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Scientific Article

Veterinary Medicine

A systematic review of flaxseed effectiveness on infertility in animal model. Systematic Review

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Abstract: According to the extensive studies carried out in the direction of the effectiveness of flax seed in solving various causes of infertility, the present study was conducted with the aim of investigating the effect of flax seed on infertility by a systematic review method. The search for studies in national and international databases was conducted in English and Persian without time restrictions. To evaluate the quality of the articles, the Cochrane Collaboration Risk of Bias tool and the Review Manager Program software (Revman 5.3) were used**.** The findings indicate that among the 1245 articles in the initial search, 56 articles (sample size: 4988) were included in the study. The articles were completed from 1998 to 2022 in Iran, America, Canada, Egypt and other countries of the world. Among the reviewed articles about the effect of flaxseed on the causes of infertility, 25 articles are about male and female reproductive system and sex hormones, 12 articles are about PCOS, 12 articles are about egg enrichment, two articles are about fertility power, two articles on uterine myoma and 1 article on cancer were studied. In most studies, the positive effect of flaxseed on infertility was confirmed. The available evidence shows the effect of flaxseed on different causes of infertility, but it is recommended to conduct more studies on different causes of male and female infertility on human samples with a strong methodology and appropriate sample size to ensure the effect of this plant on humans.

Keywords*:* Linseed, Infertility, Animal Model, Systematic Review

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Introduction:

Infertility is a significant crisis in life [1], which is defined as the failure of a couple or couples to conceive after 6 months to a year of regular sexual contact without using contraceptive methods [2]. It is estimated that 8 to 11% of couples in the world experience infertility [3], [4], [5] and in Iran at 13.2% [1].

The causes of infertility can be female or male, so that at the global level, the cause of couples' infertility in 30% of cases is related to the male factor, 40-50% is related to the female factor, and 20-30% is related to both [6], [7], [8]. Male causes of infertility include genetic disorders, blockage of genital ducts, varicocele, sperm and seminal fluid production disorders, and sexual and erectile disorders [9] and female causes of infertility include ovulation disorders, fallopian tube disorders, and uterus, endometriosis, immune system problems, low egg quality and unknown factors [10], [11], [12].Infertility treatment options include drug treatments, non-drug treatments, lifestyle modification, surgical treatments, and assisted reproductive treatments (ART) [13], [14], which one of the most popular methods of treatment in communities is the use of medicinal plants [15], [16]. Studies show that various plants are used in the treatment of infertility [17], [18], [19], [20]. One of the effective herbal medicines in the treatment of infertility is flax seed [19], [21], [22].

The botanical name of flax is Linum usitatissimum from the Linaceae plant

family, and the word flax seed is also used for this plant [23, 24]. Flaxseed contains alpha-linolenic acid (omega-3 fatty acid). It also has a high percentage of dietary fiber, both soluble and insoluble, and finally the richest plant source of lignans [24], [25], [26]. Lignan is a phytoestrogen (non-steroidal compounds found in plants).

These compounds are structurally similar to natural estrogens such as 17-beta-estradiol, which allows them to bind to estrogen receptors and thereby induce detectable biological effects [27]. Flaxseed oil is rich in stearic, oleic, linoleic and palmitic fatty acids, which contain a large amount of vitamins, including vitamin E [28].

Flaxseed has beneficial effects on the reproductive system, such as improving sperm quality and mobility and increasing testosterone levels [19, 22, 29]. Studies show that flaxseed improves sperm quality and increases sperm viability in the female reproductive system [30], [31]. Another element in flaxseed is omega-3, which is essential to produce sex hormones such as FSH, LH, and testosterone [30], [32].

There are different effective treatments for infertility, and one of these treatments is treatment with medicinal plants, which is very popular in different cultural societies. According to the extensive studies conducted in the direction of the effectiveness of flaxseed on various causes of infertility, such as improving the quality of sperm and seminal fluid, improving ovarian function and ovulation, and improving polycystic ovary syndrome. Therefore, the present study was conducted with the aim of investigating the effect of flaxseed on infertility using a systematic review method.

