

REVIEW ARTICLE

Role of *Nigella sativa* Extract in Reducing the Inflammatory Damage in Oral Submucous Fibrosis – A Review

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ABSTRACT

Nigella sativa (*N. sativa*) is an annual flowering plant, which belongs to the family Ranunculaceae. Both the seeds and oil of *N. sativa* have been researched extensively in the past few years due to the variety of its chemical ingredients and biological properties. *N. sativa* seeds contain many components including Thymoquinone (TQ), di-thymoquinone, fixed oils, alkaloids, flavonoids, proteins, fatty acids, saponins, and alpha-hederin. TQ is the most pharmacologically active component among all the constituents. The important properties exhibited by TQ are anti-inflammatory, anti-cancer, analgesic, anti-oxidant, anti-hypertensive and anti-diabetic activities. Oral submucous fibrosis (OSMF) is a chronic inflammatory disorder of the oral cavity, which has a multifactorial etiology. Areca nut consumption in the form of quid is considered as the most important factor in the development of the disease. It has a malignant transformation rate of 7-30%. A large number of researches have been conducted to investigate the outcome of using *N. sativa* and its active constituent TQ in various inflammatory conditions. This article is aimed at reviewing the literature available on various databases (Google Scholar, PubMed, and Online Journals) and to conclude if *N. sativa* can also be utilized as a potent anti-inflammatory agent for reducing the signs and symptoms of OSMF.

Keywords: *Nigella sativa*; Oral Submucous Fibrosis; Inflammation.

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INTRODUCTION

Nigella sativa (*N. sativa*) is an herb, which belongs to family Ranunculaceae¹. It is mainly cultivated in countries such as Bangladesh, Pakistan, India, Iran, Turkey, and Saudi Arabia and in several areas of Middle East and South Europe²⁻⁴. Seeds of *N. sativa* have been commonly used as a flavoring and seasoning agent for preparing different cuisines in many Asian and Eastern countries over centuries^{1,5}. Oil of *N. sativa* has also been used for treating various ailments such as diabetes, infertility, obesity, rheumatoid arthritis, hypertension and many lung diseases⁶. Its ointment has also been used to treat nasal ulcers, eczema, abscesses, orchitis and swollen joints⁶.

Oral submucous fibrosis (OSMF) is a disorder of the oral cavity which is chronic in nature⁷ and is mainly characterized by formation of fibrous bands in the subepithelial layers along with a juxtaepithelial inflammatory reaction⁸. It may affect any part of the oral cavity and may also extend into the pharynx^{9,10}. OSMF is more prevalent in India, Southeast Asia and in Asian immigrants living in United States, United Kingdom and other developed countries¹¹⁻¹³. Areca nut chewing in the form of quid is considered as the main etiological factor in the pathogenesis of OSMF¹⁴.

DISCUSSION

N. sativa seeds contain many components like Thymoquinone (TQ), di-thymoquinone, fixed oils, alkaloids, flavonoids, proteins, fatty acids, saponins, and alpha-hederin¹⁵. TQ is the most pharmacologically active component (Figure 1)^{1,16,17}. The important properties exhibited by TQ include its anti-inflammatory, analgesic,

immunomodulatory, anti-oxidant, anti-cancer, anti-diabetic and anti-hypertensive activities^{3,18-20}. Both the seeds and oil of *N. sativa* have been researched extensively in the past few years due to the variety of its chemical ingredients and biological properties, which help in improving human health, nutrition and several medical conditions²¹.

