Boletim Informativo Geum
Informative Geum Bulletin

Boletim Informativo Geum, v. 8, n. 2, p.16-25, abr/jun., 2017
ISSN 2237-7387 (Impresso)

Nigella sativa, a promising source of respiratory drugs

Muhammad Torequl Islam[1,2]

[1]Department for Management of Science and Technology Development, Ton Duc Thang University, Ho Chi Minh City, Vietnam.
[2]Faculty of Pharmacy, Ton Duc Thang University, Ho Chi Minh City, Vietnam.

***E-mail:** muhammad.torequl.islam@tdt.edu.vn

ABSTRACT

The use of *Nigella sativa* and its preparations in the treatment of various diseases is ancient. A number of lung diseases such as asthma, bronchiectasis, bronchitis, and chronic obstructive pulmonary disease are one of the major causes of morbidity and mortality in human. In most cases, these account for high prevalence of death and disability-adjusted life years lost. Several environmental, individual, and pathological conditions play important roles in gaining respiratory disorders. In many studies, *N. sativa* and its components such as carvacrol, nigellone, thymoquinone have been found act against respiratory diseases. The molecular mechanisms and the pre-clinical or clinical reports suggest that *N. sativa* may be one of the most promising sources of respiratory drugs.

Keywords: black cumin; carvacrol; nigellone; thymoquinone; lung diseases.

RESUMO

O uso de *Nigella sativa* e de preparações obtidas a partir dela é bastante comum para o tratamento de diversas doenças. Dentre estas, doenças pulmonares como asma, bronquite e doença pulmonar obstrutiva crônica constituem grandes causas de morbidade e mortalidade. Na maioria dos casos, estas representam alta prevalência de mortalidade, além de incapacitação por vários anos de vida. Várias condições ambientais, individuais e patológicas desempenham papéis importantes no desenvolvimento de distúrbios respiratórios. Em muitos estudos, *N. sativa* e seus componentes, como carvacrol, nigellona e timoquinona demonstraram ter atividade no tratamento de doenças respiratórias. Os estudos de mecanismos moleculares, estudos pré-clínicos ou clínicos sugerem que a *N. sativa* pode ser uma das fontes mais promissoras de princípios com atividade para o tratamento de doenças do trato respiratório.

Palavras-chave: carvacrol; cominho preto; doenças pulmonares; nigellona.

1. INTRODUCTION

To date, respiratory disease is a common and significant cause of illness and death around the world. The chronic lung disease or chronic obstructive pulmonary disease such as asthma, chronic bronchitis and emphysema are some forms causing serious deaths, even in developed countries (BLF, 2008). Moreover, lung cancer still occupies the leading cause of cancer-related deaths per year (PHAC, 2008). In the United States, in 2011, respiratory disease with ventilator support accounted for 93.3% of ICU utilization (BARRETT et al., 2014). Thus, respiratory diseases affect all classes of populations and cost a lot every year around the world.

Nowadays, natural lead compounds are spotlighted. The miracle herb *Nigella sativa* (Family: Ranunculaceae) and its derived components have been reported for many important biological activities (ISLAM et al., 2016). It has been demonstrated in many studies that *N. sativa* is a good remedy in the treatment of bronchial asthma and eczema (KALUS et al., 2003). Moreover, *N. sativa* alone or in combination with honey have been traditionally used for the treatment of respiratory disorders such as asthma, bronchospasm, and chest congestion (AVE-SINA, 1990).

To date, a number of *N. sativa* components such as alpha-hederin (AHN), carvacrol, nigellicine, nigellidine, thymoquinone (TQ), dithymoquinone, and thymol have been proven to act against respiratory diseases (AHMAD et al., 2013). Nigellone, a carbonyl polymer of TQ has been considered as a promising phytotherapeutic tool in asthma and bronchitis (ISLAM, 2016a). The anti-asthmatic activity may be due to its inhibitory effect of histamine release from the mast cells (CHAKRAVARTY 1993) and an anti-inflammatory effect by inhibiting the synthesis of 5-lipoxygenase products in polymorphonuclear leucocytes (EL-DAKHAKHNY et al., 2002). Interestingly, most of the activities suggested for *N. sativa* are similar to those of its promising component TQ (ISLAM et al., 2016).