Materials and Methods:

Search strategy

Studies were searched comprehensively in national and international databases such as Web of Science, Scopus, Pubmed, SID, Cochrane Library in English and Persian without time restrictions. To search for studies, the Persian keywords flax seed, infertility, female infertility factors and male infertility factors and flaxseed, Linum usitatissimum, infertility, female infertility, male infertility and OR and AND operators were used.

Primary and secondary outcomes

The primary outcomes investigated in this study included the effect of the intervention on paraclinical symptoms (hormone levels and metabolic indices) and the secondary outcomes evaluated in this

study included the effect on infertility.

Inclusion and exclusion criteria

In this study, experimental and semi-experimental studies (types of published original research articles, short forms, and scientific conferences) were included to investigate the effect of flax seeds on infertility. The inclusion criteria for studies included all types of research studies on the investigated variable. Studies that had problems in terms of statistical population or methodology were excluded from the study. The types of interventions investigated included the consumption of flax seeds in different forms of capsules, syrups, drops, powders, extracts, etc. Comparison groups also included placebo, herbal medicine or any other standard drug treatment.

Study Screening and Selection

This study was written as a systematic review based on the PRISMA 2020 checklist. At first, the search was conducted by two researchers to extract studies related to the purpose of the study in national and international databases. All the articles extracted from each database were entered in the Endnote 17 software. In the next step, after removing duplicate articles, the title and abstract of all studies were examined. Finally, the full text of relevant articles was reviewed and articles with inclusion criteria were selected.

Data Synthesis and Extraction

To extract and manage data, two researchers independently extracted data from studies with inclusion criteria. The data extraction form included: study characteristics (authors, publication year, type of study, tools used in the study), participant characteristics, and primary and secondary outcome information. In this review study, due to the heterogeneity of the data, the data were reported systematically.

Quality Assessment

The evaluation of the quality of the articles was carried out by two researchers independently, and in case of disagreement, they were resolved through negotiation with a third person. To evaluate the quality, the Cochrane Collaboration Risk of Bias tool and the Review Manager Program (Revman 5.3) were used. This tool examines the quality of articles in terms of: selection bias and implementation bias, diagnosis bias, sample attrition, selective outcome reporting bias and other biases. Each of the items examined in this tool was reported as low distortion, medium distortion, and ambiguous in terms

of distortions.

Ethical consideration

This study was approved by the ethics code IR.SKUMS.AEC.1402.018 and Grant no. 6861 in Shahrekord University of Medical Sciences.

Results:

In the initial search, 1245 articles were included in the study, and after reviewing the titles and abstracts of the articles and removing duplicate and unrelated items, 56 possible relevant articles were examined. 4 articles were excluded due to the lack of investigation of the effect of flaxseed on infertility, 10 articles due to the presence of human samples, and 2 articles due to lack of access to the full text of the article. Finally, 41 articles were included in the study (Figure 1).

Of the 41 studies included in the systematic review, 10 articles were conducted in Iran, 9 articles in the United States, 7 articles in Canada, 5 articles in Egypt, and 10 articles in other countries. The articles were published from 1998 to 2023, and the total sample size was 4988 animals. In all the articles, the effect of flax seed on various underlying causes of infertility was investigated, so that in 37 articles, the results showed that flax seed has a positive effect on infertility, while 4 articles mentioned the lack of effect of this plant on infertility. The summary of the studies is presented in Table 1.

