narra j

Short Communication

Effect of *Nigella sativa* seed extract on estradiol, FSH levels, and vaginal maturity index in menopausal women: A randomized controlled trial

Khairani Sukatendel[1*], Reni H. Hasibuan[2], Muhammad FG. Siregar[3], Dwi Faradina[4], Deri Edianto[4], Letta S. Lintang[5], Muhammad Rusda[3] and Vega Inriani[6]

[1]Division of Social Obstetrics and Gynecology, Department of Obstetrics and Gynecology, Faculty of Medicine, Universitas Sumatera Utara, Medan, Indonesia; [2]Obstetrics and Gynecology Residency Program, Department of Obstetrics and Gynecology, Faculty of Medicine, Universitas Sumatera Utara, Medan, Indonesia; [3]Division of Fertility and Reproductive Endocrinology, Department of Obstetrics and Gynecology, Faculty of Medicine, Universitas Sumatera Utara, Medan, Indonesia; [4]Division of Oncology, Department of Obstetrics and Gynecology, Faculty of Medicine, Universitas Sumatera Utara, Medan, Indonesia; [5]Division of Fetomaternal, Department of Obstetrics and Gynecology, Faculty of Medicine, Universitas Sumatera Utara, Medan, Indonesia; [6]Faculty of Medicine, Universitas Sumatera Utara, Medan, Indonesia

*Corresponding author: khairanisukatendel123@gmail.com

Abstract

Nigella sativa seed extract has been shown to have a significant effect on endometrial thickness and vaginal cytology in ovariectomized animal models, suggesting potential benefits for managing menopausal symptoms. However, to the best of the author's knowledge, no human studies have been done to support these conclusions. The aim of this study was to investigate the effects of *N. sativa* seed extract on estradiol, follicle-stimulating hormone (FSH), and the vaginal maturity index (VMI) in postmenopausal women. A single-blinded, randomized placebo-controlled experiment was carried out at Haji Adam Malik Hospital, Medan, Indonesia, with 50 eligible postmenopausal women patients randomized into three groups. Group 1 received a placebo, while groups 2 and 3 were given *N. sativa* seed extract at 910 mg/day and 1,365 mg/day, respectively. All participants were blinded to the treatment they received. The study used Shad Nigella Plus, an Indonesian herbal medicine containing 455 mg of *N. sativa* seed extract per capsule. Before the treatments, estradiol levels, FSH levels, and VMI were measured at baseline and remeasured after eight weeks of treatment. Two participants in the intervention group withdrew due to nausea, a reported side effect of *N. sativa* seed extract consumption. Both treatment groups showed significant increases in estradiol levels (p=0.01 and p=0.001) and VMI (p=0.004 and p=0.001) after eight weeks of daily *N. sativa* seed extract administration compared to the placebo group. However, no significant differences were found between the two doses in estradiol levels and VMI (p=0.12 and p=0.673, respectively). Moreover, FSH levels showed no significant difference throughout both interventions (p=0.53 and p=0.96, respectively). In conclusion, twice-daily *N. sativa* seed extract at 910 mg/day or 1,365 mg/day for eight weeks significantly increased estradiol levels and VMI in menopausal women but had no significant effect on FSH levels. These findings support the potential role of *N. sativa* seed extract as a natural treatment for menopausal symptoms.

Keywords: Estradiol, FSH, *Nigella sativa*, postmenopausal women, vaginal maturity index

Introduction

World Health Organization (WHO) predicts that by 2030, the global population of postmenopausal women will be approximately 1.2 billion. Although menopause is a natural physiological transition, not classified as a disease, its symptoms often significantly influence

https://narraj.org/

Received: August 29, 2024 - Accepted: February 4, 2025 - Published online: February 17, 2025

both physical and psychological well-being. Menopause is primarily characterized by follicular depletion, which causes a significant decline in estradiol (estrogen) and inhibin-B secretion. Consequently, circulating estrogen levels decrease, while follicle-stimulating hormone (FSH) levels rise, often exceeding 40 mIU/mL [1].

