment on reducing the unfavorable serum lipid profile of women with endometriosis, statins have been confirmed as a novel effective treatment that can reduce the symptoms of endometriosis by their antiproliferative, antioxidant, and anti-inflammatory properties **(11)**. Other mechanisms, such as inhibition of matrix metalloproteinase activity, inhibition of retinoic acid on the growth of human endometrial stromal cells **(19)**, and inhibition of monocyte chemotactic protein 1 (MCP–1), a chemokine, activating monocytes/macrophages in the peritoneum of women with endometriosis have also been suggested as mechanisms of the statins efficacy on endometriosis **(20)**. Nevertheless, these treatments are considered subsidiary, and the treatment of choice in endometriosis is surgical resection, preferably by laparoscopy, especially in patients with severe symptoms **(21)**. But the effect of this treatment on serum lipid profile and 25(OH)D has not been studied previously to be comparable to the present study results. Our results confirmed the shared mechanism of the altered lipid metabolism and endometriosis, as the unfavorable lipid profile significantly reduced after resection of the endometriotic lesions and reduced pelvic/peritoneal inflammation. It has been previously confirmed that granulocyte-macrophage colony–stimulating factor (GM–CSF), interleukin (IL)-2, IL–8, and IL–10 decreased significantly two weeks after

surgery **(22)**. The reduced inflammation after surgery has been considered the major cause of the return of the women's ability to the natural conception and fertility **(23)**. The same mechanism can be attributed to reducing TG, TC, and LDL two months after surgery in the present study.

In addition to statins, other immunomodulatory treatments have also been confirmed for endometriosis **(24)**. It has been shown that supplementation with vitamin D3 can modulate inflammation and proliferation in endometriotic cells in cellular and animal models **(15, 16)**. In another study, vitamin D supplementation could only reduce IL–6 levels in women with endometriosis **(25)**. Furthermore, women with endometriosis were found to have a lower serum level of 25(OH)D **(14)**, and women with severe endometriosis were reported to have lower 25(OH)D levels compared to the women with mild endometriosis and healthy controls **(15)**. It has also been shown that higher 25(OH)D levels and intake of dairy foods resulted in a lower risk of endometriosis **(26, 27)**. In our study, the mean serum level of 25(OH)D was below the normal limits before and after surgery and the surgical removal of endometriotic lesions could not improve the 25(OH)D levels. These results could be due to the effect of other factors on 25(OH)D levels, such as lack of sun exposure, which results in a high frequency of vitamin D deficient in women in our community **(29)**.

This study had several limitations. One of them was the non-randomized inclusion of the patients into the study, which increases the effect of confounders. We also selected patients from one center, limiting the study results' generalizability. Furthermore, we only evaluated the serum levels after two months, while these values may be different at earlier or later intervals. In addition, some patients were not accessible after surgery and had to be excluded from the study. Also, several factors, such as patients' exercise and sun exposure after surgery, could affect the study outcome but could not be controlled in this study.

Conclusion

In conclusion, the current study results showed that laparoscopic resection of endometriotic lesions resulted in a significant reduction of the unfavorable lipid profile after two months, but not 25(OH)D levels. Therefore, we suggest a revision of the indications of laparoscopic surgery to treat endometriosis and consider it for all women with endometriosis with unfavorable serum lipid profiles. This approach may also be beneficial for reducing the risk of CHD in these women. Further studies are required to study this issue.

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Conflict of Interest

The authors declared no conflict of interests.

Authors' contribution

Study concept and design: BN, and MB, drafting of the manuscript: BN and MA, critical revision of the manuscript: MA, BN, and MB, and Statistical Analysis: MB. All authors have given final approval for the version to be published.

Abbreviation

TG:Triglycerides

TC: total cholesterol

LDL: low-density lipoprotein

HDL: high-density lipoproteins

CHD:coronary heart disease

BMI:body mass index

VAS: visual analogue scale

ELISA:enzyme-linked immunosorbent assay.

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