

Effect of *Nigella sativa* supplementation on lung function and inflammatory mediators in partly controlled asthma: a randomized controlled trial

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BACKGROUND: *Nigella sativa* and its derivatives have been reported to have anti-inflammatory and bronchodilator effects, but the effects have been evaluated in only a few clinical studies.

OBJECTIVES: To determine the effect of *N sativa* supplementation on inflammation of the airways and limitation of airflow in partly controlled asthma patients.

DESIGN: Single-blind, placebo-controlled, randomized study.

SETTING: Asthma and allergy clinic of a university hospital in eastern Saudi Arabia.

PATIENTS AND METHODS: Patients were divided into three groups. A control group (n=24) received the placebo, while NS-1 and NS-2 groups (n=26 each) received 1 and 2 g/day of *N sativa*, respectively, for 3 months along with maintenance inhaled therapy.

MAIN OUTCOME MEASURE(S): Asthma control test (ACT) score, fractional exhaled nitric oxide (FeNO), peak expiratory flow (PEF) variability and other pulmonary function tests, IgE, serum cytokines, and frequency of exacerbations.

RESULTS: FEF25-75% and FEV1 (% predicted) increased significantly ($P<.05$) at both 6 and 12 weeks in the NS-2 group. PEF variability significantly improved in both NS-1 and NS-2 groups at 6 and 12 weeks as compared with the controls ($P<.05$). FeNO and serum IgE decreased significantly after 12 weeks in both the NS-1 and NS-2 groups vs baseline ($P<.05$). Both doses of *N sativa* produced a significant increase in the serum IFN- γ at 12 weeks vs baseline ($P<.05$) as well as a significant improvement in the ACT score at 6 and 12 weeks vs baseline ($P<.001$, $<.01$). Significantly fewer patients had exacerbations in the NS-1 group ($P<.05$).

CONCLUSION: *N sativa* supplementation with inhaled maintenance therapy improves some measures of pulmonary function and inflammation in partly controlled asthma.

LIMITATIONS: No bronchoalveolar lavage or sputum samples taken for measurement of asthma markers.

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Asthma is a serious public health problem throughout the world, with a global prevalence ranging from 1% to 18%.^{1,2} All manifestations of asthma are now known to result from an underlying chronic inflammatory process mediated and orchestrated by products of certain immune cells.³⁻⁵ So far

there are no effective preventive measures or cure for asthma, and the goal of current treatment is to achieve and maintain clinical control of the manifestations.¹ Furthermore, current treatment does not modify the underlying loss of lung function and does not stop the progression of chronic inflammation and remodeling

associated with asthma.^{6,7} The need for new effective, safe and tolerable treatment modalities for asthma is apparent.

Many herbs have been evaluated for their beneficial effects on asthma in human or animal models.⁸ Among them is *Nigella sativa*, which grows in the Middle East, Eastern Europe, Western and Middle Asia. *N sativa* has been used as a natural remedy for many ailments for over 2000 years.^{9,10} Several potential anti-asthmatic effects have been reported in studies conducted in humans as well as animals. They include relaxant effects on different smooth muscle preparations,¹¹ anti-cholinergic effects,¹² an inhibitory effect on histamine (H1) receptors,¹³ and a stimulatory effect on β -adrenergic receptors.¹⁴ Anti-inflammatory, immunomodulatory and anti-oxidant effects have also been documented for this herb.¹⁵

Only a few clinical studies have evaluated the effect of *N sativa* on asthmatic patients.¹⁶⁻¹⁸ A significant improvement in subjective feelings and pulmonary function tests were observed in those studies, but limitations of the studies were the small number of patients and lack of clear outcome measures on the effect on airway inflammation. The aim of this study was to evaluate the effects of *N sativa* supplementation on clinical outcome measures and indicators of airway inflammation in patients with partly controlled asthma.