Up to 75% of post-menopausal women have genitourinary symptoms, such as dyspareunia, genital tract damage, decreased libido, and uterovaginal prolapse [2]. If not treated, these symptoms may progressively worsen. Estrogen deficiency is a major factor in this process, resulting in diminished vascularity, a decline in vaginal maturity index (VMI), as well as decreased glandular secretion [3]. Hormone replacement therapy (HRT) with conjugated equine estrogen is the primary treatment for these symptoms. While HRT can be beneficial for these menopausal symptoms, it has side effects such as an increased risk of stroke and cardiovascular disease [4].

Phytoestrogens, plant-derived compounds with a structure similar to 17-β-estradiol, can bind to estrogen receptors and produce estrogenic effects in animal models and cell cultures [5]. *Nigella sativa*, a Ranunculaceae family plant native to Southwest Asia, the Middle East, and Southeast Asia, is a rich source of phytoestrogens [6,7]. Thymoquinone, the primary bioactive compound (30–48%), as well as flavonoid and phenolic compounds, contribute to its uterotrophic activities. These compounds may help regulate hormonal imbalances and reduce menopausal symptoms due to their phytoestrogen and antioxidant properties [6,7].

N. sativa seed extract has been reported to have a significant effect on endometrial thickness [8] and vaginal cytology in ovariectomized rats (*Rattus norvegicus*) [9], suggesting its potential benefits for treating menopausal symptoms [8,10-14]. While several previous animal studies have shown its efficacy, clinical data in postmenopausal women remains limited. To the best of the author's knowledge, the present study is the first study to assess the *N. sativa* effects in postmenopausal woman patients, as previous studies have mostly used animal models, which may not directly reflect human physiological responses. Therefore, the aim of this study was to investigate the effects of *N. sativa* seed extract on estradiol levels, FSH levels, and the VMI in postmenopausal women.

Methods

Study design and setting

This study used a single-blind, randomized, placebo-controlled experiment to carefully evaluate its aims. The experiment was conducted at Haji Adam Malik Hospital, Medan, Indonesia, and specifically targeted postmenopausal women as participants. The study was held from September to November 2023.

Sample size and sampling method

According to the minimum sample size calculation, 48 participants were required for this study. However, the sample size was increased to 50 to account for potential loss of follow-up and participant withdrawals. The study used successive sampling to enroll participants. To guarantee adherence to the regular drug regimen, all participants received daily telephone follow-ups.

Patient criteria

The inclusion criteria for participants of this study included postmenopausal women with a menopause duration of 2–10 years, a body mass index (BMI) ≤30 kg/m², and no history of unilateral or bilateral hysterectomy or oophorectomy. Moreover, participants were required to have no smoking history or no chronic diseases such as diabetes mellitus, autoimmune disorders, hypertension, and malignancy. In addition, participants who had taken HRT during the previous three months were eliminated, as were those who consumed herbal remedies containing phytoestrogens and antioxidants (vitamins E and C). Moreover, participants who had a history of vaginal cleaning or intravaginal suppository use, as well as sexual activity within three days before sample collection were also eliminated. The exclusion criteria for participants of this study included participants who had defects in their vaginal smear cytology. While, the dropout criteria included participants who were lost to follow-up, participants who withdrew from the

experiment, and participants who suffered from drug intolerant such as nausea, vomiting, and dizziness.

Randomization and treatments of participants

A total of 50 participants were chosen and randomly allocated to one of three groups using a simple randomization method. Each participant drew a tiny ball labeled with the numbers 1, 2, or 3, according to their group assignment. Group 1 served as the placebo group, while groups 2 and 3 received 910 mg/day and 1,365 mg/day of *N. sativa* seed extract, respectively. All participants remained blinded to their assigned treatment. This study used Shad Nigella Plus, an Indonesian herbal medicine containing *N. sativa* seed extract, with each capsule containing 455 mg of *N. sativa* seed extract. Participants were distributed as follows: Group 1 received two placebo capsules per day (n=10). Group 2 received two capsules of *N. sativa* seed extract per day (910 mg, n=20). Group 3 received three capsules of *N. sativa* seed extract per day (1,365 mg, n=20). The treatment was administered for eight weeks, maintaining a consistent dosage throughout the study.

