Artigo Original

Premenstrual syndrome: clinical assessment of treatment outcomes following Borago officinalis extract therapy

Síndrome da tensão pré-menstrual: avaliação clínica dos resultados de tratamento após terapia com extrato de Borago officinalis

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Unterms: premenstrual syndrome, Borago officinalis, borage oil, g-linolenic acid, physical and emotional symptoms of premenstrual syndrome.

Summary

With the objective of evaluating the use of Borago officinalis oil in the treatment of premenstrual syndrome (PMS) symptoms, we assessed 180 patients who presented a clinical diagnosis of PMS and who were treated with one daily 900mg borage oil capsule. Efficacy and safety assessments were performed based on data obtained prior to the start of treatment (Pretreatment), and after each menstrual cycle (Assessment 2 - first menstrual cycle after the start of treatment; Assessment 3 - second menstrual cycle; Assessment 4/Final Assessment - third menstrual cycle). Efficacy assessments included a PMS questionnaire evaluating emotional and physical PMS symptoms and the Patient and Physician Assessments, in which both the subject and the physician rated the patient's overall condition on a scale of 1-10 points. The primary safety and tolerability measures included any changes in vital signs and physical exam in relation to pretreatment, changes in laboratory exams, and the occurrence of adverse events after the first dose of study medication. We observed a statistically significant reduction in the total scores of the PMS assessment ($p<0.0001$), with

95.4% of all patients displaying some reduction in scores at Assessment 4 in relation to Pretreatment values. The emotional symptoms and physical symptoms scores also decreased significantly from Pretreatment to Assessment 4 ($p<0.0001$ for both). In the Patient and Physician Assessments, there was a statistically significant increase in the both the patient's and physician's scores throughout the study in relation to pretreatment values ($c2= 328.4$; $DF= 4$; $p<0.0001$ for Patient Assessment and $c2= 355.3$; $DF= 24$; $p<0.0001$ for Physician Assessment). The majority of the adverse events (AEs) recorded were mild to moderate in intensity, and none were considered serious. The most common AEs were related to the digestive/gastrointestinal tract, specifically stomach/abdominal upset. Physical assessments did not vary significantly throughout the treatment period. Based on the results of this study, we conclude that the use of Borago officinalis oil was safe and effective in the treatment of the physical and emotional symptoms of premenstrual syndrome in the patient population evaluated.

Introduction

The term Premenstrual Syndrome (PMS) refers to the cyclic group of physical and behavioral symptoms that occur repetitively during the luteal phase of the menstrual cycle and subside after the start of menstrual bleeding1. It is commonly classified as a psycho-neuro-endocrine disorder of unknown etiology, whose symptoms are not caused by organic disease 2.

The symptoms of PMS vary in intensity, ranging from mild discomfort to severe symptoms that significantly impact the patient's daily routine. PMS is common, and it is estimated that 30-40% of all women of reproductive age suffer from symptoms strong enough to impact daily life to some degree, while 3-8% suffer a more severe form of PMS, known as premenstrual dysphoric disorder, in which the disability caused by the symptoms is severe1,3-6.

Over 150 symptoms associated with PMS have been identified, however the number of symptoms commonly observed in affected patients is significantly smaller. Among the most common symptoms (reported among 80-92% of affected patients) are fatigue, irritability, abdominal distension, anxiety/tension, mastalgia, mood swings, and depression. Other common symptoms (reported by 50-80% of affected patients) are cravings for specific types of foods (predominantly chocolate), acne, increased appetite, hypersensitivity to environmental stimuli, swelling, anger, easy crying, feeling of emotional isolation, headache, and memory lapses7. The prevalence of PMS appears to be independent of ethnicity and socioeconomic or cultural conditions 8,9.

Current evidence suggests that PMS arises from the interactions between the cyclic changes in ovarian steroids (progesterone and estrogen) and central neurotransmitters. Serotonin is identified as the principal neurotransmitter involved in the manifestations of PMS, although there is evidence of the involvement of other neurotransmitters, including beta-endorphin, gamma-aminobutyric acid (GABA), as well as the autonomic nervous system. Some of the systemic manifestations, such as abdominal distension, may arise from peripheral mechanisms 10-14.