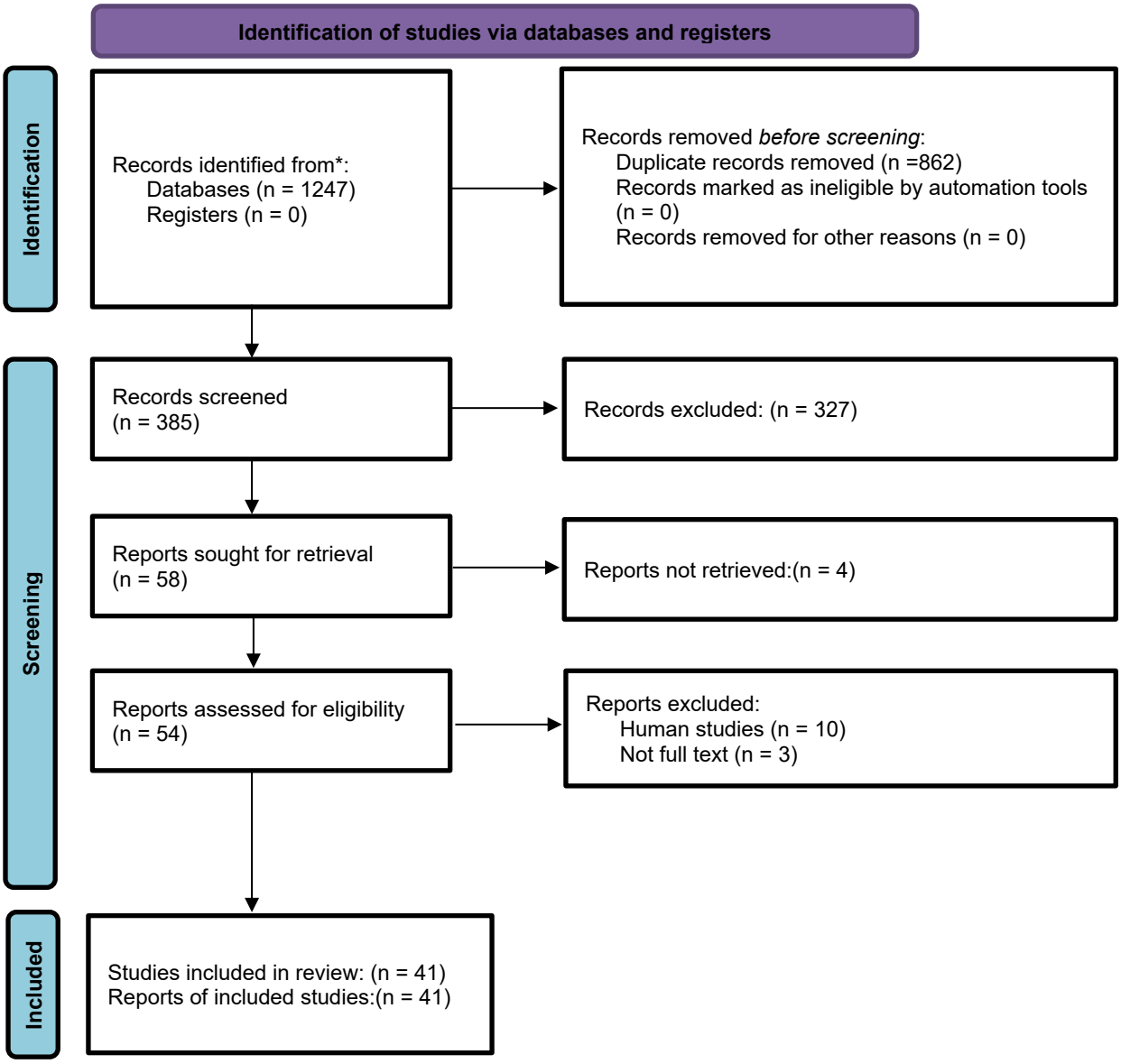

Figure 1: Flowchart of entering studies into systematic review

Table 1: Summary of animal studies on the effect of flaxseed on infertility

First Author	Study aim	Study design
Dikshit et al. (2016) [33]	The effect of flaxseed on estrogen levels in ovarian neoplasia	Reduction of ovarian cancer and reduction of inflammation and chemical changes caused by estrogen following consumption of flaxseed
Selo et al. (2016) [34] Bahmanpour et al. (2016) [35] Zanussi et al. (2019) [30] Perumal et al. (2019) [31] Paul et al. (2021) [36] Shah et al. (2016) [37] Sally et al. (2019) [38] Nikolaev et al. (2020) [39] Sohrabipour et al. (2013) [40] Corrêa et al. (2017) [41]	The effect of flaxseed on the male reproductive system	A significant increase in sex hormones such as testosterone, Follicle-Stimulation Hormone (FSH), and (LH), and by affecting various parameters, has caused an increase in fertility in rats: positive effect on sperm and seminal fluid and libido, increased testosterone levels, increased fertility after conception. Laboratory, protection of sperms against oxidative stress and improvement of varicocele, no adverse effect on spermatogenesis and testicular tissue and epididymis, increase of estradiol levels, and beneficial effect on testicular weight gain.
Janet et al. (1998) [42] Ahmad et al. (2012) [43] Petit et al. (2006) [44] Pourjafari et al. (2019) [45] Pourjafari et al. (2020) [46] Pourjafari et al. (2022) [47] Moallem et al. (2013) [48] Attia et al. (2022) [49] Vlčková et al. (2022) [50]	The effect of flaxseed on the female reproductive system	- The lack of effect of flaxseed on pregnancy outcomes, except that it causes low birth weight. - The effect of flaxseed on the reproductive system of the fetus has been seen, such that in the female fetus, the anogenital distance is shortened and the weight of the uterus and ovary is lighter, while the relative weight of the prostate and gonads in the male fetus is increased. - It has had a positive effect on the levels of estrogenic hormones. - It caused an increase in sex hormones such as estradiol and progesterone and caused a decrease in ovarian cholesterol. - Reducing the amount of casualties in pregnancies - Increase in the diameter and weight of the ovaries - Increase of estradiol and LH, FSH - Reduction of anti-apoptotic genes - Increase of follicles and positive effect on folliculoneogenesis

