

Effect of Borago Officinalis Extract on Moderate Persistent Asthma: A Phase two Randomized, Double Blind, Placebo-Controlled Clinical Trial

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Background: Borago officinalis and its derivatives are used in folk medicine to treat asthma because of its special effect on allergic disorders. It suppresses the tumor necrosis factor-alpha (TNF-alpha) and delivers gamma-linolenic acid. The objective of this clinical trial was to determine the effect of Borago officinalis on clinical and physiological findings in moderate persistent asthma.

Materials and Methods: This prospective, randomized, double blind, placebo-controlled, clinical trial was conducted on patients aged 15-90 years with moderate asthma and forced expiratory volume in one second (FEV1) of 60-79% of predicted who presented to a sub-specialty clinic of pulmonary medicine. We randomly allocated subjects to receive either Borago extract (5 mL three times a day) or a matched placebo for one month. The primary outcome was the asthma control test (ACT) score and fractional exhaled nitric oxide (FENO) test. Secondary outcomes included clinical findings, spirometry, and sputum cytology including inflammatory cells.

Results: Thirty-eight subjects with a mean age of 46.8±15.3 years and mean duration of asthma of 71±103 months were enrolled in our study. Cough, dyspnea, wheezing, nocturnal symptoms, and airway hyper-responsiveness reduced significantly in the Borago group after the treatment and ACT scores improved significantly (10.8±5.26 before and 15.4±5.12 after the trial). Flare up of asthma and emergency department visits in the Borago group also decreased significantly (3.6±2.33 to 2±1.86 flare ups per month and 0.62±0.9 to 0.05±0.23 for emergency department visits per month). Physiological parameters including spirometry, FENO, and sputum cytology including eosinophil and neutrophil did not change significantly.

Conclusion: Borago improved the clinical findings of asthma, but it was not able to suppress the inflammation involved in asthma.

Key words: Borago officinalis, Echium amoenum, Asthma, Gamma-linolenic acid, Tumor necrosis alpha

INTRODUCTION

Current treatments of asthma are usually directed at suppressing bronchial eosinophilic inflammation. Many different treatments have been used in folk medicine for asthma and the mechanisms of action of traditional medicines may introduce new strategies for treatment of resistant asthma.

Borago officinalis and its Iranian species Echium amoenum contain several fatty acids including palmitic, linoleic, stearic, and γ-linolenic acids (1). The flower of this plant is traditionally used in folk medicine as a bronchodilator (1). Several mechanisms have been

described for this herbal drug; some of them are involved in asthma and allergic reactions. The anti-oxidative activity of herbal drugs was evaluated in Mediterranean herbal drugs, and *Borago officinalis* was found to contain the highest amount of sterols with anti-oxidative and anti-inflammatory activities (2). Thus, *Borago officinalis* has been effectively used for the topical treatment of atopic dermatitis (3). The authors attributed the defective production of gamma-linolenic acid (GLA) to impaired activity of delta-6 desaturase enzyme that could be compensated by use of *Borago* extract rich in GLA. The benefit of *Borago officinalis* in treatment of inflammation was more than its local effect on atopic dermatitis. Its GLA content is able to suppress TNF-alpha via increasing cAMP and prostaglandin E levels in rheumatoid arthritis (4).

TNF has been proven to increase in uncontrolled asthma (5). Although TNF blockers (infliximab, etanercept and adalimumab) are not listed as standard therapy for asthma, they were used in experimental models of asthma and a small case series (6), but their wide range of side effects and cost prevent their wide spread use for asthma (7). *Borago* extract in this regard shows some benefits and by limiting its dosage we can control its side effects. A recent study showed its effect on arachidonic acid metabolism, which is very important in asthma (8).

According to the promising results of experimental studies (7,8), use of *Borago* extract in clinical trials has been recommended. The aim of this study was to evaluate the effect of *Borago* extract on asthma.

MATERIALS AND METHODS

a. Participants:

Forty-two asthmatic patients older than 15 years were enrolled in this study. Two criteria were used to confirm the diagnosis of asthma: a) positive history of airway hyper-reactivity and history of recurrent episodes; and b) significant post-bronchodilator improvement (9).