PATIENTS AND METHODS

This participant-blinded, randomized and placebo-controlled clinical trial was carried out at the University of Dammam in the Eastern Province of Saudi Arabia and its affiliated teaching hospital in accordance with the principle of the Declaration of Helsinki.¹⁹ The trial was approved by KACST (King Abdulaziz City of Science & Technology) as well as by the Institutional Review Board of the university and it was registered in an international clinical trial registry, ISRCTN (BioMed Central (ISRCTN48853858)). Written informed consent was obtained from all patients who volunteered for the trial. Previously diagnosed adult asthmatic patients (18-65 years old) were recruited from the pulmonary outpatient clinic of the university hospital. Asthma in all patients was diagnosed according to the criteria of the National Institute of Health (NIH).²⁰

The inclusion criteria were a diagnosis of partly controlled asthma according to Global Initiative for Asthma (GINA) guidelines,¹ non-smoking status and having prior treatment for at least 3 months with daily maintenance therapy of inhaled corticosteroids without any concomitant asthma medications except for short-acting β agonist (SABA) as needed. Exclusion criteria

included use of additional asthma medications (e.g. leukotriene modifiers, oral steroids), severe exacerbation or hospitalization for asthma within one month prior to or during the study period, chronic diseases (e.g. diabetes mellitus, autoimmune diseases), compliance <90% of the assigned medication, pregnancy or lactation.

Partly controlled asthmatic patients who agreed to participate in the study were selected by non-probability (convenience) sampling. Patients who met inclusion criteria were allocated to three groups by random allocation. Based upon 5 years of records from the asthma clinic, it was anticipated that about 150 subjects with partly controlled asthma would be reporting in at least four asthma seasons (about 2 years). After observing the selection criteria and accommodating dropouts, this number was determined to be sufficient to derive a reasonable sample size with a close to normal distribution. The first patient who met selection criteria was coded as subject 1 and subsequent patients were numbered sequentially, up to subject 150. The RANDBETWEEN (1,150) command of Microsoft Excel 2010 (Microsoft Company, Redmond, WA, USA) was used to create a list of random numbers for the three study groups (control, NS-1 and NS-2). The lists were sorted in numerical order. In addition, keeping in mind time and cost constraints, recruitment was limited to a maximum 2 years (approximately four asthma seasons).

The initial screening visit to the clinic was followed by a one-week run-in period to determine eligibility for recruitment, as well as to establish the baseline values for the different study measurements. At the end of the run-in-period, patients attended an entry visit (visit-0) when they were assigned to the respective groups following the distribution order of random numbers generated (**Figure 1**). The control group received 1 capsule of placebo twice daily; the NS-1 group received 1 gram *N sativa* daily in two divided doses (500 mg cap twice daily), and the NS-2 group received 2 grams *N sativa* daily in two divided doses (2 capsules of 500 mg twice daily). Subsequently they were followed with two visits scheduled 6 weeks (visit 1) and 12 weeks (visit 2) after starting the treatment. Clinical assessment, spirometry, measurement of fractional exhaled nitric oxide (FeNO) and serum 'total immunoglobulin E' (IgE) were performed at all visits. Blood samples were taken to measure serum levels of cytokines during visit-0 and visit-2 only.

All the medications were taken orally for 12 weeks in addition to the maintenance inhaler therapy (mostly budesonide 400 μ g daily and in some cases fluticasone propionate 500 μ g daily into two divided doses).

Nigella sativa (black seeds) in the form of 500 mg capsules of whole ground seeds (Bio Extracts (Pvt) Ltd, Sri Lanka), were used in this study. Charcoal powder in the form of capsules (260 mg) supplied by Arkopharma Pharmaceutical Laboratories Carros, France, was used as placebo. The participants were blinded; none knew whether he or she was receiving placebo or *N sativa*.