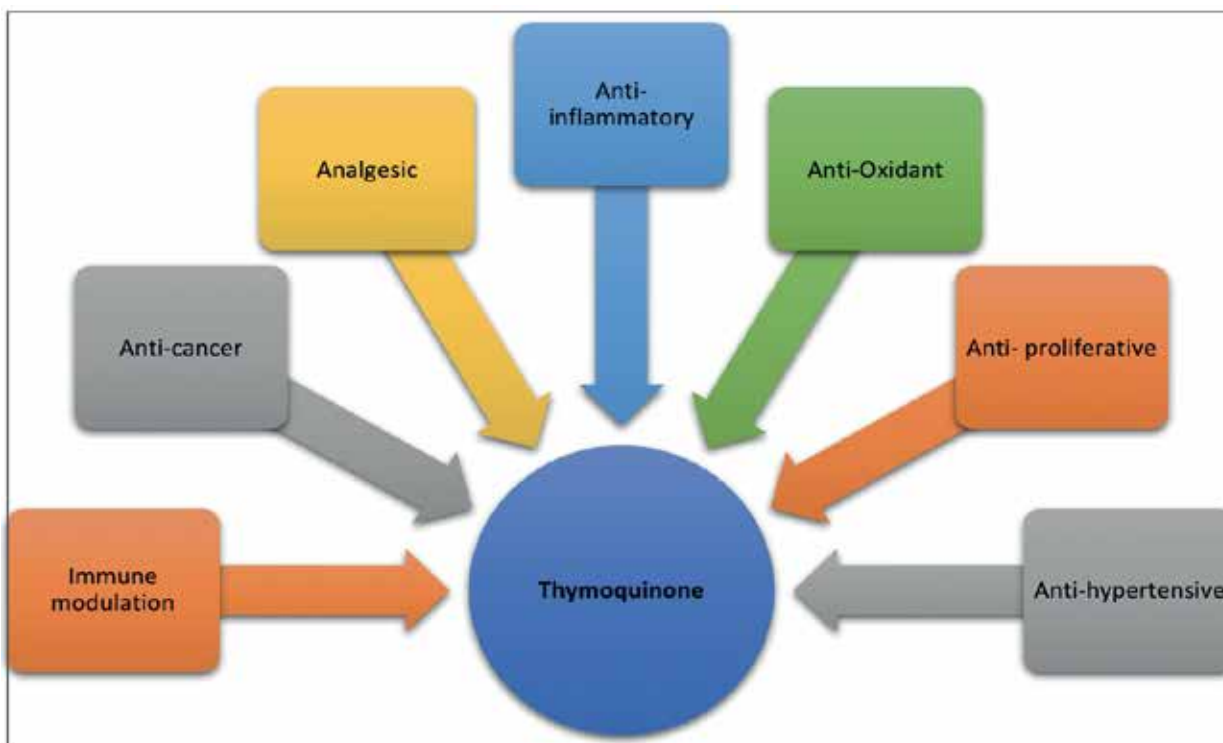


Figure 1: Various properties exhibited by Thymoquinone (TQ)

Inflammation is a complex biological response of body tissues, which occur because of injury, several pathogens, toxic compounds, damaged cells and irradiation²². Inflammation plays an important part in the development of various diseases such as cancer, arthritis, asthma, osteoarthritis and cystic fibrosis⁵. The process of inflammation involves release of certain inflammatory cytokines and mediators. A variety of stimuli such as bacteria, viruses, stress and cytokines induce a transcription factor known as NF- κ B, which in turn is responsible for regulating immune and inflammatory response²³.

NF- κ B mediates the induction of various pro-inflammatory genes and it causes activation, differentiation and effector function of inflammatory T-cells²⁴. Interleukins (IL), transforming growth factor- β (TGF- β), natural killer cells (NK), tumor necrosis factor- α (TNF- α) and major histocompatibility complex (MHC) are a few major cytokines among which IL and TNF- α plays an important role in most of the inflammatory conditions²⁵. Many trials have been carried out using *N. sativa* to demonstrate its strong anti-inflammatory activity²⁶ as shown in Table 1.

Table 1: Trials conducted on different diseases using *Nigella sativa* oil.

S. No.	Authors' Name	Study Design	Intervention
1	Hussain et al. (2019) ²⁷	Randomized controlled trial	Intervention group: <i>N. sativa</i> oil Placebo group: Magic mouthwash
2	Ameen et al. (2019) ^{28,29}	Randomized controlled trial	Intervention group: <i>N. sativa</i> oil Placebo group: Magic mouthwash
3	Hadi et al. (2016) ²⁶	Randomized control trial	Placebo group: Starch filled capsules NS group: <i>N. sativa</i> oil capsules

***Nigella sativa* in Preventing Oral Mucositis in Acute Myeloid Leukemia**

A trial was conducted in 2019 in which *N. sativa* oil was used as a mouth rinse to improve oral mucositis in chemotherapy treated acute myeloid leukemia (AML) patients. Total 54 AML patients were enrolled for the trial and were divided into Magic mouthwash group and *N. sativa* mouth rinse group. Participants in both the groups were asked to do the Magic mouthwash and *Nigella* oil mouthwash rinses (10ml) topically four times a day for four weeks. The severity of Oral mucositis was assessed using Oral mucositis assessment scale (OMAS) While Visual Analogue scale (VAS) was used to measure the pain intensity and swallowing and levels of IL-6 and TNF- α were assessed before starting the trial. At the end of the trial, it was concluded that *N. sativa* mouth rinse was more effective as compared to Magic mouthwash in reducing the inflammation of oral mucosa. It also reduced the pain and improved swallowing in participants belonging to *N. sativa* group. IL-6 and TNF- α levels in saliva were also found to be reduced in the *Nigella* oil mouthwash group as compared to the other group²⁷.