A correlation between the symptoms of PMS and nutritional deficiency has also been postulated, with controversial results. No difference in vitamins A, E, or B6 levels were detected between women with and without PMS 15-18. While there is evidence that magnesium supplementation may improve PMS symptoms19, no difference was detected between affected and non-affected women in intracellular magnesium levels throughout the menstrual cycle, and studies of serum levels presented diverging results 20-24. Contrary to popular belief, several studies have indicated that stress does not play a central role in the severity of PMS, and it appears more likely that PMS causes stress rather than the possibility of stress causing PMS 25-28.

There is no single diagnostic test to confirm the presence of PMS. Diagnostic confirmation of PMS should take into account the specific symptoms, the timing of the symptoms (they should occur during the luteal phase), symptom severity, as well as the absence of hormone intake, use of narcotic drugs and medications, as well as exclusion of other possible diagnoses. Differential diagnosis for PMS includes premenstrual exacerbation of existing psychiatric disorders, perimenopause, as well as medical conditions such as hypo- or hyperthyroidism. The differentiation between these conditions is important because the therapeutic approach to each condition is individual 7,29,30.

The treatment of PMS depends on the severity of the symptoms. Conservative therapies are often recommended prior to drug therapy, and may offer additional benefits to patients undergoing drug treatment. These include physical exercise to combat stress, tension, anxiety, and depression, as well as relaxation therapy, and dietary modifications. In cases of more severe symptoms, drug therapy may be indicated. The most commonly used medications in the treatment of PMS include the selective serotonin reuptake inhibitors (the most widely used of which is fluoxetine), anxiolytics (the most commonly prescribed is alprazolam), oral contraceptives, spironolactone, and in more severe cases, gonadotropin-releasing hormone agonists, which temporarily interrupt the menstrual cycle 4,31.

The search for non-drug alternatives that are safe and effective is common among patients with moderate symptoms, for a variety of reasons. The use of dietary supplements is frequently adopted by women with moderate symptoms in whom: a) lifestyle changes did not impact PMS symptoms satisfactorily; b) symptoms are not severe enough to warrant drug

treatment; c) there is a preference to avoid long-term drug therapy due to side effects; and d) patients trying to get pregnant32. Although their efficacy has not been confirmed in controlled clinical trials, commonly used dietary supplements include calcium, manganese, magnesium, vitamin B6, vitamin E, multivitamins, carbohydrate supplements, herbal extracts, and essential free fatty acids 32.

Supplements containing long-chain fatty acids include primrose oil, black currant oil, and borrage oil. Borrage oil is extracted from the seeds of the Borago officinalis plant, and its main component is g-linolenic acid (GLA). GLA is an unsaturated omega-6 fatty acid, that in addition to acting as a precursor in prostaglandin synthesis, was experimentally proven to reduce production of interleukin 1-beta (IL-1beta), which may play a role in inflammation and diseases such as rheumatoid arthritis. GLA may also affect cAMP levels which in turn inhibit synthesis of tumor necrosis factor-alpha, the central inflammatory mediator which also regulates joint destructive processes in rheumatoid arthritis 33,34.

Prostaglandins are formed by the conversion of linolenic acid to gamalinolenic acid via arachidonic acid, and a reduction in this conversion has been observed in several clinical conditions, including PMS. The use of borage oil and other similar supplements is based on the theory that women with PMS have a GLA-related deficiency, and that this deficiency can lead to abnormalities in prostaglandin synthesis, which may contribute to the symptoms of PMS 31,32.

Objectives

To evaluate the use of Borago officinalis in the form of borage oil capsules (900mg) in the treatment of the symptomatology specific to PMS. To evaluate the efficacy and safety of the use of borage oil in the treatment of PMS-specific symptoms in terms of clinical assessments and questionnaires completed by the patient and the investigating physician, physical exam results, and incidence of adverse events, including clinically significant changes in laboratory exams.