Study procedure

Each participant in this study received a medical history and general physical assessments, and the data were recorded. A 5 mL blood sample was collected to measure serum estradiol and FSH levels before the treatment for the baseline data. Serum estradiol and FSH levels were analyzed at the Laboratory of Gatot Subroto, Medan, Indonesia. Moreover, a vaginal smear was performed to assess the VMI. Vaginal smears were obtained by swabbing the proximal one-third of the lateral vaginal wall using an Ayre spatula, then transferring the material to a slide and fixing it with 96% alcohol. Samples were stained with hematoxylin-eosin at the Laboratory of Anatomical Pathology, Haji Adam Malik Hospital, Medan, Indonesia, and observed under a light microscope at 200× magnification to determine vaginal cell types. Vaginal cell types (parabasal, intermediate, and superficial) were then counted, and the VMI was calculated as: (0.2 × % parabasal cells) + (0.6 × % intermediate cells) + (1.0 × % superficial cells) [15,16]. A VMI below 52 indicates vaginal atrophy [16].

After the baseline assessment of estradiol levels, FSH levels, and VMI, participants received their assigned treatment for eight weeks. Group 1 (placebo group) took one capsule twice daily, group 2 (910 mg of *N. sativa* seed extract) took one capsule twice daily, and group 3 (1,365 mg of *N. sativa* seed extract) took one capsule three times daily after meals. Compliance was monitored through thrice-weekly telephone follow-ups and weekly home visits, with any adverse reactions recorded. After eight weeks, blood sampling and vaginal smear examinations were repeated to assess changes in estradiol levels, FSH levels, and VMI.

Statistical analysis

Data was analyzed using SPSS version 21.0 (IBM SPSS, Chicago, USA) with a statistical significance level of $p<0.05$. For quantitative data, mean and standard deviation (SD) were calculated. Bivariate analysis of pre- and post-study groups used the paired t-test, Chi-square test, or analysis of variance (one-way ANOVA) for normally distributed data, with the least significant difference (LSD) as a post hoc test after one-way ANOVA for comparison between groups. Non-normally distributed data were analyzed using the Wilcoxon test for paired data (pre- and post-study comparisons), and the Mann-Whitney test for independent data comparisons.

Results

Characteristics of participants

Initially, 50 participants participated in this study. However, after two follow-ups, two participants withdrew due to unacceptable side effects, while one was eliminated for failing to comply with the specified drug regimen. Medication adherence, measured by the number of remaining capsules in returned packs was 95% in the intervention group and 100% in the placebo group. The individuals' baseline characteristics are presented in **Table 1**. There were no significant differences between the groups in terms of maternal age, age of menarche, duration of menopause, or parity.

Table 1. Characteristics of the study participants

Variables	Total, mean±SD	Post-menopausal women			*p*-value
		Placebo, mean±SD	*Nigella Sativa* 910 mg/day, mean±SD	*Nigella Sativa* 1,365 mg/day, mean±SD	
Maternal age	55.40±3.17	55.80±2.93	55.94±2.93	54.63±3.33	0.4[a]
Menarche	13.70±144	13.50±1.71	13.89±1.24	13.63±1.53	0.76[a]
Duration of menopause	5.20±2.28	5.50±2.32	5.37±2.19	4.89±2.42	0.74[a]
Parity					0.6[b]
Nulliparity	4 (8.33%)	0 (0.00%)	1 (2.08%)	3 (6.24%)	
Primiparity	3 (6.25%)	0 (0.00%)	1 (2.08%)	2 (4.16%)	
Multiparity	41 (85.42%)	10 (20.83%)	17 (35.42%)	14 (29.17)	