***Nigella sativa* in Preventing Oral Mucositis in Head and Neck Squamous Cell Carcinoma (HNSCC)**

An open label clinical trial was conducted in 2017 on patients who were diagnosed with squamous cell carcinoma of the head and neck to check the effect of *Nigella sativa* oil mouthwash (NSO) on reducing oral mucositis caused by chemo-radiation. Levels of salivary interleukin 6 (IL-6) and TNF- α were also assessed. Forty patients were recruited for this trial and were divided through randomization into Group A and Group B respectively. Patients in Group A were asked to use NS oil mouthwash 5 times a day whereas participants in Group B were given Magic mouthwash and were instructed to use it in a similar manner. The results of this trial showed decrease in the severity of oral mucositis, which was assessed using the radiation therapy oncology group (RTOG) guidelines in chemotherapy or chemo-radiotherapy, treated HNSCC patients after using *N. sativa* oil mouthwash²⁸. Another clinical trial was conducted in 2017 until 2018 to assess the outcome of using *N. sativa* oil on oral mucositis in HNSCC patients. Forty participants who met the inclusion criteria were enrolled for this trial.

After randomization, the patients were split up into Group A, Group B. Groups A participants were given NS oil mouthwash (10ml), and group B participants were treated with magic mouthwash (10ml). All the patients were instructed to use the mouthwash 6 times daily for a period of 7 weeks. Participants in both the groups were treated with radiation or chemo-radiation according to the hospital protocol and were assessed weekly for the development of oral mucositis using RTOG guidelines. The results of this trial showed no significant difference in RTOG grading between the

two groups after 3 weeks of intervention. From week 4 until week 6, a very few number of patients in Group A indicated the development of mucositis as compared to Group B. In addition, the severity of OM was reduced in those patients suggesting the ability of NS oil to act as a potent anti-inflammatory agent. Patients in group A also showed betterment in swallowing function as compared to group B²⁹.

An *in-vitro* study was also conducted to see the effects of thymoquinone on head and neck squamous cell carcinoma (HNSCC) cell lines individually or in combination with cisplatin and radiation. In this study, two HNSCC cell lines (SCC25 and CAL27) were used to observe the effects of TQ on cell proliferation in combination with radiation and cisplatin. The results of this study suggested that TQ has strong anti-proliferative properties and due to its lesser toxicity, it can be used in combination with radiotherapy to treat HNSCC. However, the combined use of TQ and cisplatin showed no significant results. The combination of TQ and radiation also resulted in reduced clonogenic survival of the HNSCC cells³⁰.

***Nigella sativa* in Improving Clinical Parameters in Oral Submucous Fibrosis (OSMF)**

OSMF is a premalignant condition with great potential to convert into malignancy³¹. The malignant transformation rate of OSMF ranges from 7-30%³². A large number of treatment modalities have been suggested for treating OSMF but they all are only effective in reducing the signs and symptoms of the disease with no curative value^{10,33,34}. In OSMF, inflammation occurs as a response to tissue injury caused by areca nut chewing which in turn causes the release of certain cytokines such as IL-6, TNF- α , and TGF- β by the macrophages³⁵. Among these cytokines, TGF- β is the most important cytokine involved in the formation of fibrosis¹⁰. It increases the collagen production by activating pro-collagen genes while it decreases collagen degradation by activating the tissue inhibitor of matrix metalloproteinase gene (TIMP) and inhibiting the collagenase activity¹⁰.

A randomized preliminary study was conducted to assess the effect of using turmeric with black pepper and *N. sativa* in OSMF. This study was conducted in two phases (I and II). Interventional part was carried out in phase I in which a total number of 40 patients were recruited and were divided in to two subgroups: Group A and group B respectively. Group A was given turmeric with black pepper in capsule form (400mg) and group B was given *N. sativa* capsules (500mg) for a period of three months. Both the groups were recalled after every 15 days for follow up visit and four different clinical parameters were assessed on each visit including burning sensation, Interincisal mouth opening (IMO), tongue protrusion and cheek

flexibility. In phase 2, Levels of superoxide dismutase (SOD) were also checked before and after treatment in both the groups and their values were compared with the healthy controls³⁶. SOD is an antioxidant, which acts as a first line defense against injuries caused by the production of reactive oxygen species³⁷. The results of this trial showed significant betterment in clinical parameters of both the groups (<0.01) while it was observed that the overall treatment response was a little higher in group A. The mean value for SOD was also increased in both groups at the end of the trial³⁶.