Included subjects had to have moderate persistent asthma that was determined by two criteria: 1- moderate stage asthma proven by respiratory symptoms including

daily cough, wheezing and/or dyspnea, 2- spirometry that revealed an obstructive pattern and FEV1 in the range of 60-79% of predicted value. Exclusion criteria: 1- severe asthma due to frequent acute attacks of severe asthma. 2- Having a positive history of respiratory tract infections, bronchiectasis, sinusitis, vocal cord dysfunction (evaluated by plateau in the flow volume curve or MEF50%/MIF50%>2.2) (10), and cigarette or water pipe smoking. All patients gave their written informed consent.

b. Study design

The study was a prospective, phase 2b randomized, double blinded, placebo-controlled, parallel group clinical trial. The ethical committee of our university approved the study and it was registered in the Iranian clinical trial registry (IRCT201209302695N4).

c. Randomization and masking:

Computer generated random tables were used to randomly divide subjects into *Borago* extract and placebo groups. Assignment of subjects to each group was done by an independent assistant who was blinded to the study groups.

d. Herbal drug and placebo:

Hydroalcoholic extract of *Borago officinalis* was obtained from 8.4 kilograms of the flower and leaves of *Borago officinalis* in the form of a syrup by a local herbal drug company (Exir Gol Sorkh Company, Mashhad, Iran). The herbs were percolated using 4 L of water and 3.5 L of ethanol in 40-45°C for three hours. The placebo was produced and bottled identically in shape, color and size to the original drug (provided by the same company). Before the trial, a sample of the herbal drug was tested for level of effective and toxic alkaloids by a pharmacist. The dosage was one 5 cc administered by spoon three times a day for one month.

e. Blinding:

The drug and placebo were coded by a blinded colleague and the drugs were prescribed by another pharmacist who was blinded to the code of the drug and

placebo. This pharmacist kept the sealed code of the package until the end of the trial and the code was opened only in case of emergencies. Patients and clinical investigators were unaware of group assignment throughout the study and outcome variables were evaluated by physicians and technicians who were unaware of the studied groups. Inhaled salbutamol was allowed to be used freely by patients as a reliever medication throughout the study.

f. Outcome variable measurement:

Primary end points were improvement in the score of the ACT questionnaire, which is a valid questionnaire for evaluating the activity of asthma (11) and translated to Farsi and improvement in the FENO score, which was measured by No Breath (Bedfont Medical Instruments, London, England). Secondary end points were frequency of complete improvement of cough, dyspnea, changes in spirometry parameters including the FEV1, FEV1/FVC, and fraction of inflammatory cells in sputum. Spirometry was done by a turbine spirometry device (Superspiro, Micomedical Company, London, UK) according to the American Thoracic Society/European Respiratory Society guidelines (12).

g. Sputum collection:

As the first step, we asked the subjects to expel sputum from their throat and try to expectorate sputum from their lungs. In case of no sputum, sputum induction was carried out by 5% saline inhalation in a compressor type nebulizer (CX3, Omron, Japan) according to the European Respiratory Society guidelines (13).

h. Sputum processing:

Sputum preparation and decontamination of the saliva were performed by a liquid base commercial kit (E-prep Plus sol, Tehran, Iran). Two slides were prepared and stained by Papanicolaou stain for each subject and 300 non-squamous cells were counted in each slide. The mean values of the results of the two slides were recorded. A predefined method for classification of inflammatory cells was used (10,14) as follows: subjects with eosinophilic

percentage more than 3% were classified as eosinophilic, neutrophilic percentage more than 76% as neutrophilic, both of them as mixed type and none of them as paucigranulocytic.

i. Time of outcome measurement:

The outcome variables were assessed at the time of the first visit and over the 30-day period of treatment. Frequency of side effects to Borago extract and acute attacks including emergency department visits were evaluated at the final step of assessments.

j. Follow up:

Patients were followed up every two weeks by phone calls. Every subject who complained of coughing and unexpected side effects were excluded from the study and started on standard asthma treatments.

k. Statistical analysis

Sample size was calculated to detect 20% difference of primary outcome (ACT score) between Borago and placebo groups by means of 80% power and less than 5% error. Normal distribution was assessed by the Kolmogorov-Smirnov test. Comparison of outcome measures between the Borago and placebo groups was done by chi square, Fisher's exact, Mann-Whitney U (for non-normally distributed subjects), and student t tests (for normally distributed subjects). The treatment results were analyzed by McNemar's test, ANCOVA, paired t and Wilcoxon signed ranks tests. SPSS 19 was used for statistical analysis. All tests were two-sided and level of significance was set at $P < 0.05$.