Material and methods

Based on the medical records of patients presenting PMS and attended atHospital das Clínicas de Teresópolis, Fundação Educacional Serra dos Órgãos until May 2010, 180 subjects were selected for the study, following Ethical Committee approval (approval no. 462-10). Inclusion criteria specified for female patients of reproductive age with a previous clinical diagnosis of PMS, treated with Borago officinalis in the form of 900mg borage oil capsules (standardized to a minimum of 180mg GLA/capsule, one capsule per day, commercially available in Brazil as Gamaline V - Herbarium) for a minimum of three menstrual cycles, and who were not pregnant or breastfeeding and using adequate birth control. Patients presenting hypersensitivity to borage oil, who had received medication for treatment of specific PMS symptoms within 60 days of the pretreatment assessment, and patients with a previous diagnosis of premenstrual dysphoric disorder were excluded from the study.
The clinical research form contained physical exam and clinical laboratory test results obtained from before, during, and at the conclusion of treatment. Efficacy assessments were performed based on data obtained prior to the start of treatment (Pretreatment), and after each menstrual cycle (Assessment 2 - first menstrual cycle after the start of treatment; Assessment 3 - second menstrual cycle; Assessment 4/Final Assessment - third menstrual cycle).

The primary efficacy measures included a PMS questionnaire evaluating emotional and physical PMS symptoms. Patients assessed the presence of symptoms with: "0" if the symptom was absent; "1" if the symptom was barely noticeable; "2" if the symptom inhibited activities; and "3" if the symptom altered their lifestyle. The highest possible score for the emotional symptom checklist was 54 points, and the highest possible score for the physical symptom checklist was 45. The sum of the emotional + physical symptom checklist was recorded as the total score of the questionnaire.

Secondary efficacy measures included the Patient and Physician Assessments, in which both the subject and the physician rated the patient’s overall condition on a scale of 1-10 points, with "1" corresponding to the worse assessment and "10" the best. At Assessment 4, the patient’s willingness to continue treatment with the borage oil capsule was also rated on a scale of 1-10 points, with "10" corresponding to most willing to continue treatment. At Assessment 4, the study physician also evaluated the overall efficacy of the study medication as "Very Good", "Good", "Fair", or "Poor".

The primary safety and tolerability measures included any changes in vital signs and physical exam in relation to pretreatment, and any changes in clinical laboratory exams in relation to pretreatment, and the occurrence of adverse events after the first dose of study medication. Any laboratory exams out of reference range were recorded as adverse events. The secondary safety measure was the evaluation of overall tolerability of the study medication performed at Assessment 4 by the study physician, using the same classifications of "Very Good", "Good", "Fair", or "Poor" as were used for the overall efficacy assessment.

The clinical research form was filled, coded and the data were analyzed using GraphPad Prism v. 5.1. software. Frequency tables were generated and central tendency measures were calculated (mean, median, mode). As appropriate, we used the Student’s T-test or the repeated-measures analysis of variance (ANOVA) for continuous variables and Fisher’s test or

the c2 test for categorical variables. Results were compared between each assessment and throughout the study.

Efficacy endpoints included the percentage of patients with a reduction in the total score of the PMS questionnaire, as well as in the individual emotional and physical symptoms scores. The percentage of patients with Patient and in the Physician's Assessments scores of 8-10 at the final Assessment were also efficacy endpoints, along with the percentage of patients receiving an assessment of "Very Good" in the overall assessment of efficacy performed at the study end by the investigating physician.

The safety endpoints for this study included the percentage of patients presenting adverse events, the percentage of patients presenting laboratory alterations, and the percentage of patients receiving an assessment of "Very Good" in the overall assessment of tolerability performed at the study end by the investigating physician.

Table 1 - Demographic and baseline characteristics

Observation	Result
Age (years)	30.1 (±)7.43
Ethnicity	
Asian	4
Black	32
Caucasian	67
Mulatto	77
Marital Status	
Divorced	28
Married	73
Single	74
Widowed	5
Dietary habits	
Good	113
Moderate	63
Poor	4
Smoking	
Nonsmoker	104
< 10 cigarettes / day	42
≥ 10 cigarettes / day	34
Alcohol consumption	
None	103
< 2 drinks/day	58
≥ 2 drinks /day	19
Physical exercise	
Regular (at least 1x/week)	51
Irregular	75
None	51

Results

A total of 180 patients were included in the study, in accordance with the protocol. Figure 1 demonstrates the flow of patients through the study. Table 1 summarizes the demographic and baseline data collected at the start of the study. The most common type of contraceptive reported was hormonal pills, followed by intrauterine devices, diaphragm, condoms, and calendar-based contraception. At Pretreatment, 43 patients (23.9%) reported dysmenorrhea, while 8 (4.4%) patients reported intermenstrual bleeding, and 3 (1.7%) reported amenorrhea.