The degree of control of asthma symptoms was assessed using Asthma Control Test (ACT)²¹ which has been found to correlate well with GINA guidelines for control of asthma.²²⁻²⁴ Information on daytime symptoms, limitation of activities, nocturnal symptoms, need for reliever/rescue treatment, frequency of exacerbations and lung functions was recorded in a proforma at baseline. At each visit, the occurrence of moderate or severe exacerbations, defined according to the American Thoracic Society (ATS) and European Respiratory Society (ERS) criteria,²⁵ were recorded. The percentage of patients with at least one exacerbation, over the whole treatment period, was calculated in each group.

Spirometry was carried out following ATS guidelines for standardization of spirometry,^{26,27} using a calibrated computerized pneumatograph spirometer (Vitalograph) that included software for a BTPS (body temperature and pressure saturated) correction factor. The predicted values of forced expiratory volume at one second (FEV1 % predicted), forced vital capacity (FVC), and forced expiratory flow (FEF25-75%) were calculated automatically based on age, gender, height and ethnicity. The procedure was fully explained to the patients before the test, and subjects were encouraged to perform at least three valid maneuvers during each measurement, with the best values used for analysis.

The participants were trained to measure their peak expiratory flow (PEF) at home using a handheld portable peak expiratory flow meter (Mini -Wright standard peak flow meter-Clement Clarke international limited-London, UK) provided free of cost at the start of study. PEF was measured twice daily during the week of the run-in-period preceding the start of the trial and was repeated during the week preceding every follow up visit. PEF was measured first thing in the morning and at night before medicines were taken. The participants were reminded by phone and readings were plotted in a PEF chart. An index value for PEF variability was calculated by obtaining the lowest morning reading (AM PEF) over the week, divided by the highest PEF reading in that week expressed as a percent. The higher the index, the lower the PEF variability, and control of asthma symptoms is better (and vice versa).¹

Fractional exhaled nitric oxide (FeNO) was mea-

sured according to ATS/ERS guidelines^{28,29} using a small portable hand held NO analyzer (Niox Mino- Aerocrine AB, Solna, Sweden). This device has a measurement range of 5-300 ppb, and reproducibility of <3 ppb for measured values below 30 ppb and <10% for measured values ≥ 30 ppb. In addition, FeNO was measured before spirometry to avoid the effect of the spirometric maneuver on FeNO.

Eight milliliters of blood were collected in a plain tube from every patient by venipuncture. The blood was allowed to clot for at least half an hour at room temperature and then centrifuged at 4°C at 3000 rpm for 15 minutes. Serum was collected into small aliquots and stored at -80 °C for total IgE and cytokines analysis. Total IgE was measured using a fluoroenzyme immunoassay technique using ImmunoCAP total IgE (Phadia AB, Uppsala, Sweden). The cytokines (IL-4, IL-10, IL-17, IFN- γ and eotaxin) were measured in serum using specific quantitative sandwich ELISA technique using kits supplied by IBL company (Germany). Samples were analyzed in single measurements since the IBL cytokines assay has good precision in pretests with a stated intra-assay mean coefficient of variance (CV) of less than 10%.

Statistical analysis

Data were reported as arithmetic means and standard deviation for all variables except for cytokines where the data were transformed to logarithmic scale to assume normal distribution and reported as geometric mean. Analysis of variance (ANOVA) was used to compare the demographic and biometric characteristics of the patients. Comparison of proportions between groups was done using the chi-square (χ^2) test. Analysis of covariance (ANCOVA) was used for between the groups comparison of pulmonary function tests, serum cytokines, FeNO and IgE. The covariate for ANCOVA was the respective baseline value, the test being compared as the dependent variable and the groups as fixed variable. Wilcoxon rank test and Kruskal Wallis tests were used for analysis of ACT score within and between groups, respectively. For within group comparisons the paired t test was used. Data were analyzed using Statistical Package of Social Science (SPSS) software version 16. *P* values of <.05 were considered significant. The intention to treat methodology was used for the number of participants finally analyzed with missing values being ascribed by last observation carried forward.