RESULTS

a. Basic demographical findings

Forty-two subjects suffering from moderate stage asthma were recruited in this study, and 38 subjects (15 females and 23 males, mean age of 46.8 ± 15.3 years) completed the course of study. Most of them were living in the city and only nine subjects reported exposure to air pollution. Mean age, distribution of gender, occupation and exposure to air pollution were not significantly

different between the two groups. The mean duration of asthma from the first manifestation of the disease was 71 ± 103 months (range: 1-480 months) with no significant difference between the two groups (81 ± 120 months in the Borago group and 61 ± 77 months in the placebo group, $P=0.77$).

b. Comparison of outcome variables between the Borago and placebo groups

Before the trial: The most frequent symptoms before the trial in both groups were dyspnea, cough, nocturnal symptoms, and wheezing on physical exam (Table 1). All subjects reported aggravation of symptoms after exercise, and sputum and airway hyper-reactivity, which were mentioned by more than half of subjects. Gastro-esophageal reflux (GERD) and post-nasal drip (PND) as adjunct clinical findings was reported in less than half of the subjects. The mean number of attacks per week and emergency department visits per month were 3.6 ± 2.33 and 0.62 ± 0.9 , respectively in the Borago group and 3.63 ± 2.87 and 0.53 ± 0.83 , respectively in the placebo group, which were not significantly different. The differences in ACT scores between the two groups before the trial were not significant (Table 2).

Table 1. Comparison of effects of hydro-alcoholic extract of Borago with the placebo on demographic and clinical findings of asthmatic subjects

	Total	Before trial		After trial	
		Borago	Placebo	Borago	Placebo
Cough	33 (86%)	18 (95%)	15 (79%)	3 (16%)*†	16 (84%)
Dyspnea	37 (98%)	18 (95%)	19 (100%)	8 (42%)*†	18 (94%)
Wheezing	37 (97%)	19 (100%)	18 (95%)	13 (68%)*	17 (89%)
Sputum	22 (58%)	13 (68%)	10 (53%)	7 (36%)‡	13 (68%)
Nocturnal Symptoms	32 (84%)	16 (84%)	16 (84%)	10 (52%)*	16 (84%)
AHR	24 (63%)	13 (68%)	11 (58%)	7 (36%)*†	11 (58%)
GERD	19 (50%)	8 (42%)	11 (58%)	5 (26%)‡	12 (63%)
PND	15 (40%)	6 (31%)	9 (47%)	5 (26%)	9 (47%)
Aggravation After exercise	38 (100%)	19 (100%)	19 (100%)	15 (79%)	19 (100%)

AHR= Airway hyper-responsiveness, GERD= Gastro-esophageal reflux, PND= Post- nasal drip

*= Significant difference before and after the trial in the Borago group (McNemar's test)

†= Significant difference between Borago and the placebo after the trial (chi square test)

‡= Significant difference after the trial in the placebo group (McNemar's test)

Table 2. Comparison of overall asthma control, spirometry and physiological evaluation of subjects enrolled in the trial for treatment of moderate persistent asthma by Borago extract.