Table 2 - Safety measures

Variable	Pretreatment	Assessment 2	Assessment 3	Assessment 4
Weight (kg)	61.06 (±6.42)	61.09 (±6.47)	61.22 (±6.47)	61.24 (±6.5)
BMI	23.54 (±2.37)	23.57 (±2.38)	23.64 (±2.46)	23.66 (±2.42)
Systolic BP (mmHg)	119.5 (±9.05)	119.4 (±9.65)	119.9 (±9.73)	119.7 (±9.19)
Diastolic BP (mmHg)	72.57 (±10.24)	72.09 (±10.44)	72.04 (±11.02)	71.94 (±11.28)
Heart rate (bpm)	65.19 (±6.05)	64.60 (±5.98)	65.22 (±54.18)	65.44 (±5.89)

Data are mean (+SD).

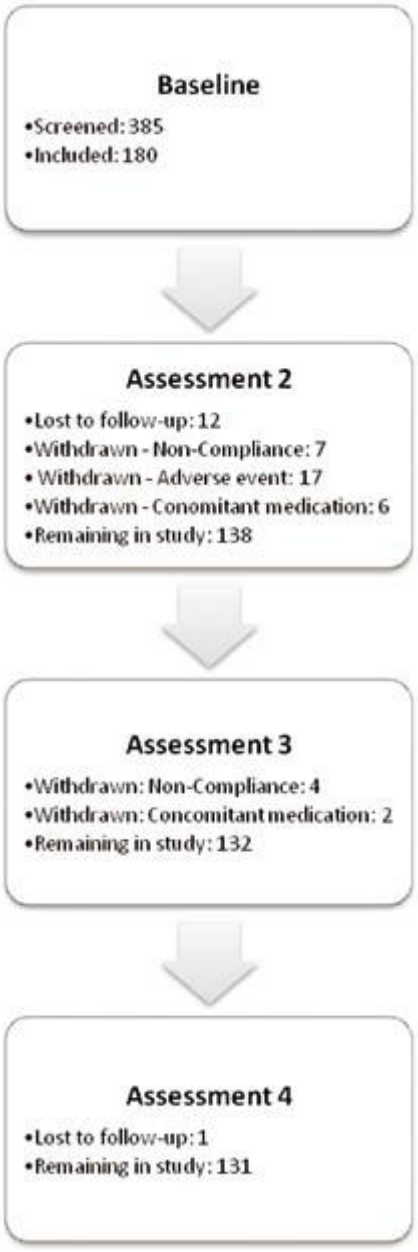

Figure 1 - Flowchart of patients in the study.

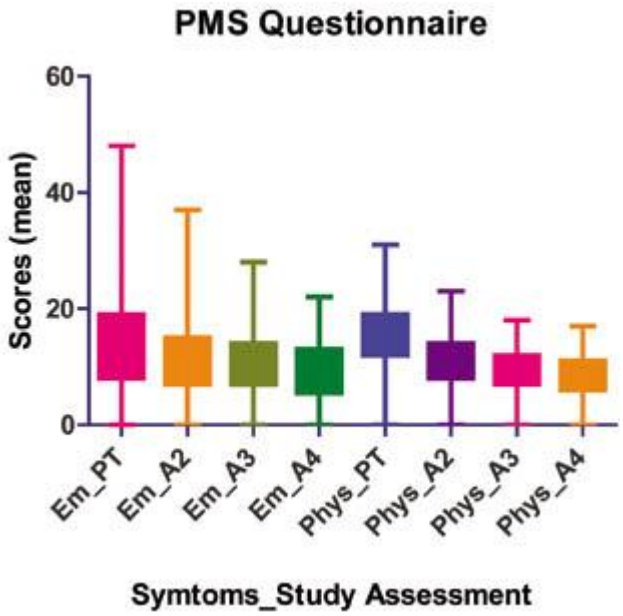

Figure 2 - PMS questionnaire scores.