RESULTS

One hundred and forty-one partly controlled asthmatic patients participated in the study (**Figure 1**). Sixty-

five participants were excluded. Before the start of the study, 7 of those patients withdrew when briefed about methodology for informed consent. Seventy-six patients were subsequently allocated to three groups by random allocation (Table 1). Of the 76 patients enrolled in the study, two patients from the control group discontinued because of severe exacerbations, one patient from group NS-1 discontinued because of an urgent (unrelated) surgery and one patient from group NS-2 was lost to follow. Table 2 summarizes the results of lung function tests for all patients in the three groups, at baseline, after 6 weeks and after 12 weeks of treatment. There were no significant differences between the three groups at baseline in any of the pulmonary function tests. Treatment with placebo for 6 or 12 weeks was not associated with statistically significant changes from baseline in any of the lung function tests. In the group treated with the high-dose *N sativa* (NS-2) at 6 and 12 weeks, FEV1 (% predicted), FEF25-75 and FEF75-75 (% predicted) were significantly higher than baseline values. In addition, the ratio of FEV1/FVC showed a significant increment at the 6 week measurement in NS-2 group, compared with the corresponding baseline values. PEF variability was significantly higher at 6 weeks and 12 weeks of treatment with the low dose *N sativa* (NS-1) and at 12 weeks treatment with the high dose *N sativa* (NS-2) compared to their corresponding baseline values. Furthermore, the improvement in PEF variability from baseline by both *N sativa* doses after 12 weeks of treatment was significantly higher than the corresponding placebo change.

In levels of cytokines, no significant differences between the three groups were observed at the baseline (Table 3). The levels of IL-4, IL-10, IL-17A and the eotaxins did not change significantly in any of the three groups at any of the follow up visits. However, for both *N sativa* groups (NS-1 and NS-2), the serum level of IFN- γ at 12 weeks was significantly higher than at baseline.

In fractional exhaled nitric oxide (FeNO) and serum levels of immunoglobulin E (IgE), no significant differences between the three groups at baseline were found for both FeNO and IgE (Table 4). The low dose of NS (NS-1) produced a significant reduction in FeNO at weeks 6 and 12, while the higher dose group (NS-2) showed significant reductions in serum IgE compared with baseline.

In ACT scores, no significant differences were found between the three groups at baseline and the scores for the three groups were below the cut-off value of 20, which separates controlled from uncontrolled asthmatic patients (Figure 2). ACT scores increased significantly from the respective baseline values after 6 and 12

weeks of treatment in placebo and both test groups. The increment in ACT score in the placebo group, however, remained below the cut-off value of 20, while both *N sativa* groups exceeded this barrier. Also, ACT scores at 6 weeks and 12 weeks of treatment with any of the two doses of *N sativa* were significantly higher than the corresponding values in the group treated with placebo (control group). Nine patients (37.5%) in the control group, 3 (11.5%) in the NS-1 group and 4 (15.4%) in the NS-2 group developed severe exacerbations dur-

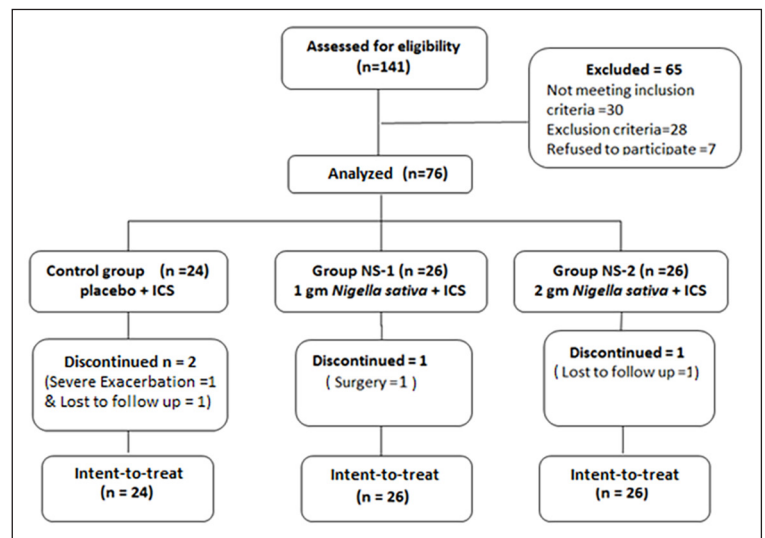


Figure 1. Flow diagram of recruitment and dispensation of subjects. ICS=Inhaled corticosteroid

Table 1. Demographic and biometric characteristics of the patients.