This review aims to sketch the therapeutic promises of *N. sativa* and its components in respiratory diseases.

2. METHODOLOGY

Online literature searches were done in the following databases: Pubmed, Science Direct, Scopus, and Google Scholar till December 2016 to identify articles, editorials, and reviews about preventive and relieving effects of *N. sativa* and its components on respiratory diseases.

3. FINDINGS

The chemical composition and overall activities of *N. sativa* and its seed oil have been discussed in one of my recent studies (ISLAM, 2016b). Moreover, two individual reviews have been also done on nigellone (ISLAM, 2016a) and TQ (ISLAM et al., 2016). Therefore, this paper is focused only on the findings on *N. sativa* and its components in respiratory diseases.

In lung inflammation, oxidative stress, pro-inflammatory and inflammatory mediators as well as immunological activities play vital roles. However, viral and bacterial agents are also evident to cause lung infection. *N. sativa* and its components, especially the carvacrol, TQ and nigellone are evident to act as antioxidant, anti-inflammatory, antimicrobial and immunomodulatory agents in a number of studies (GHOLAMNEZHAD et al., 2015; ISLAM, 2016a,b; ISLAM et al., 2016).

3.1. *N. sativa* in respiratory diseases

N. sativa (0.1-3.0 mg/ml) in isolated rabbit jejunum and guinea pigs tracheal preparations caused a dose-dependent relaxation of spontaneous contractions in rabbit jejunum and inhibited K^+-induced contractions, suggesting calcium channel blockade. The effect was verified with verapamil (a standard calcium channel blocker). In the guinea pig trachea, it was found to cause relaxation of carbachol-, histamine- or K^+-induced contractions, indicating a spasmolytic and bronchodilator activity mediated, possibly through calcium channel blockade pathway (GILANI et al., 2001). In a study, the *N. sativa* oil in 152

(capsules: 40 to 80 mg/kg/day) patients with allergic diseases (allergic rhinitis, bronchial asthma, atopic eczema) decreased ~~in~~ plasma triglycerides and increased ~~in~~ high-density lipoprotein (HDL) cholesterol levels. Although the lymphocyte subpopulations, endogenous cortisol levels and adrenocorticotropic hormone (ACTH) release remained unchanged, but IgE, eosinophil count, total cholesterol, and LDL cholesterol were found to decrease (KALUS et al., 2003). Furthermore, the aqueous extract of *N. sativa* showed an inhibitory effect on the pre-contracted tracheal chains in the presence of both ordinary and calcium free Krebs solution in guinea pigs (BOSKABADY et al., 2004).

N. sativa and dexamethasone treated in conalbumin (i.p.)-induced allergic asthmatic mouse were significantly reduced the peripheral blood eosinophil count, immunoglobulin (Ig)G1 and IgG2a levels, cytokine profiles and inflammatory cells in lung tissue (ABBAS et al., 2005). The essential oil of *N. sativa* seeds and its main components investigated on human neutrophil elastase activity suggests exerting an antielastase effect (half-minimal inhibitory concentration, IC_{50}: 12 µM). The author supposed that the responsible component may be carvacrol, which can be used in the treatment of injuries appeared gained from chronic obstructive pulmonary disease and emphysema (KACEM; MERAIHI, 2006).