The total scores of the PMS assessment decreased in a statistically significant manner from Pretreatment to Assessment 4 ($p<0.0001$), with a total of 95.4% of all patients displaying some reduction in scores at Assessment 4 in relation to Pretreatment values. Figure 2 summarizes the results of the PMS questionnaire scores for emotional symptoms and physical symptoms at each study assessment. The mean emotional symptom scores decreased significantly from Pretreatment to Assessment 4 ($p<0.0001$). At Assessment 4, 77.9% of patients showed improved emotional symptom scores in relation to Pretreatment values, with a mean percentage score reduction of 27.69%. The physical symptom scores of the PMS questionnaire also decreased significantly from Pretreatment to Assessment 4 ($p<0.0001$), with 93.9% of patients showing improvement. The percentage score reduction was 42.3% from Pretreatment to Assessment 4 in the PMS questionnaire scores of physical symptoms.

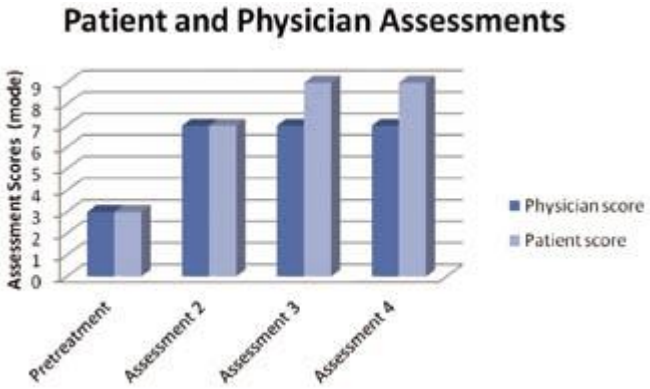

Figure 3 - Patient and physician assessment of overall condition.

Table 3 - Adverse events	
Adverse Event	**Number of affected patients**
Abdominal discomfort	2
Abdominal distension	3
Diarrhea	3
Diuresis	1
Eructation	6
Flatulence	4
Halitosis	1
Headache	3
Heartburn	1
Increased number of evacuations	1
Indigestion	2
Laboratory alteration	7
Meteorism	9
Migraine	1
Nausea	11
Palate changes	1
Pruritus	1
Smelly stool	1
Soft stool	4
Stomach pain	3
Vomiting	7

Data are n.

Figure 3 summarizes the results of the evaluations of overall condition performed by the patients and the study physician at each study assessment. There was a statistically significant increase in the patient's scores throughout the study in relation to pretreatment values (c2= 328.4; DF= 4; p<0.0001), with 48.1% of patients responding with scores of 8-10 at Assessment 4. The scores of the physician's assessment of overall condition also showed significant improvement in relation to pretreatment scores (c2= 355.3; DF= 24; p<0.0001). At Assessment 4, 43.5% of the patients received scores of 8-10 in the physician's assessment of overall condition.

The physician's assessment of overall efficacy at Assessment 4 was given as "Very Good" for 42 (32.06%) patients, "Good" for 50 (38.17%) patients, "Acceptable" for 22 (16.79%), and "Poor" in 17 (12.97%) patients.

Table 2 summarizes the results of the physical evaluations performed at each assessment and used as safety measures. Throughout the 4-month treatment period, there were no significant changes in weight (p=0.598), body mass index (BMI) (p=0.508), blood pressure (BP) (p=0.432 for systolic BP; p=0.055 for diastolic BP), or pulse (p=0.331).

A total of 39 (21,7%) patients presented adverse events (AEs), for which 17 patients were withdrawn from treatment. The AEs observed are summarized in Table 3. The majority of the AEs recorded (77.7%) were mild to moderate in intensity, and none were considered serious. The most common AEs were related to the digestive/gastrointestinal tract, specifically stomach/abdominal upset. The laboratory alterations recorded were as follows: elevated transaminases (ALT/AST) (n=3); elevated blood glucose (n=2); elevated amylase, bilirubins, and BUN (n=1 for each).

Overall tolerability was considered "Very Good" among 50 (38.17%) subjects in the physician's assessment of overall tolerability, while it was considered "Good" in 55 (41.98%) patients, "Acceptable" among 17

(12.97%) patients, and "Poor" in 9 (6.87%) patients.