		- Reduction of atretic follicles - Improving the In Vitro Fertilization (IVF) process and the quality of the eggs - Positive effect on ovulation function - Positive effect on eggs and ovulation
Isgur et al. (2015) [51] SIZEMORE et al. (2018) [52]	The effect of flaxseed on polyps and fibroids of the female genital tract	- Reduction of ovarian and fallopian tube polyps - Improvement of uterine fibroids - Beneficial effect on the treatment of uterine tube leiomyomas
Janet et al. (1999) [53]	Dose, Timing, and Duration of Flaxseed Exposure Affect Reproductive Indices and Sex Hormone Levels in Rats	- Early puberty - Increase in ovarian weight and estradiol and testosterone levels - Growth of genital organs - Prostate weight loss
Jelodar et al. (2018) [54] Abd El-Galil et al. (2021) [55] Osman et al. (2019) [56] Ghasemi et al. (2021) [57] Mehraban et al. (2020) [58] Wang et al. (2020) [59]	The effect of flaxseed on polycystic ovarian syndrome (PCOS) sufferers	- Effective as a therapeutic method to help modify lifestyle in PCOS (polycystic ovarian syndrome) disease - Progesterone increase and testosterone decrease and no change in estrogen and Dehydroepiandrosterone (DHEA) - Increasing the number of antral follicles and decreasing the number of cystic follicles - Improvement of PCOS/weight loss in PCOS sufferers - Improvement of metabolic and hyperandrogenic symptoms in PCOS sufferers - Decrease of testosterone and increase of sex hormone bound globulin
Janet et al. (2000) [60]	The effects of flaxseed and ii's components on reproductive indices and cancer risk in rats	- No effect on pregnancy and breastfeeding - Positive effect on the reproductive system of female and male fetuses
Saber et al. (2020) [61]	Effect of including n-3/n-6 fatty acid feed sources in diet on fertility and hatchability of broiler breeders and post-hatch performance and carcass parameters of progeny	- No effect on the number of fertile eggs and fertility rate - Reducing the rate of death after birth
Gambonini et al. (2019) [62]	The effect of flaxseed on milk production and conception success	- Improvement of conception rate in prime cows - Increasing milk production