The boiled extract of *N. sativa* (15 ml/kg for 3 months) in asthmatic adults (n = 14/15) was found to improve all asthma symptoms, frequency of asthma symptoms/week, chest wheezing, and pulmonary function test (PFT) values in comparison to the control group (BOSKABADY et al., 2007). Moreover, in forty (40) chemical war victims (n = 20) *N. sativa* boiled extract (0.375 ml/kg for 2 months) significantly improved all respiratory symptoms, chest wheezing, and PFT values (BOSKABADY; FARHADI, 2008). The boiled extract of *N. sativa* (50 and 100 mg/kg) in 15 asthmatic patients at 30 min to 3 h showed relatively potent anti-asthmatic effect in comparison to 6 mg/kg theophylline (BOSKABADY et al., 2010). Allergic rhinitis is the most common chronic and allergic disease, especially in children. Nikakhlagh et al (2011) demonstrated that, the *N. sativa* oil (30 days treatment) in 66 allergic rhinitis patients (22 males and 44 females) significantly reduced the nasal mucosal congestion, nasal itching, runny nose, sneezing attacks, turbinate hypertrophy, and mucosal pallor. Moreover, the *N. sativa* oil (intranasal) in 42 geriatric patients with nasal dryness and related symptoms was given for 3 weeks, where the oil was found to improve the nasal dryness, obstruction and crusting significantly in comparison to the saline treated group (OYSU et al., 2014).

Four cumulative concentrations of n-hexane, dichloromethane, methanol and aqueous fractions of *N. sativa* (0.8, 1.2, 1.6 and 2.0 g%) and theophylline (0.2, 0.4, 0.6 and 0.8 mM) were examined for their relaxant effects on precontracted tracheal chains of guinea pig, where a significant relaxing effect was observed in the methanol and dichloromethane fraction groups (BOSKABADY et al., 2008). *N. sativa* (0.08 g/d for 14 days treatment) in 100 mg/m^3 inhaled sulfur mustard guinea pigs (n = 6) was also decreased in tracheal responsiveness and lung inflammation along with the eosinophl, monocyte, and lympocytes in animals (HOSSEIN et al., 2008). In a study, the *N. sativa* volatile oil (400 mg/kg, i.g. for 7 days) in hydrochloric acid (2 ml/kg *via* injection) induced lung injured male Wistar rats, inhibited the inflammatory pulmonary responses, reduced peribronchial inflammatory cell infiltration, alveolar septal infiltration, alveolar edema, alveolar exudate, alveolar macrophages, interstitial fibrosis, granuloma and necrosis formation. The volatile oil also reduced the activity of inducible nitric oxide synthase (iNOS) in animals (KANTER, 2009).

The aqueous extract of *N. sativa* in BLAB/c and C57/BL6 primary cells significantly enhanced the splenocyte proliferation in a dose-responsive manner. In addition, it favored the secretion of T-helper cell (Th)-2, versus Th1, cytokines by splenocytes, while suppressing interleukin (IL)-6, tumor necrosis factor alpha (TNF-α), nitric oxide (NO), key pro-inflammatory mediators, primary macrophage levels (MAJDALAWIEH et al., 2010). On the other

hand, the hydro-ethanolic extract of *N. sativa* in 1% ovalalbumin (OA)-sensitized (i.p.) guinea pigs (n = 7) significantly decreased in tracheal responsiveness, while increasing in total white blood corpuscle (WBC) and eosinophil counts in the lung lavage fluid (BOSKABADY et al., 2011a). *N. sativa* (14 days treatment) in sulfur mustard-exposed guinea pigs (n = 7) significantly decreased tracheal responsiveness and neutrophil number, while increasing in eosinophil, monocyte, lymphocyte, IL-4 and interferon gamma (IFN-γ) levels animals (BOSKABADY et al., 2011b). In another study, Boskabady et al (2011c) also found a similar result for *N. sativa*, where they observed significant pathological changes along with an increase in blood IL-4 and IFN-γ levels in OA-sensitized guinea pigs (n = 8).

N. sativa (NS) in cecal ligation and puncture (CLP)-induced septic rats significantly decreased the proinflammatory cytokine levels in serum; lipid peroxidase (LPO) level, myeloperoxidase (MPO) activity, and pathological changes in lung tissues, while the increase in reduced glutathione (GSH) levels and superoxide dismutase (SOD) activity in the lung tissue. Furthermore, NS also reduced the CLP-induced in mortality of rats (BAYIR et al., 2012). Moreover, *N. sativa* oil (oral treatment) in ovalbumin (OA, 50 µg, i.p. for 30 days) BALB/c mice showed a significant decrease in airway hyperresponsiveness, the number of total leukocytes, macrophages and eosinophils, levels of IL-4, -5 and -13 in the bronchoalveolar lavage fluid (BALF), serum levels of total IgE, OVA-specific IgE and IgG1, and significant increase in BALF level of IFN-γ and serum level of OA-specific IgG2a, indicating the restoration of local Th1/Th2 balance. Interestingly the oil abrogated the histopathological changes of the lungs as like the control group (BALAHA et al., 2012).