At the end of the treatment period, subjects who completed the treatment cycle were asked to rate their willingness to continue treatment on a scale of 1 (very unwilling) to 10 (very willing). A total of 70 patients (53.44%) responded with scores of 9-10.

Discussion

The results of this study indicate a beneficial effect of borage oil in the treatment of PMS symptoms. While borage oil is widely marketed for use in the treatment of PMS symptoms, references in the literature of studies specific for this indication are scarce. Much of what is known about GLA is based on studies involving evening primrose oil, derived from the seeds of the Oenothera biennis plant, which contains a slightly lower GLA concentration in relation to borage oil35.

The oil extracted from the seeds of the Borago officinalis plant is a rich source of g-linolenic acid (GLA), an essential fatty acid of the omega-6 series which represents the first product of the n-6 polyunsaturated fatty acid pathway. The underlying mechanism of action of GLA is believed to result from its downregulation of prostaglandin E2 production, which takes place by a rapid conversion of GLA to dihomo-g-linolenic acid (DGLA). This conversion increases PGE1 production, and consequently increases intracellular cAMP levels, which in turn inhibits phospholipase, thus limiting the release of arachidonic acid (AA)32.

GLA and its biosynthesis are crucial to n-6 polyunsaturated fatty acid metabolism. GLA is synthesized in mammals from dietary linoleic acid by the action of D6-desaturase, a rate-limiting enzyme. It is then converted to DGLA through the action of a polyunsaturated fatty acid-specific elongase. The enzyme D5-desaturase converts DGLA to arachidonic acid, but both DGLA and arachionic acid can be metabolized to form eicosanoids (including prostaglandins). While oxidation of DGLA yields 1-series of prostaglandins by cyclooxygenase, arachidonic acid is converted to 2-series prostaglandins (also by cyclooxygenase) or 4-series leukotrienes (by 5-lipoxygenase). These metabolites are essential in the regulation of many biological activities, and also exert modulatory effects in a variety of diseases. They act in suppression of chronic inflammation, inhibition of platelet aggregation and thrombosis, suppression of vasodilation, lowering of blood pressure, and also inhibit the development of smooth muscle cell proliferation-associated atherosclerotic plaque 32,35,36.

Reduced conversion rates of LA to GLA and consequent reduction in production of n-6 polyunsaturated fatty acids has been attributed to symptoms associates various conditions, including diabetes, aging, atopic dermatitis, rheumatoid arthritis, alcoholism, cancer, cardiovascular disease, and PMS. These observations have led to the theory, corroborated in clinical studies, that dietary supplementation with GLA which effectively bypasses D6-desaturation, effectively alleviates many of these symptoms 36.

At the end of the study, 95.4% of the patients displayed reduction in the total PMS assessment scores in relation to pretreatment values. In the analysis of the individual sub-items (emotional symptoms and physical symptoms), while there was a statistically significant score improvement at Assessment 4 in relation to pretreatment for both items, a higher percentage of patients displayed improvements in the scores of the physical symptoms section of the assessment (93.9%) as compared to the percentage of patients displaying improvement in the emotional symptoms (77.9%). The physical symptoms assessed in the PMS questionnaire included pain and discomfort such as lower back pain, headache, and breast tenderness common among women suffering from PMS. The anti-inflammatory properties of the borage oil may have better addressed these symptoms as compared to the emotional symptoms assessed, which included mood swings, anxiety, and irritability, among others.

With regards to the adverse effects recorded during treatment, the majority of these affected the GI tract, and were mild or moderate in severity. Gamolenic and linoleic acids from evening primrose oil, and presumably similar sources such as borage oil, have been reported to produce minor gastrointestinal disturbances and headache35,37. The laboratory alterations recorded above reference ranges were transitory and none were considered severe. However, long-term dietary supplementation with essential fatty acids should take into account the effect of these compounds on lipid indexes.