	Before trial		After trial	
	Borago	Placebo	Borago	Placebo
FVC (L)	2.4±0.5	2.2±0.8	2±0.6	2.4±0.8
FVC percent	70.4±11.2	71.8±11.7	75.8±9.5	72.8±9.8
FEV1 (L)	1.75±0.43	1.66±0.54	1.8±0.7	1.5±0.5
FEV1 percent	66.6±8.8	64.6±8.2	63.9±15.6	60.5±3.2
FEV1/FVC	73.3±7.4*	75.3±7.3	72.1±7.5	76.7±11.3
FEV1/VC	8.3±12.4	81±15.3	73.2±16	73.3±10.3
FEF25-75 (L/S)	1.32±0.3	1.36±0.5	1.47±1.6	1.42±0.7
FEF25-75 percent	37±11.5	38.8±8.9	38.5±17.5	37.9±15.4
FEF25-75/FVC	0.54±0.15	0.63±0.17	0.58±0.2	0.67±0.22
PEFR (L/S)	2.2±0.7	2.3±1.4	3.1±1.9	3.2±2.6
PEFR percent	45±13.3‡	52.3±22.4	47.2±17.7	53.2±35.4
FENO (PPM)	56.8±45.2	51.3±46.4	62.3±43.3	52.7±37.1
ACT	10.8±5.26	13.1±4.3	15.4±5.12*†	11.2±5.62
Acute attack	3.6±2.33	3.63±2.87	2±1.86*	3.42±2.87
Emerg. Dep. Visit	0.62±0.9	0.53±0.83	0.05±0.23*	0.42±1.17

*= Significant difference before and after the trial in the Borago group (paired t test)

†= Significant difference between Borago and the placebo groups after the trial (student t test or Mann-Whitney U test)

‡= Significant difference after the trial in the placebo group (paired t test)

After the trial: Cough, dyspnea, wheezing, nocturnal symptoms and airway hyper-responsiveness significantly decreased in the Borago group (Table 1). ACT scores also improved significantly (Table 2) and patients' reports of asthma acute attacks and emergency department visits decreased in the Borago group. Sputum production, symptoms aggravated by exercise, GERD, PND, spirometry parameters and FENO did not change significantly in the two groups (Tables 1 and 2).

c. Comparison of Borago with the placebo after the trial

Clinical findings including cough, dyspnea, sputum production, airway hyper-responsiveness, and GERD improved significantly in the Borago group compared to the placebo. ACT scores were also significantly higher in the Borago group, but other clinical findings including acute attack, spirometry parameters and FENO were not significantly different with the placebo group (Tables 1 and 2).

Comparison of sputum inflammatory cells in Borago and placebo groups before and after the trial showed no significant difference in proportion of inflammatory cells. However, there were significant increments in lymphocyte percentage in the Borago group after the trial compared to lymphocyte in the Borago group before the trial and placebo group after the trial (Figure 1).

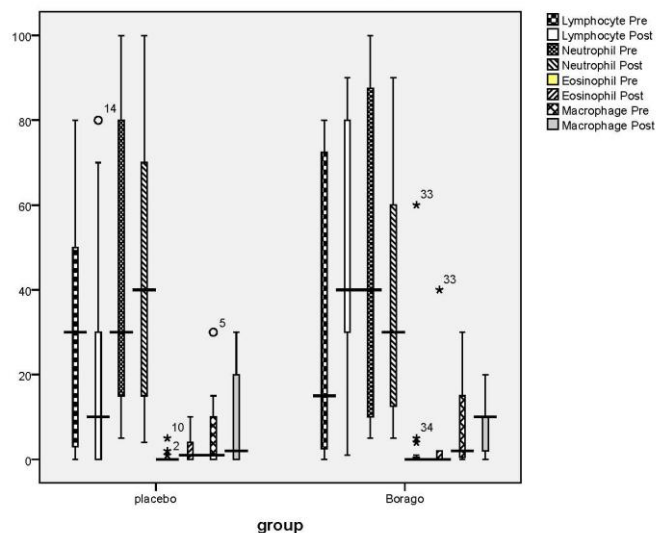


Figure 1. Comparison of sputum inflammatory cells in Borago and placebo groups

Neutrophils were the most frequent inflammatory cells found in the sputum of both groups before the trial. Neutrophil count decreased in the Borago group after the trial, although it was not significant (Figure 1). Eosinophils as the most important inflammatory cells were higher in the Borago group ($3.2 \pm 1.4\%$) than the placebo group ($2.6 \pm 2.7\%$), but this difference was not significant. Frequency of macrophages was in the range of 6.5-8%, which did not change after the trial in both groups.

Figure 2 shows the cumulative classification of inflammatory cells into four major inflammatory classifications. In the case group, the neutrophilic pattern including neutrophilic and mixed patterns decreased, and in the other groups, eosinophilic and paucigraneulocytic patterns increased ($X^2 = 34, P = 0.001$). In contrast, the eosinophilic pattern including mixed pattern developed in the placebo group ($X^2 = 29, P = 0.003$).