Two flavonoids (20-20% and 21-20% fractions) from methanolic fraction of *N. sativa* in guinea pig (n = 6) were found to exert a potent bronchodilatory effect (KEYHANMANESH et al., 2013). The *N. sativa* oil (4 ml/kg, i.p.) in hyperoxia-induced lung injured rats was also evident to decrease the severity of lung damage, malonaldehyde (MDA) and MPO levels, while increased SOD and glutathione peroxidase (GSH-Px levels (TAYMAN et al., 2013). On the other hand, the seed extract and seed oil (0.25, 0.5 and 1 mg/ml) of *N. sativa* were found to reduce cell viability of a human lung cancer (A-549) cell line in 3-(4, 5-dimethylthiazol-2yl)-2, 5-biphenyl tetrazolium bromide (MTT) assay (AL-SHEDDI et al., 2014).

N. sativa (500 mg/kg) and its essential oil (EO) (5 ml/kg) were given orally in lipopolysaccharides (LPS, 1 mg/kg, i.p.) treated and to determine the lung inflammation, 18F-fluoro-deoxy-D-glucose (0.8 ml/kg) was administrated in animals under the anesthesia before the 1 h of PET-scanning. Both *N. sativa* and EO significantly decreased in 18F-FDG uptake, DNA fragmentation and NO levels (ENTOK et al., 2014). Additionally, the *N. sativa* fixed oil also exhibited an immunomodulatory and anti-inflammatory effect of significant reduction of peripheral blood eosinophils (PBE) of allergic mice (n = 6 to 10) (ABDEL-AZIZ et al., 2014).

3.2. *N. sativa*-derived components against pulmonary diseases

The volatile oil of *N. sativa* (4-32 µl/kg) in urethane-anaesthetized guinea pigs significantly antagonized the effects of mepyramine, atropine and reserpine. Furthermore, an intravenous (i.v.) administration of TQ (1.6-6.4 mg/kg) was found to increase the intra-tracheal pressure without affecting the respiratory rate, suggesting it may act *via* release of histamine involving histaminergic mechanisms and indirect activation of muscarinic cholinergic mechanisms (EL TAHIR et al., 1993). In a study, both AHD and TQ were found to act against human lung cancer (A549) and larynx epidermoid (HEp-2) cell lines (ROONEY; RYAN, 2005). TQ is also evident to inhibit 5-lipoxygenase (the main enzyme in the biosynthesis of leukotriene) (EL GAZZAR et al., 2006a), decrease tension in the tracheal muscle and inhibition of lipoxygenase products of arachidonic acid metabolism as well as non-selective blocking of the histamine and serotonin receptors (AL-MAJED et al., 2001) in rodents. Moreover, TQ has been shown to be useful in the treatment of acute

respiratory distress syndrome in rats (ISIK et al., 2005).

In a review works, Butt and Sultan (2010) demonstrated that, the *N. sativa* and its derived components such as TQ, AHD (saponin), carvacrol, proteins, and alkaloids (nigellicines and nigelledine) can be used in various maladies, including lung disease. Khan et al (2011) also suggested that *N. sativa*, crude oil and TQ can be used in lung disease. An intraperitoneal injection of TQ in OA-sensitized allergic mice was found to prevent Th2 cytokines and eosinophil infiltration and goblet cell hyperplasia in the airways. Additionally, TQ down-regulated IL-4, -5 and -13, while up-regulated IFN-γ levels (EL GAZZAR et al., 2006b). In another study, an intraperitoneal TQ injection for 5 days is also evident to exert an almost similar result, additionally with the inhibition of cyclooxigenase (COX)2 protein expression and prostaglandin (PG)D2 production (EL MEZAYEN et al., 2006). Suddek (2010) supposed that, the TQ-mediated relaxation effect in phenylephrine-induced pulmonary artery contraction, is possibly *via* ATP-sensitive channels and probably by non-competitive blocking of serotonin, α1 and endothelin receptors.