Fetter et al. (2017) [63] Nanas et al. (2023) [64]		- Increased lactose and milk fat
Feng et al. (2015) [65]	Effects of dietary omega-3/omega- fatty acid ratios on reproduction in the young breeder rooster	- Lack of influence on testicular indicators - Positive effect on spermatogonia and germ cells - Positive effect on the levels of sex hormones testosterone, LH , FSH, GNRH - Increasing reproductive performance of young roosters
Abdel Moniem et al. (2010) [66]	Protective role of flaxseed oil against lead acetate induced oxidative stress in tests of adult rats	- Restoring the testicular index and increasing the level of testosterone - Treatment of abnormal cases in sperm such as degenerative changes - Reducing the harmful effects of lead on the testicles
Naji et al. (2014) [67]	The Histological Effects of Cucurbita pepo, Silybum marianum, Linum usitatissmum, Vitex agnus_castus 17β estradiol on ovarian tissue in three Spot Gorami (Trichogaster trichopterus)	Increasing the diameter of the eggs (study on the tissue and in terms of histology)
Himanshu et al. (2022) [68]	Supplementation effect of dietary flaxseed and coconut oil on antioxidant enzyme activities, LPO seminal plasma protein profiling in adult ram	- Positive effect on the quality of sperm and seminal fluid - Increased testosterone levels - No effect on sperm motility and speed - Lack of influence on testicular indicators
Vlckova et al. (2018) [69]	Supplemental flaxseed modulates ovarian functions of weanling gilts via the action of selected fatty acids	- No effect on steroid hormones - Increase in insulin-like hormone 1
Yousif et al. (2019) [32]	Effect of Flaxseed on some hormonal profile and genomic DNA concentration in Karadi lambs	Decrease in testosterone and increase in estradiol, thyroid stimulating hormone (TSH) and growth hormone

Makawy et al. (2018) [70]	Flaxseed oil as a protective agent against bisphenol-A deleterious effects in male mice	Reducing testicular tissue changes and abnormal sperm conditions and reducing bisphenol-A (BPA) genotoxicity

1. The effect of flaxseed on the genitals and sex hormones of male and female

The genitals and male and female sex hormones are beneficially affected by flaxseed, which contains compounds like plant phytoestrogens, alpha-linolenic acid, and omega-3 fatty acids [36]. The effects of flaxseed on the male genitalia include increased sperm number and semen quality, increased libido, sperm protection against oxidative stress and increased serum testosterone and estradiol levels [30], [31], [35], [36], [39], [40], [41], [71].

And the effects of flaxseed on the female genital tract include increased diameter and weight of the ovary, positive effect on ovulation and increased folliculonoids, improved egg quality and IVF process, increased serum levels of estrogen and progesterone, and decreased serum cholesterol levels [42-50, 72].

While some studies have expressed the impact of flaxseed on sex hormones [69]as well as lack of effect on testicular indicators [65, 68].

2. The effect of flaxseed on metabolic disorders associated with polycystic ovary syndrome

Polycystic ovary syndrome (PCOS) is the most common endocrine disorder in fertility ages [73, 74]. The most common clinical manifestation of this syndrome includes irregular menstruation and infertility cycles [75]. Based on Rotterdam criteria, PCOS detection depends on the identification of at least two of the following three properties: menstruation, hyperogenism, and polycystic ovary in ultrasound [76]. In addition, patients with PCOS are at increased risk of metabolic syndrome, type 2 diabetes and cardiovascular disease (CVD) [77].

Lifestyle correction, such as dietary pattern, exercise, and behavioral therapies, is the first line of treatment for PCOS [78]. However, attention to medicinal plants has recently spread as an alternative therapy [79]. Linum usitatissium is a rich source of several biological active compounds, including alpha-linolenic acid (ALA), phytoestrogenic lignons and dietary fibers [80].

Previous studies have shown that flaxseed can improve the metabolic symptoms of PCOS and by modifying ovulation, growth of follicles and sex hormone levels will also affect the problem of infertility of these patients [54-59, 81-88].

3. The effect of flax seeds on fertility

Studies have demonstrated that flaxseed has a beneficial effect on fertility [30, 31, 35-38, 41, 55-59, 62-64, 68]. In this regard, the results of Paul et al.'s study (2021) showed that flaxseed increases fertility in mice by improving the structure of ovaries and ovarian follicles [36]. The results of Himanshu et al.'s study (2022) also showed that flaxseed improved fertility in male rams by increasing the number of sperm and the quality of semen volume [68]. The findings of Jelodar et al.'s study (2018) also showed the positive effect of flaxseed on fertility in rats with polycystic ovary syndrome [54].

While, Saber et al. concluded in their study that flax seed had no effect on the number of fertile eggs and fertility of broiler chickens [61].