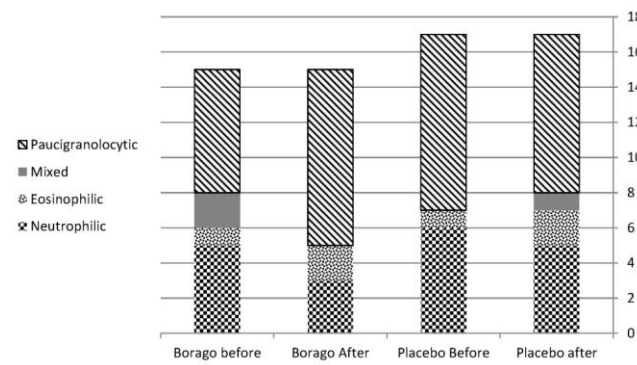


Figure 2. The cumulative classification of inflammatory cells into four major inflammatory groups

d. Side effects

Allergic skin rash was observed in two subjects consuming Borago extract, but this complication did not prompt the halt of treatment. Acute attack of asthma in one subject in Borago group was severe and this forced the halt of Borago treatment.

DISCUSSION

a. Main findings:

In this study, hydroalcoholic extract of Borago officinalis was obtained and its effect on clinical findings, spirometry, FENO, and sputum inflammatory cells of 52 subjects suffering from moderate persistent asthma was evaluated in a phase 2 prospective double-blind placebo controlled clinical trial.

Results of the study showed that 5 mL of Borago extract three times daily for one month was able to suppress major clinical findings of asthma including cough, dyspnea, airway hyper-responsiveness, and night symptoms, and it also improved the control of this disease based on the ACT score. Physical exam revealed significant improvement of wheezing. Gastro-esophageal reflux as an accompanying symptom also improved by Borago extract. Borago extract as a controller drug was able to reduce the frequency of acute asthmatic attack requiring emergency room visits and use of drugs. However, this drug did not suppress inflammatory markers including FENO, sputum neutrophils or eosinophils. Physiological parameters

including spirometry did not significantly change after the treatment.

b. Interpretation of findings in relation to previously published studies

Previous research about the beneficial effect of Borago extract showed interesting effects on TNF, which is important in asthma (5,6). Anti-TNF drugs are used in asthma (15) with substantial side effects including susceptibility to infection (16). Borago extract was able to inhibit TNF, but interestingly in this study and in another published study, it did not cause serious side effects (4). The mechanism of action of Borago extract in asthma should be elucidated more, but this study showed that the effect of Borago extract on clinical asthma is transient and rapid and it can be classified only as a reliever drug.

c. Strengths and limitations of this study:

This study was the first clinical trial about the efficacy of Borago extract for asthma. The subjects were randomly divided into two similar groups and treated similarly. The patients, clinicians, and researchers were blinded to the treatment. The drug (Borago extract) was produced by a standard method by the company, and the placebo was produced with a similar shape and taste. No serious side effect or flare up occurred in the case and control groups; therefore, it did not impact on the results of this study. Analysis of study was performed by standard parameters commonly used in clinical trials. In this study, inflammatory cells and FENO as markers of inflammation in asthma did not significantly change. Therefore, Borago extract did not show a significant effect on pathophysiology of asthma, but it showed optimal efficacy in improving the clinical findings of asthma. The placebo effect was not seen because asthma worsened in the placebo group. Future studies should be directed to elucidate the mechanism of action of Borago extract in asthma and mediators involved in asthma that were not evaluated in this study. Another interesting topic for future studies is evaluation of the possible effect of gamma-linolenic acid, which is found in high amounts in Borago

extract, on subjects suffering from deficiency of this essential compound.

CONCLUSION

In conclusion, it was shown that temporary relief of asthma symptoms was achieved in symptomatic asthmatics who used Borago extract, and so it can be prescribed for asthma as an adjunct for patients who desire herbal drug treatment alternatives. Future studies about the mechanisms of action of Borago extract involved in asthma relief are recommended especially regarding the mechanism related to gamma-linolenic acid.

Declaration of interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of this paper.

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