Variables*	Control Group (n=24)	Group NS-1 (n=26)	Group NS-2 (n=26)
Age (years)	37.1 (11.2)	37.5 (12.7)	39.2 (13.6)
Male, n (%)	9 (37.5)	8 (30.8)	9 (34.6)
Female, n (%)	15 (62.5)	18 (69.2)	17 (65.4)
Height (m)	1.58 (0.09)	1.60 (0.1)	1.60 (0.07)
Weight (kg)	75.49 (15.5)	77.12 (19.5)	78.28 (15.2)
BMI (kg/m ²)	30.1 (6.0)	30.6 (8.8)	30.4 (5.2)
Asthma duration (years)	13.3 (8.5)	10.3 (8.9)	13.1 (6.4)
Atopic asthma n (%)	14 (58.3)	12 (46.2)	12 (46.2)

Data is presented as (mean±SD) for all variables. No statistically significant differences between treatment groups. Control: Placebo group, NS-1: 1 g/day *Nigella sativa*, NS-2: 2 g/day *Nigella sativa*

Table 2. Pulmonary function tests in patients treated with placebo, low-dose *Nigella sativa* (NS-1) and high-dose *Nigella sativa* (NS-2) at baseline, and after 6 and 12 weeks of treatment.

Variable	Group	Baseline	6 weeks	12 weeks
FEV1 (L)	Control group	2.3 (0.6)	2.3 (0.7)	2.3 (0.6)
	Group NS-1	2.5 (0.8)	2.6 (0.8)	2.6 (0.8)
	Group NS-2	2.3 (0.7)	2.4 (0.8)	2.4 (0.7)
FEV1 (% predicted)	Control group	81.1 (19.1)	79.8 (20.6)	80.8 (20.6)
	Group NS-1	85.5 (17.3)	86.8 (15.3)	87.7 (15.8)
	Group NS-2	78.1 (21.4)	83.8 (21.4)*	85.5 (22.9)*
FVC (L)	Control group	3.0 (0.7)	3.0 (0.7)	3.0 (0.7)
	Group NS-1	3.2 (1.0)	3.2 (1.0)	3.3 (1.0)
	Group NS-2	3.0 (0.9)	3.1 (0.9)	3.2 (0.9)
FVC (% predicted)	Control group	90.1 (13.7)	88.8 (17.1)	89.1 (13.7)
	Group NS-1	92.8 (17.3)	93.3 (16.8)	94.8 (14.8)
	Group NS-2	88.7 (21.9)	91.4 (14.8)	93.0 (22.9)
FEV1/FVC (%)	Control group	76.2 (13.2)	76.2 (11.8)	77.2 (12.2)
	Group NS-1	78.6 (9.1)	79.1 (9.2)	79.0 (9.1)
	Group NS-2	74.3 (9.7)	77.4 (9.7)*	76.7 (7.6)
FEF 25-75 (L/sec)	Control group	2.3 (1.3)	2.2 (1.2)	2.3 (1.5)
	Group NS-1	2.5 (1.1)	2.6 (1.2)	2.7 (1.3)
	Group NS-2	1.9 (0.9)	2.3 (1.2)**	2.3 (1.0)**
FEF 25-75 (% predicted)	Control group	67.5 (38.7)	66.2 (35.3)	68.2 (40.7)
	Group NS-1	74.5 (29.1)	76.5 (30.1)	77.3 (31.6)
	Group NS-2	56.3 (25.0)	66.5 (31.6)**	66.4 (27.5)
PEF variability	Control group	76.6 (7.3)	78.3 (8.3)	78.5 (8.8)
	Group NS-1	73.5 (10.7)	80.9 (9.7)**	83.6 (8.7)***
	Group NS-2	73.7 (11.2)	75.3 (11.7)	81.4 (8.7)**