SD: standard deviation
[a]Analyzed using a one-way ANOVA test
[b]Analyzed using the Chi-square test

Estradiol levels

In the placebo group, estradiol levels showed a non-significant decrease, falling from 15.67±9.11 pg/mL before the intervention to 13.32±4.87 pg/mL afterward (p=0.508). In contrast, both *N. sativa* seed extract intervention groups (910 mg/day and 1,365 mg/day) experienced significant increases in estradiol levels. The 910 mg/day group's levels increased from 34.22±30.95 pg/mL to 52.51±32.05 pg/mL (p=0.01), while the 1,365 mg/day group showed an increase from 28.85±24.48 pg/mL to 71.32±46.11 pg/mL (p=0.01). These findings suggest that *N. sativa* seed extract at both doses may have a positive effect on estradiol levels, unlike the placebo (**Table 2**).

Table 2. Comparison of the mean estradiol levels of postmenopausal women among study groups before and after intervention

Groups	Levels of estradiol (pg/mL), mean±SD		*p*-value[a]
	Before	After	
Placebo	15.67±9.11	13.32±4.87	0.508
Nigella sativa seed extract 910 mg/day	34.22±30.95	52.51±32.05	0.01[*]
Nigella sativa seed extract 1,365 mg/day	28.85±24.48	71.32±46.11	0.001[**]

SD: standard deviation
[a]Analyzed using the Wilcoxon test
[*]Statistically significant at $p<0.05$
[**]Statistically significant at $p<0.01$

Moreover, *N. sativa* seed extract supplementation at both 910 mg and 1,365 mg daily for eight weeks showed similar effects on estradiol levels, with no statistically significant difference observed between the two groups (p=0.12). This suggests that either dose provides comparable benefits regarding estradiol levels (**Table 3**).

Table 3. Comparison of estradiol levels of post-menopausal women in the intervention group of *Nigella sativa* seed extract 910 mg/day and 1,365 mg/day

Groups	Estradiol (pg/mL), mean±SD	*p*-value[a]
Before		0.24
N. sativa seed extract 910 mg/day	34.22±30.95	
N. sativa seed extract 1,365 mg/day	28.85±24.48	
After 8 weeks		0.12
N. sativa seed extract 910 mg/day	52.51±32.05	
N. sativa seed extract 1,365 mg/day	71.31±46.11	

SD: standard deviation
[a]Analyzed using the Mann-Whitney test

Follicle-stimulating hormone (FSH) levels

In the placebo group, FSH levels increased significantly from 59.20±16.53 pg/mL to 70.62±17.44 pg/mL (p=0.04). In contrast, *N. sativa* seed extract supplementation, at both 910 mg/day and 1,365 mg/day, did not significantly alter FSH levels ($p>0.05$). The 910 mg/day group experienced a slight, non-significant decrease from 60.23±18.43 pg/mL to 57.59±16.80 pg/mL (p=0.53), while the 1,365 mg/day group showed minimal change, from 56.39±13.20 pg/mL to 56.17±22.09

pg/mL (p=0.96). No significant difference in FSH levels was observed between the two *N. sativa* seed extract treatment groups (p>0.05) (**Table 4**).

Table 4. Comparison of follicle-stimulating hormone (FSH) levels of postmenopausal women among study groups before and after intervention

Groups	FSH (pg/mL), mean±SD	p-value[a]
Placebo		0.04*
Before	59.20±16.53	
After 8 weeks	70.62±17.44	
Nigella sativa seed extract 910 mg/day		0.53
Before	60.23±18.43	
After 8 weeks	57.59±16.80	
Nigella sativa seed extract 1,365 mg/day		0.96
Before	56.39±13.20	
After 8 weeks	56.17±22.09	

SD: standard deviation
[a]Analyzed using paired t-test
*Statistically significant at p<0.05

Vaginal maturity index (VMI)

The VMI increased significantly after 8 weeks of intervention in both the *N. sativa* seed extract at 910 mg/day (p=0.004) and 1,365 mg/day (p=0.001) groups. In contrast, the placebo group showed no significant change in VMI levels over the same period (p=0.06) (**Table 5**).