4. The effect of flaxseed on cancer

Flaxseed is mainly cultivated as an oil crop and is a rich source of omega-3 fatty acid, alpha-linolenic acid, dietary fiber, plant lignans and a very rich source of nutrients [89]. Flaxseed also has anti-inflammatory effects and is known as a strong antioxidant [90]. Extensive research has emphasized that flaxseed can reduce the severity of ovarian cancer by reducing inflammatory and carcinogenic factors [91]. In the study of Dikshit et al. (2016), the results showed that flaxseed increased the metabolism of estradiol and as a result, the serum level of estradiol decreases and causes a decrease in the severity of ovarian cancer [33].

5. The effect of flaxseed on breastfeeding

Flaxseed contains compounds such as plant phytoestrogens, alpha-linolenic acid and omega-3 fatty acid [36]. The high concentration of alpha-linolenic acid has made flaxseed a common fat supplement in dairy cattle diets [62]. The information obtained about the effect of flaxseed on breastfeeding has been contradictory, so that some studies have reported that consumption of flaxseed increases milk production [62, 63]. While other studies emphasize the lack of effect of flaxseed on breastfeeding [53, 92].

Discussion:

The present systematic review was conducted with the aim of evaluating and summarizing the results of animal studies on the effect of flaxseed on infertility. Based on the criteria of Cochran Group's Risk of Bias tool, most of the studies had a strong

methodology. Among the reviewed studies, 42 articles had the necessary conditions to be included in the present study.

According to the results of most of the reviewed studies (20 articles), flaxseed influences male and female reproductive system. The effects of flaxseed on the male reproductive system include increasing the number of sperm and the quality of semen, increasing libido, protecting sperm against oxidative stress, and increasing the serum level of testosterone and estradiol [30, 31, 35, 36, 39-41, 71]. And its effects on the female reproductive system include increasing the diameter and weight of the ovary, positive effect on ovulation and increasing folliculoneogenesis, improving the quality of eggs and the IVF process, increasing the serum level of estrogen and progesterone, and reducing the serum level of cholesterol [42-50, 72]. Also, flaxseed has a positive effect on metabolic disorders and ovulation disorders in patients with polycystic ovary syndrome and can increase the fertility of these patients [54-59, 81-88]. In addition, this plant has beneficial effects in the treatment of uterine leiomyomas and uterine polyps [52, 86]. However, there have been studies with different results, which indicate that flaxseed has no effect on the level of sex hormones [69] and testicular indices [65, 68].

Among the other effects of flaxseed on infertility treatment is its positive effect on fertility, so the results of the study by Paul et al. (2021) showed that flaxseed improves the structure of the ovary and ovarian follicles and increases fertility in mice [36]. On the other hand, the study by Himanshu et al. (2022) concluded that flax seed improves fertility in male rams by increasing the number of sperm and the quality of semen volume [68].

The findings of the study by Jelodar et al. (2018) are also in line with most of the studies conducted in this field, so that the results of this study report the positive effect of flaxseed on fertility in rats with polycystic ovary syndrome [54]. While Saber et al. concluded in their study that flax seed had no effect on the number of fertile eggs and fertility of broiler chickens [61].

Among the other useful effects of flaxseed in the treatment of infertility, we can refer to the results of the study by Dikshit et al. The results of this study showed that flaxseed increases the metabolism rate of estradiol, and as a result, the serum level of estradiol decreases and causes the severity of ovarian cancer to decrease [33]. Also, flaxseed has a positive effect on increasing milk production and breastfeeding success [62, 63].

Conclusion:

The available evidence shows the effect of flaxseed on the levels of sex hormones in both sexes and subsequently their fertility. But because most of the studies conducted in this field are based on animal samples and there are few clinical trial studies on human samples. It is recommended to conduct more studies on different causes of male and female infertility on human samples with a strong methodology and appropriate sample size to ensure the effect of this plant on humans.

Ethical considerations:

This study was registered by the Research Vice-Chancellor of Shahrekord University of Medical Sciences and has the code of ethics under the number IR.SKUMS.AEC.1402.018.

Financial support:

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Conflict of interest:

There is no conflict of interest among the authors of the present study.

Data availability:

The data collected and the results of this study are available to everyone.

Limitations:

In the current review article, it was not possible to perform a meta-analysis because the methodology of the studies was heterogeneous in terms of quality.

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