Moreover, TQ exerted an anti-apoptotic effect and attenuated lung injury in chronic toluene exposure rats (KANTER, 2011). In another study, TQ significantly decreased the progression of pulmonary fibrosis in belomycin-induced rats, where it counteracted emphysema in air alveoli, inflammatory cell infiltration, lymphoid hyperplastic cell activation surrounding the bronchioles as well as over-expression of NF-κB in lung tissue. Furthermore, TQ also restored the antioxidant enzymes SOD and glutathione-S-tranferase (GST) levels toward normal value (EL-KHOULY et al., 2012). In cyclophosphamide-induced lung injured rats, TQ (7 days treatment) significantly improved histopathological changes in lung tissue of the animals. Moreover, it also decreased the serum biomarkers such as total protein, LDH, TNF-α, and lipid peroxidation, while increasing the antioxidant enzyme levels (SUDDEK et al., 2013).

In a recent study, both TQ (3 mg/kg, i.p.) and AHD (0.3 and 3.0 mg/kg i.p.) in OA-sensitized guinea pigs were evident to decrease WBC and eosinophil counts, while an increase in neutrophil, lymphocyte and monocyte counts along with a bronchodilatory effect (SAADAT et al., 2015). Moreover, in OA-sensitized guinea pigs (n = 8) TQ and AHD significantly decreased the levels of IL-4 and -17 with an increase in IFN-γ in comparison to the control group (KEYHANMANESH et al., 2015). Furthermore, both TQ (3 mg/kg, i.p.) and AHD (0.02 mg/kg, i.p.) were found to decrease the levels of micro-RNA (miRNA)-126, IL-13 mRNA and cause pathological changes in the lungs of the OA-sensitized rats (n = 8) (FALLAHI et al., 2016). Ebrahimi et al (2016) suggested that, TQ (3 mg/kg) or OA-sensitized pretreated with AHD (0.02 mg/kg) in rats altered the OA-mediated pathological changes. Additionally, both of them increased serum antioxidant enzyme levels and miRNA-133a gene expression, while decreasing IL-2, and -17 mRNA. In a recent study, TQ was found to improve the macrophage phagocytosis *via* modulation of the S1P system and protect bronchial epithelial cells from cigarette smoke or LPS-induced apoptosis, demonstrating a potential therapeutic agent for smoking-related lung diseases (BARNAWI et al., 2016).

Moreover, nigellone and TQ were also evident to inhibit tracheal contraction with a modulatory effect on mucociliary clearance in rodents (WIENKÖTTER et al., 2008), while carvacrol (20, 40 or 80 mg/kg) in LPS-induced endotoxemia and acute lung injury in mice exerted an anti-inflammatory effect, possibly *via* inhibition of nuclear factor kappa B (NF-κB) and mitogen-activated protein kinases (MAPKs) signaling pathways, thereby inhibiting inflammatory cytokines TNF-α, IL-6 and -1β production (FENG; JIA, 2014). In another study, carvacrol (73 mg/kg, i.p.) in methotrexate (20 mg/kg, i.p. for 8 days)-induced lung injury in rats increased in total antioxidant capacity, while decreasing in MDA and oxidative stress index in rat lung tissue (SELIMOĞLU ŞEN et al., 2014). On the other hand, carvacrol on systemic inflammation (cigarette smoke for 3 months) in guinea pig (n = 6) decreased in serum IL-8 and MDA, total WBC, eosinophil, neutrophil and

lymphocyte counts (MAHTAJ et al., 2015). Boskabady et al (2015) demonstrated that, carvacrol (60, 120, and 240 µg/ml) in drinking water in a 3 months cigarette smoked guinea pigs (n = 6) was found to increase the total WBC, eosinophils, and neutrophils counts as well as the levels of IL-8, while Gholami Mahtaj et al (2015) found an improved tracheal response and pathological changes in the lung in chronic obstructive pulmonary disease (COPD) guinea pigs (n = 6).