Although some previous studies of polyunsaturated fatty acids from evening primrose oil or other GLA-rich supplements in the treatment of PMS symptoms have had controversial results, these may have been due to variations in the concentrations of the s used, as well as flaws in the trial designs 38-41. However, in a recent study, Rocha Filho et al. (2011) reported improvement in PMS symptoms using the Prospective Record of the Impact and Severity of Menstruation (PRISM) calendar among patients receiving 1 or 2g daily of polyunsaturated fatty acids (at a cyclical dosing schedule of a 15-day treatment period beginning on the 15th day of the menstrual cycle). The results of this double-blind, placebo-controlled study demonstrated a significant symptomatic improvement among subjects who received the active treatment, with a greater improvement among subjects who received the higher dose. The authors also monitored cholesterol levels throughout the 6-month treatment period, reporting no change in laboratory parameters 42.

Conclusion

Based on the results of this study, we conclude that the use of Borago officinalis oil was safe and effective in the treatment of the physical and emotional symptoms of premenstrual syndrome in the patient population evaluated.

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Bibliografia

1. Steiner M, Born L. Diagnosis and treatment of premenstrual dysphoric disorder: an update. Int Clin Psychopharmaol 2000; 15(Suppl 3):S5-S17.
2. Friedman DE, Laffe A. Influence of lifestyle on the premenstrual syndrome. J Reprod Med 1985; 30: 715-719.
3. ACOG: Premenstrual Syndrome (ACOG Committee opinion). Int J Gynaecol Obstet 1995; 50:80-84.
4. Daugherty JE. Treatment strategies for premenstrual syndrome. Am Fam Phys 1998; 58:183-192.
5. Borenstein J, Chiou CF, Dean B, et al. Estimating direct and indirect costs of premenstrual syndrome. J Occup Environ Med 2005; 47:26-33.
6. Deuster PA, Adera T, South-Paul J. Biological, social, and behavioral factors associated with premenstrual syndrome. Arch Fam Med 1999; 8:122-128.
7. Mortola JF, Girton L, Beck L, et al. Diagnosis of premenstrual syndrome by a simple prospective reliable instrument. Obstet Gynecol 1990; 76:302-307.
8. Mongale L, Dan A, Krogh V, et al. Perimenstrual symptom prevalence rates: an Italina-American comparison. Am J Epidemiol 1993; 138:1070=1081.
9. Raja SN, Feehan M, Santon WR, et al. Prevalence and correlates of the premenstrual syndrome in adolescence. J Am Acad Child Adolesc Psychiatry 1992; 31:783-789.
10. Budieri DJ, Li-Wan Po A, Dornan JC. Clinical trials of treatment of premenstrual syndrome: entry criteria and scales for measuring treatment outcomes. Br J Obstet Gynaecol 1994; 101:689-695.
11. Schmidt PJ, Nieman LK, Danaceau M, et al. Differential behavioral effects of gonadal steroids in women with an in those without premenstrual syndrome. N Engl J Med 1998; 338:209-216.
12. Majewska MD, Harrison ML, Schwartz RD, et al. Steroid hormones are barbiturate-like modulators f the GABA receptor. Science 1986; 232:1004-1007.
13. Bethea C. Regulation of progestin receptors in raphe neurons of steroid-treated monkeys. Neuroendocrinology 1994; 60:50-61.
14. Warldlaw SL, Thoron L, Frantz AG. Effects of sex steroids on brain beta-endorphin. Brain Res 1982; 245:327-331.
15. Chuong CJ, Dawson EB, Smith ER. Vitamin A levels in premenstrual syndrome. Fertil Steril 1990; 54:643-647.
16. Chuong CJ, Dawson EB, Smith ER. Vitamin E levels in premenstrual syndrome. Am J Obstet Gynecol 1990; 163:1591-1595.