Data are presented as mean and standard deviation. Control: Placebo group (n=24), NS-1: 1g/day *Nigella sativa* (n=26), NS-2: 2 g/day *Nigella sativa* (n=26), FEV1: Forced expiratory volume at one second, FVC: Forced vital capacity, FEF: Forced expiratory flow, PEF: Peak expiratory flow.

* $P < .05$, ** $P < .01$, *** $P < .01$ for between groups comparison PEF variability at 12 weeks. For ANCOVA Tests of between-subjects effects: $F = 5.016$, $P = .009$, adjusted R-squared = .300. The Levene test indicated equality of variances for the PEF variability tests ($P = .563$). For within group analysis using paired t test FEV1 for group NS-2 at 6 weeks as well as 12 weeks and PEF variability for Group NS-1 at 6 weeks shows significant difference when compared with the baseline.

ing the follow-up period. The frequency of exacerbations in the NS-1 group was significantly less than the placebo group.

DISCUSSION

N sativa has been studied for the control of bronchial asthma using in vitro methods,³⁰ animal models,³¹ and

clinical trials.¹⁷ These studies have reported several positive effects on different aspects of asthma control like clinical outcome, pulmonary function tests, cytokines, IgE and FeNO. However, to the best of our knowledge, the current study is the first in which all of these aspects have been studied simultaneously. Evaluation of these aspects together could increase the reliability of conclusions and elucidate a more mechanistic basis for therapeutic effect. A special feature of our study was an active effort to reinforce compliance through frequent contact with participants in between the visits. Thus we achieved a good compliance percentage among our patients (>90%). In general, *N sativa* was well tolerated and no side effects were reported by any of our participants.

Our results support two previous studies, which reported a favorable effect of *N sativa* on lung function tests in asthmatic patients.^{16,17} The improvement in lung function tests points to a possible bronchodilator effect of *N sativa*. However, our study also showed improvement in PEF variability with administration of *N sativa*, which better reflects bronchial hyper-responsiveness than other lung function tests. This finding indicates that *N sativa* supplementation specifically improves expiratory flow during the mid-part of vital capacity (FEF25-75%). FEF25-75% is known to reflect the function of the small airways³² and has recently been implicated in the pathogenesis of asthma.³³

Our results suggest that *N sativa* might minimize the underlying inflammation. This might prevent progression of remodelling of bronchial tissue. This potential effect on remodeling is further supported by the finding that most of the effects of *N sativa* in this study are more remarkable after 12 weeks of continuous use of this herb. Remodelling is a long-term process and thus *N sativa* might be ameliorating the inflammatory reaction. We could not ascertain this as our study was not designed to assess actual remodelling due to the invasive procedures involved.

Our study is the first to show that administration of *N sativa* to patients with asthma is associated with a reduction in FeNO, an indicator of the inflammatory reaction underlying pathogenesis of bronchial asthma.²⁹ However, it is not clear why the reduction in FeNO at the 1 g dose disappeared at the 2 g dose of NS. This observation seems to be in line with a recent in vitro study on the effect of an alcoholic extract *N sativa* on NO production in rat mix glial cells inflamed by lipopolysaccharide, which were treated with different doses of black seeds extract. The authors observed a significant reduction in NO production at lower doses of the extract, which was lost at higher doses.³⁴ In addition,