Table 5. Comparison of vaginal maturity index (VMI) in postmenopausal women across groups before and after intervention

Groups	Vaginal maturity index, mean±SD		p-value[a]
	Before	After	
Placebo	46.94±16.53	44.44±15.32	0.06
Nigella sativa seed extract 910 mg/day	51.52±19.41	59.96±12.29	0.004**
Nigella sativa seed extract 1,365 mg/day	48.46±17.38	58.71±11.58	0.001**

SD: standard deviation
[a]Analyzed using paired t-test
**Statistically significant at p<0.01

After eight weeks of intervention (post-treatment), VMI was significantly elevated (mean±SD: 56.23±13.77; p=0.007). Post hoc analysis showed significant differences between the placebo group and both *N. sativa* seed extract treatment groups: 910 mg/day (p=0.003) and 1,365 mg/day (p=0.006). However, no significant difference was found between the two *N. sativa* seed extract groups (910 mg/day vs 1,365 mg/day; p=0.763). This indicates that *N. sativa* seed extract supplementation, at both doses, significantly impacts VMI compared to placebo, but the two doses have comparable effects.

Table 6. The vaginal maturity index (VMI) after eight weeks of intervention among groups

Groups	n	Vaginal maturity index				p-value[a]	Post hoc	
		Mean	SD	Median	Min-max		NS 910	NS 1,365
Placebo	10	44.44	14.95	41.60	25.20–70.00	0.007**	0.003**	0.006**
NS 910	19	59.96	12.29	62.00	38.00–79.60			0.763
NS 1,365	19	58.71	11.58	62.80	41.20–81.20			
Total	48	56.23	13.77	57.20	25.20–81.20			

NS: *Nigella sativa* seed extract; SD: standard deviation
[a]Analyzed using a one-way ANOVA test
**Statistically significant at p<0.01

Discussion

This study demonstrated that *N. sativa* seed extract, at both doses of 910 mg/day and 1,365 mg/day, significantly improved estradiol levels in postmenopausal women compared to placebo. While both doses exhibited similar efficacy, a dose of 910 mg/day is recommended. These findings align with previous studies demonstrating the estrogenic effects of *N. sativa* seed extract. For instance, Sana *et al.* observed a significant increase in estradiol with 500 mg of *N. sativa* seed

extract [17]. Another study by Parhizkar *et al.* also reported that *N. sativa* seed extract significantly elevated estrogen levels, impacting uterotrophic assays and vaginal cornification [18]. Moreover, this study found no significant difference in FSH levels following intervention across all groups (placebo and both *N. sativa* seed extract dosages). These results are consistent with previous findings by Sukatendel *et al.*, which demonstrated no effect of *N. sativa* seed extract on FSH levels in rats [12].

This study found a significant increase in the VMI following *N. sativa* seed extract intervention (both 910 mg/day and 1,365 mg/day) compared to placebo. No significant difference was observed between the two *N. sativa* seed extract dosages. The placebo group exhibited a mean VMI <52, indicative of low estradiol and increased vaginal atrophy, whereas both *N. sativa* seed extract groups demonstrated a mean VMI >52, suggesting reduced atrophy. These findings are consistent with Lumbanraja *et al.*, who reported significant VMI differences in groups experiencing vaginal dryness and dyspareunia [19]. Furthermore, a study by Andriana and Lestari demonstrated a dose-dependent effect of *N. sativa* seed extract on VMI in ovariectomized rats, with the highest VMI observed in groups receiving the highest *N. sativa* seed extract doses [9].