***Nigella sativa* in Preventing Inflammation in Arthritis**

A randomized clinical trial was carried out to check the effect of *N. sativa* oil extract on oxidative stress and inflammatory cytokine response in rheumatoid arthritis patients. The study participants were divided into two groups; an intervention group and the control group. Patients in the intervention group were given *N. sativa* oil capsules and the other group was given paraffin capsules. Serum levels of TNF- α and IL-10 and oxidative stress parameters were assessed in whole blood at the baseline and end of the trial. The results of this study showed significant decrease in the disease activity score following the assessment of 28 joints (DAS28) of intervention group while it remained unchanged in the placebo group. In addition, the serum IL-10 levels were found to be high at the end of the trial in the intervention group whereas the levels of malondialdehyde and nitric oxide (NO) were significantly decreased compared to the baseline values in the intervention group. Moreover, the levels of Super oxide dismutase (SOD), TNF- α , catalase and total antioxidant capacity (TAC) within or between the groups were not found to be significant statistically²⁶.

***Nigella sativa* in Preventing Inflammation in Asthma and COPD**

Ikhsan et al. carried out a trial in 2018 to evaluate the effectiveness of ethanol extract of *N. sativa* on reducing the inflammatory process in Wistar rat mast cells. The results of this experiment indicated that the extract of *N. sativa* inhibited the release of histamine from peritoneal Wistar rat mast cells and was effective in reducing inflammation without producing any side effects³⁸. Another double blinded, randomized controlled trial was done to estimate the effect of using *N. sativa* oil (NSO) capsules in asthmatic patients and to check for reduction in their inflammatory parameters. A total of 80 adult asthmatic patients were enrolled in this study. Treatment group received NS oil capsules while the placebo group was given virgin olive oil capsules twice a day for four weeks. At the end of the trial significant improvement was seen in patients with low basal predicted FEV₁ % (forced expiratory volume) in the NSO group as compared

to the placebo group. In addition, significant decrease in the absolute peripheral blood eosinophil count was observed in the NSO group while the level of total serum IgE remained statistically insignificant between the two groups³⁹.

A trial was done on clinically diagnosed COPD patients in 2019 to assess the beneficial effects of using *N. sativa* oil in such patients. Total 100 patients with mild to moderate COPD were enrolled for the trial and were placed in two different groups after randomization; Control group and Black seed oil (BSO) group respectively. Patients in the control group were given conventional COPD treatment while participants in BSO group were advised to take *Nigella* oil capsules along with the conventional therapy. The results of this trial showed statistically significant increase in the values of pulmonary function tests in comparison to the control group. IL-6 and TNF- α levels were also found to be decreased significantly in participants of BSO group when compared to the control group after the completion of the trial⁴⁰.

***Nigella sativa* in Preventing Inflammation in Pancreatic cancer**

A trial was performed to check the anti-inflammatory activity of TQ on pancreatic cancer cells (HS766T) and to analyze the pro-inflammatory cytokine and chemokines expression using real-time PCR. The result of this study showed that TQ reduces the constitutive and TNF- α induced activation of Nuclear factor kappa beta (NF- κ B) and reduces its transport into the nucleus. In addition, there was a dose and time dependent decrease in the formation of MCP-1, interleukin-1 β , TNF- α and Cyclooxygenase-2 suggesting a strong anti-inflammatory activity exhibited by TQ⁴¹.

CONCLUSION

The most common anti-inflammatory agent used for the treatment of OSMF is steroid. However, it has been observed that the prolonged use of steroids can lead to water and salt retention, gastric ulceration and bone marrow depression when given systemically. *N. sativa* is a natural herb with many health benefits. It has a very strong anti-inflammatory activity so it can be utilized as a potent anti-inflammatory agent with almost no adverse effects for treating OSMF.

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CONFLICT OF INTEREST

Authors have no conflict of interest to declare.

AUTHOR'S CONTRIBUTION

HB conceptualized the study and wrote the manuscript, SA helped in writing the manuscript, SS helped in collecting the information via online sources, MH overall supervised and proof read the manuscript.

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