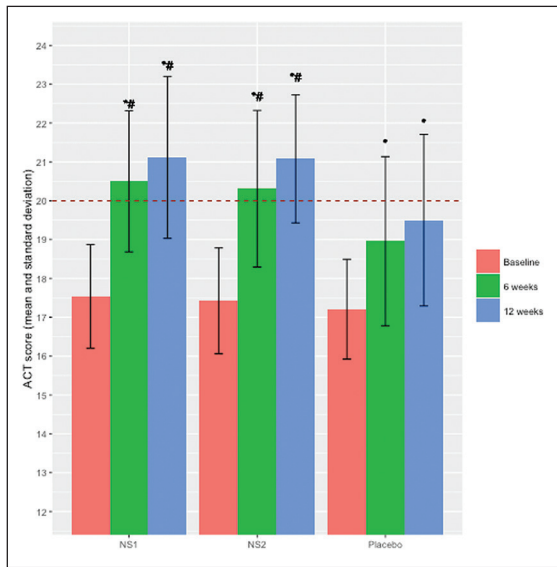


Figure 2. ACT score (mean and standard deviation) within each treatment group at baseline and at 6 and 12 weeks (the horizontal line at the ACT score of 20 indicates the cut-off point score). *Significant difference ($P < .001$) compared to corresponding baseline value within each group. #Significant difference ($P < .01$) between groups in the mean ACT scores at both 6 and 12 weeks visits compared to the corresponding measurements in the placebo group. ACT=asthma control test, NS=*Nigella sativa*

earlier studies on the effect of *N sativa* on other health disorders reported the same observation. A higher dose of *N sativa* cells (3 g/day) was less effective than a lower dose (2 g/day) in eradicating *H pylori* in patients with non-ulcer dyspepsia³⁵ and in controlling blood glucose in diabetic patients.³⁶ A possible explanation lies in the fact that the whole black seed contains more than 100 constituents, many of which have not been chemically identified nor have they been pharmacologically tested. Some of these ingredients of *N sativa* might have been maximally active against NO production at a 1 g/day dosage; the effect was minimized at higher doses.

Most aspects of the pathogenesis of bronchial asthma have been linked to an underlying chronic inflammatory process that is dependent on a balance between two groups of mediators, pro-inflammatory and anti-inflammatory. In our study we included IL-4 (produced by Th2) and IFN- γ (produced by Th1) as representative of these two groups. It appears that this balance can be tipped towards increased or decreased inflammation by certain cytokines. It has been suggested that IL-4 tips this balance towards increased inflammation while IFN- γ has the opposite effect.³⁷ Interestingly, our study shows that IFN- γ is increased by both doses of *N sativa*, which might partially account for its anti-

Table 3. Serum cytokine levels in the control (placebo) and *Nigella sativa*-treated groups at the baseline, and after 6 and 12 weeks of treatment.

Cytokines	Group	Baseline	12 weeks	P
IL-4 (pg/mL)	Control group	1.6 (5.7)	1.6 (5.7)	.50
	Group NS-1	2.4 (6.7)	2.3 (6.8)	.90
	Group NS-2	2.2 (6.5)	2.1 (6.4)	.96
IL-10 (pg/mL)	Control group	2.2 (5.8)	1.6 (6.3)	.07
	Group NS-1	2.4 (6.1)	2.8 (6.3)	.20
	Group NS-2	1.7 (6.6)	1.5 (6.6)	.40
IL-17A (pg/mL)	Control group	1.2 (6.4)	1.0 (6.6)	.30
	Group NS-1	1.8 (7.2)	2.9 (7.3)	.13
	Group NS-2	1.9 (7.0)	1.6 (7.4)	.34
IFN- γ (pg/mL)	Control group	3.0 (5.5)	2.6 (5.4)	.26
	Group NS-1	3.8 (5.8)	4.7 (6.0)	.03*
	Group NS-2	2.8 (5.8)	3.3 (6.0)	.03*
Eotaxin (pg/mL)	Control group	98.1 (238.6)	100.6 (256.2)	.26
	Group NS-1	99.4 (104.5)	100.9 (40.8)	.91
	Group NS-2	96.2 (42.3)	100.7 (30.1)	.42

All variables are represented as geometric mean (SD), except eotaxin which is represented as arithmetic mean (SD). *Significant difference ($P < .05$). IL=interleukin; IFN- γ =interferon gamma. NS: *Nigella sativa*, Control placebo group (n=24), NS-1: 1 g/day *Nigella sativa* (n=26), NS-2: 2 g/day *Nigella sativa* (n=26) * $P < .05$

For between groups comparison IFN- γ (interferon gamma) at 12 weeks, for ANCOVA of between subjects effects: $F=4.287$, $P=.017$, adjusted R-squared=0.560. The Levene test indicated equality of variances for IFN- γ tests ($P=.321$).