The underlying physiology of postmenopausal vaginal epithelial changes remains incompletely understood. Decreased estrogen levels are associated with reduced paracellular permeability of the vagina and cervix, potentially contributing to vaginal dryness [19]. Estrogen deficiency during menopause leads to genitourinary tract atrophy, including the vaginal mucosa. This atrophy manifests as decreased vaginal wall vascularity and elasticity, shortening, and loss of rugae, potentially causing dyspareunia. In addition, thinning of the vaginal epithelium increases the risk of post-coital trauma and bleeding. Furthermore, decreased glycogen and lactic acid production result in a more alkaline vaginal pH, increasing susceptibility to infection and symptoms such as irritation, dryness, burning, and itching. Estrogen reduction also impacts the connective tissue of the bladder, urethra, and pelvic wall, potentially leading to dysuria, urinary frequency, nocturia, urgency, recurrent urinary tract infections, and pelvic organ prolapse [20].

N. sativa seed extract has demonstrated promising effects in mitigating menopausal symptoms. Sana and Parhizkar reported increased estradiol levels and improved menopausal rating scale scores following *N. sativa* seed extract supplementation, enhancing quality of life in postmenopausal women [17]. Nagy *et al.* observed estrogenic activity in female rats administered *N. sativa* seed extract, with histological findings revealing active endometrial hyperplasia and uterine gland dilation in the treatment group compared to controls [22]. These findings suggest a potential mechanism for *N. sativa* seed extract's beneficial effects on menopausal symptoms.

Beyond menopause, *N. sativa* seed extract has shown promise in other gynecological conditions. Mahmoudian *et al.* found that short-term *N. sativa* seed extract treatment (1000 mg/day for 16 weeks) may improve ovarian volume, hormonal balance, and menstrual abnormalities in teenagers with polycystic ovary syndrome (PCOS) [23]. Khani *et al.* observed significant changes in serum factors and improved ovarian tissue structure in PCOS rats treated with *N. sativa* seed extract, noting a dose-dependent effect [24]. Several studies have also indicated a protective effect of *N. sativa* seed extract against uterine and ovarian damage [12,25]. Further study has explored the potential benefits of *N. sativa* seed extract in primary dysmenorrhea [26], hyperprolactinemia [27], candidiasis[28], cervical cancer [29] breast cancer, ovarian cancer, premenstrual syndrome and pre-eclampsia [30].

N. sativa seed extract rich in flavonoids and phenolic compounds, exhibits significant estrogenic activity [31]. While the precise mechanism of action of phytoestrogens remains incompletely understood, evidence suggests both estrogen receptor-dependent and -independent pathways may be involved [8,32]. It is important to note that much of the existing study on *N. sativa* seed extract has been conducted in animal models. Therefore, extrapolating these findings to humans requires caution, as the therapeutic effects may differ. This study demonstrated comparable beneficial effects between the minimum and higher doses of *N. sativa* seed extract. Consequently, the minimum effective dose is recommended.

Conclusion

Supplementation with *N. sativa* seed extract (910 mg or 1,365 mg daily for eight weeks) significantly increased estradiol levels and VMI in postmenopausal women compared to placebo. However, no significant difference in FSH levels was observed. Therefore, *N. sativa* seed extract may offer a safe and effective alternative therapy for managing menopausal symptoms related to estrogen decline.

Ethics approval

This study was approved by the Ethical Committee of Universitas Sumatera Utara on 26th September 2023 (Approval No: 467/KEPK/USU/2023).

Acknowledgments

We would like to thank the participants, the pathology anatomy department of Adam Malik General Hospital, and the staff of Gatot Subroto Laboratory, Medan, Indonesia.

Competing interests

All the authors declare that there are no conflicts of interest.

Funding

This study received no external funding.

Underlying data

Derived data supporting the findings of this study are available from the corresponding author on request.