17. Stewart A. Vitamin B6 in the treatment of the premenstrual syndrome: Review. Br J Obstet Gynaecol 1991; 98:329-330.
18. Kleijnen J, Ter Reit G, Knipschild P. Vitamin B6 in the treatment of premenstrual syndrome - a review. Br J Obstet Gynaecol 1990; 97:847-852.
19. Facchinetti F, Borella P, Sances G, et al. Oral magnesium successfully relieves premenstrual mood changes. Obstet Gynecol 1991; 78:177-181.
20. Sherwood RA, Rocks BF, Steward A, et al. Magnesium and the premenstrual syndrome. Ann Clin Biochem 1986; 23:667-670.
21. Facchinetti F, Borella P, Fioroni L, et al. Reduction of monocyte's magnesium in patients affected by premenstrual syndrome. J Psychosom Obstet Gynaecol 1990; 11:221-229.
22. Rosenstein DL, Elin RJ, Hosseini JM, et al. Magnesium measurements across the menstrual cycle in premenstrual syndrome. Biol Psychiatry 1994; 35: 557-561.
23. Cerin A, Collins A, Landgren BM, et al. Hormonal and biochemical profiles of premenstrual syndrome. Treatment with essential fatty acids. Acta Obstet Gynecol Scand 1993; 72:337-343.
24. Posaci C, Erten O, Uren A, et al. Plasma copper, zinc and magnesium levels in patients with premenstrual tension syndrome. Acta Obstet Gynecol Scand 1994; 73:452-455.
25. Fontana AM, Palfai TG. Psychosocial factors in premenstrual dysphoria: stressors, appraisal, and coping processes. J Psychosom Res 1994; 38:557-567.
26. Beck LE, Gevirtz R, Mortola JF. The predictive role of psychosocial stress on symptom severity in premenstrual syndrome. Psychosom Med 1990; 52: 536-543.
27. Gallant SJ, Popiel DA, Hoffman DM, et al. Using daily ratings to confirm premenstrual syndrome/late luteal phase dysphoric disorder. Part II. What makes a "real" difference? Psychosom Med 1992; 54:167-181.
28. Gallant, SJ, Popiel, DA, Hoffman, DM, et al. Using daily ratings to confirm premenstrual syndrome/late luteal phase dysphoric disorder. Part I. Effects of demand characteristics and expectations. Psychosom Med 1992; 54:149-166.
29. American Psychiatric Association. Diagnostic and statistical manual Edition IV, American Psychiatric Association, Washington, DC. 1994; p. 715.
30. Hay AG, Bancroft J, Johnstone EC. Affective symptoms in women attending a menopause clinic. Br J Psychiatry 1994; 164:513-516.
31. Johnson SR. Premenstrual syndrome therapy. Clin Obstet Gynecol 1998; 41:405-421.
32. Bendich A. The potential for dietary supplements to reduce premenstrual syndrome (PMS) symptoms. J Am Col Nutrition 2000; 19(1):3-12.
33. Kast RE. Borage Oil Reduction of Rheumatoid Arthritis Activity May Be Mediated by Increased cAMP That Suppresses Tumor Necrosis Factor-Alpha. Int Immunopharmacol, 2001, 1(12):2197-2199.
34. Furse RK, Rossetti RG, Seiler CM, et al. Oral Administration of Gammalinolenic Acid, an Unsaturated Fatty Acid With Anti-Inflammatory Properties, Modulates Interleukin-1Beta Production by Human Monocytes. J Clin Immunol 2002; 22(2):83-91.
35. Sweetman SC, ed. Martindale: The complete drug reference. 37th ed. London: Pharmaceutical Press, 2011.
36. Huang YS, Ziboh VA. Gamma Linolenic Acid: Recent advances in biotechnology and clinical applications. AOCS Publishing, 1st ed. 2001. 259pp.
37. Foster S, Tyler VE. Tyler's Honest Herbal. 4th ed. New York: Haworth Press, 1999.
38. Budeiri D, Li Wan Po A, Dornan JC. Is evening primrose oil of value in the treatment of premenstrual syndrome? Control Clin Trials 1996; 17(1):60-68.
39. Labruzzo BA, Chausk R, Kendall S. Which complementary therapies can help patients with PMS? J Fam Pract 2009; 58(10):552-559.
40. Collins A, Cerin A, Coleman G, et al. Essential fatty acids in the treatment of premenstrual syndrome. Obstet Gynecol 1993; 81(1):93-98.
41. Khoo SK, Munro C, Battistutta D. Evening primrose oil and treatment of premenstrual syndrome. Med J Aust 1990; 153(4):189-192.
42. Rocha Filho EA, Lima JC, Pinho Neto JS, et al. Essential fatty acids for premenstrual syndrome and their effect on prolactina and total cholesterol levels: a randomized, double-blind, placebo-controlled study. Reprod Health 2011; 8(1):2.