Table 4. FeNO and serum Ig E in the control (placebo) and in the *Nigella sativa*-treated groups at the baseline, and after 6 and 12 weeks of treatment.

Variable	Group	Baseline	6 weeks	12 weeks
FeNO (ppb)	Control group	34.9(32.8)	38.2 (37.7)	34.8 (26.9)
	Group NS-1	23.0 (13.3)	20.8 (11.2)	18.1 (8.2)*
	Group NS-2	27.6 (30.6)	24.4 (21.4)	26.9 (29.1)
IgE (kU/L)	Control group	623.8 (802.5)	579.6 (672.6)	595.9 (752.5)
	Group NS-1	453.5 (711.8)	401.3 (621.6)	391.5 (616.0)
	Group NS-2	393.8 (470.1)	345.3 (392.6)	322.0 (373.2)**

Data is presented as (mean \pm SD), NS=*Nigella sativa* * $P < .05$, ** $P < .01$; FeNO: fractional exhaled nitric oxide, Ig E: immunoglobulin E; Control placebo group (n=24), NS-1: 1 g/day *Nigella sativa* (n=26), NS-2: 2 g/day *Nigella sativa* (n=26)

inflammatory effect. Other studies have shown that *N sativa*³⁸ as well as thymoquinone (an active ingredient of NS),³⁹ suppress the release of IL-4 and augment production of IFN- γ . This opens room for further study on a possible mechanism for the anti-asthmatic potential

of *N sativa*—restoration of the balance between Th1/Th2 cytokines.

The reduction in serum IgE produced by the 2-g dose of NS supports a previous study in the mouse model of asthma.⁴⁰ This reduction in serum Ig E level might be linked to the high levels of IFN- γ associated with both doses of *N sativa* used in our study. Noteworthy is the fact that IFN- γ is known to suppress the inflammatory reaction in asthma.⁴¹

The scores on the ACT improved in all groups, including those who received placebo. This placebo effect has been reported⁴² and is expected in a subjective test like ACT. The ACT score has been validated by multiple studies. It includes items on shortness of breath, night time waking, interference with activity, rescue treatment use and rating of asthma control by the patient. It correlates with the degree of airways inflammation reflected by FeNO⁴³ and with the frequency of exacerbation and the degree of control of asthma.⁴⁴

None of our subjects reported any unpleasant or untoward effects. We did not statistically evaluate other reported effects. Some of our patients who had co-existing allergic rhinitis reported a marked improvement in the allergic symptoms such as itching, running nose, sneezing and nasal blockage. Others reported an improvement in oral thrush (a common side effect of inhaled corticosteroid therapy (ICS)). These observations

point to the need for further studies to evaluate the use of *N sativa* in allergic rhinitis and as complementary therapy to ICS in asthma.

One limitation of this study was the difficulty in obtaining bronchoalveolar lavage or induced sputum samples for inflammatory cells and marker analysis. Both of these procedures are uncomfortable and have a significant risk of bronchospasm. Most patients refuse these once explained. Furthermore, the processing techniques for these samples are complicated with a limited success rate even in highly qualified centers. We compensated for this limitation by substituting these procedures with the non-invasive and novel measurement of fractional exhaled nitric oxide, which has a comparable reliability and specificity to invasive techniques.^{45,46}

We conclude that addition of the *N sativa* to the controller ICS therapy improves the overall level of control and decreases exacerbations in partly controlled asthma.

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Conflict of interest

The authors report no conflict of interest.

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