Declaration of artificial intelligence use

We hereby confirm that no artificial intelligence (AI) tools or methodologies were utilized at any stage of this study, including during data collection, analysis, visualization, or manuscript preparation. All work presented in this study was conducted manually by the authors without the assistance of AI-based tools or systems.

How to cite

Sukatendel K, Hasibuan RH, Siregar MFG, *et al*. Effect of *Nigella sativa* seed extract on estradiol, FSH levels, and vaginal maturity index in menopausal women: A randomized controlled trial. Narra J 2025; 5 (1): e1399 - http://doi.org/10.52225/narra.v5i1.1399.

References

1. Orlowski M, Sarao MS. Physiology, follicle stimulating hormone. In: Ackley WB, Adolphe TC, Aeby TC, *et al*., editors. StatPearls. Treasure Island: StatPearls Publishing; 2023.
2. Gandhi J, Chen A, Dagur G, *et al.* Genitourinary syndrome of menopause: An overview of clinical manifestations, pathophysiology, etiology, evaluation, and management. Am J Obstet Gynecol 2016;215(6):704-711.
3. Latiff LA, Parhizkar S, Dollah MA, *et al.* Alternative supplement for enhancement of reproductive health and metabolic profile among perimenopausal women: A novel role of *Nigella sativa*. Iran J Basic Med Sci 2014;17(12):980-985.
4. Rossouw JE, Anderson GL, Prentice RL, *et al.* Risks and benefits of estrogen plus progestin in healthy postmenopausal women: Principal results from the Women's Health Initiative randomized controlled trial. JAMA 2002;288:321-333.
5. Rietjens IMCM, Louisse J, Beekmann K. The potential health effects of dietary phytoestrogens. Br J Pharmacol 2017;174(11):1263-1280.
6. Wani JA, Tsagkaris C, Majid S, *et al.* Therapeutic effects of *Nigella sativa* on hormonal dysfunctions. In: Khan A, Rehman M, editors. Black seeds (*Nigella sativa*): Pharmacological and therapeutic applications. Amsterdam: Elsevier; 2021.
7. Bauld R, Brown RF. Stress, psychological distress, psychosocial factors, menopause symptoms and physical health in women. Maturitas 2009;62(2):160-165.
8. Andriana K, Lestari DY. Effect of extract black cumin (*Nigella sativa*) on the vaginal cytology of ovariectomized white mouse (*Rattus novergicus* strain Wistar). AHSR 2017;2:357-361.

9. Andriana K, Nelasari H, Arsana WIW. Effect of extract black cumin (*Nigella sativa*) on the LDL/HDL ratio of ovariectomized white mouse (*Rattus novergicus* strain Wistar). Adv Environ Biol 2015;9(23):229-232.
10. Parhizkar S, Latiff LA, Parsa A. Effect of *Nigella sativa* on reproductive system in experimental menopause rat model. Avicenna J Phytomed 2016;6(1):95-103.
11. Kamil ZH. Spectacular black seeds (*Nigella sativa*): Medical importance review. Med J babylon 2013;10(4):77749739
12. Sukatendel K, Siregar MFG, Natadisastra M, *et al.* Benefits of *Nigella sativa* extract protecting ovary due to cisplatin chemotherapy. Maced J Med Sci 2021;9(A):680-687.
13. Parhizkar S, Latiff LA, Rahman SA, *et al.* Assessing estrogenic activity of *Nigella sativa* in ovariectomized rats using vaginal cornification assay. African J Pharm Pharmacol 2011;5(2):137-142.
14. Desmawati D, Sulastri D. Phytoestrogens and their health effect. Open Access Maced J Med Sci 2019;7(3):495-499.
15. Cline JM, Paschold JC, Anthony MS, *et al.* Effects of hormonal therapies and dietary soy phytoestrogens on vaginal cytology in surgically postmenopausal macaques. Fertil Steril 1996;65(5):1031-1035.
16. Speroff L. Efficacy and tolerability of a novel estradiol vaginal ring for relief of menopausal symptoms. Obstet Gynecol 2003;102(4):823-834.
17. Sana S, Umair HM, Saeed M. Effect of *Nigella sativa* on estradiol levels and menopausal symptoms in post-menopausal females. Proceedings SZMC 2021;35(4):36-40.
18. Parhizkar S, Manzouri L. A randomised controlled trial on climacteric symptoms management and quality of life improvement of *Nigella sativa* seeds powder in menopausal women. Maturitas 2015;81(1):179.
19. Lumbanraja IL, Siregar MFG, Lumbanraja SN, *et al.* Association of vaginal maturation index and vaginal pH with the most bothersome symptoms of genitourinary syndrome of menopause. J South Asian Feder Obst Gynae 2021;13(5):288-291.
20. Simon JA, Kokot-Kierepa M, Goldstein J, Nappi RE. Vaginal health in the United States: Results from the vaginal health: Insights, views & attitudes survey. Menopause 2013;20(10):1043-1048.
21. Munno GM, La Verde M, Lettieri D, *et al.* Pelvic organ prolapse syndrome and lower urinary tract symptom update: What's new?. healthc 2023;11(10):1513.
22. Nagy AM, Abdelhameed MF, Elkarim ASA, *et al.* Enhancement of female rat fertility via ethanolic extract from *Nigella sativa* L. (black cumin) seeds assessed via HPLC-ESI-MS/MS and molecular docking. Molecules 2024;29(3):735.
23. Mahmoudian A, Ashouri A, Bilandi RR, *et al.* The possible short-term of *Nigella sativa* – L in the management of adolescent polycystic ovarian syndrome: Results of a randomized controlled trial. J Ovarian Res 2024;17(1):1-10.
24. Khani S, Abdollahi M, Khalaj A, *et al.* The effect of hydroalcoholic extract of *Nigella sativa* seed on dehydroepiandrosterone-induced polycystic ovarian syndrome in rats: An experimental study. Int J Reprod Biomed 2021;19(3):271-282.
25. Abdullah WS, Chelabal-nailey KG. *Nigella sativa*-based protection against uterus and ovary-related histological damages generated by cimetidine in mice. J Glob Pharma Technol 2018;10(08):284-289.
26. Samadipour E, Rakhshani MH, Kooshki A, *et al.* Local usage of *Nigella sativa* oil as an innovative method to attenuate primary dysmenorrhea: A randomized double-blind clinical trial. Oman Med J 2020;35(5):1-6.
27. Pakdel R, Hadjzadeh H, Sadegh M, *et al.* The effects of hydroalcoholic extract of *Nigella sativa* seeds on serum estradiol and prolactin levels and obstetric criteria due to hypothyroidism in rat. Adv Biomed Res 2017;6(1):166.
28. Fard FA, Zahrani ST, Bagheban AA, *et al.* Therapeutic effects of *Nigella sativa* Linn (black cumin) on Candida albicans vaginitis. Arch Clin Infect Dis 2015;10(1):1-5.
29. Hasan TN, Shafi G, Syed NA, *et al.* Methanolic extract of *Nigella sativa* seed inhibits SiHa human cervical cancer cell proliferation through apoptosis. Nat Prod Commun 2013;8(2):213-216.
30. Deepthika KSH, Karunagoda KPKR, Perera PK. Medicinal effects of *Nigella sativa* in gynecological disorders: A systematic review. IJPSR 2022;13(8):2977-2987.
31. Elghareeb MM, Elshopakey GE, Rezk S, *et al. Nigella sativa* oil restores hormonal levels, and endocrine signals among thyroid, ovarian, and uterine tissues of female Wistar rats following sodium fluoride toxicity. Biomed Pharmacother 2024;170(12):116080.
32. Chavda VP, Chaudhari AZ, Balar PC, *et al.* Phytoestrogens: Chemistry, potential health benefits, and their medicinal importance. Phyther Res 2024;38(6):3060-3079.