~~C~~arvacrol/thymol is also evident to exert cytotoxic effects towards BEAS-2B transformed human bronchial epithelial cells and in A549 and H292 lung carcinoma cells (KHOSRAVI; ERLE, 2016), while carvacrol in OA-sensitized mice was found to decrease in IL-4, IFN-γ, tumor growth factor-beta (TGF-β), forkhead box P3 protein (FOXP3), and IL-17 genes expressions, which indicates its possible therapeutic value in allergy, autoimmunity, and infectious diseases (KIANMEHR et al., 2016). Chemical structures of the *N. sativa* respiratory acting phytochemicals have been shown in **Figure 1**.

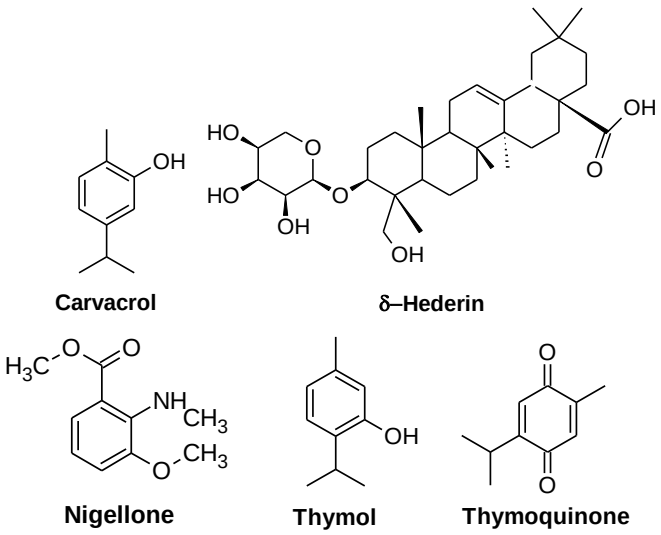

Figure 1 – Chemical structures of *N. sativa* respiratory acting compounds.

4. CONCLUSION

A number of non-clinical, pre-clinical and clinical reports suggest that different preparations of *N. sativa* and its isolated components have pulmonary system protective capabilities. The dietary consumption of the plant seed and its oil or as adjuvant therapy may bring some important therapeutic values in the treatment of many diseases, including pulmonary diseases. However, further investigations are needed to determine the bioavailability and other pharmacokinetic properties as well as safety ranges of each preparation of *N. sativa*. Advanced pharmaceutical concerns are also highly appreciated to the synthesis of analogues and prepare nano-encapsulated formulations, especially with carvacrol, TQ, and nigellone.

CONFLICT OF INTEREST

None declared.

REFERENCES

ABBAS, A.T.; ABDEL-AZIZ, M.M.; ZALATA, K.R.; ABD AL-GALEL, TEL.-D. Effect of dexamethasone and *Nigella sativa* on peripheral blood eosinophil count, IgG1 and IgG2a, cytokine profiles and lung inflammation in murine model of allergic asthma. Egypt. J. Immunol. v. 12, p. 95-102, 2005.

ABDEL-AZIZ, M.; ABASS, A.; ZALATA, K.; ABD AL-GALEL, T.; ALLAM, U.; KARROUF, G. Effect of dexamethasone and *Nigella sativa* on inducible nitric oxide synthase in the lungs of a murine model of allergic Asthma. Iran. J. Allergy Asthma Immunol. v. 13, p. 324-334, 2014.

AHMAD, A.; HUSAIN, A.; MUJEEB, M.; KHAN, S.A.; NAJMI, A.K.; SIDDIQUE, N.A.; DAMANHOURI, Z.A.; ANWAR, F. A review on therapeutic potential of *Nigella sativa*: A miracle herb. Asian Pac. J. Tropic. Biomed. v. 3, p. 337-352, 2013.

AL-MAJED, A.A.; DABA, M.H.; ASIRI, Y.A.; AL-SHABANAH, O.A.; MOSTAFA, A.A.; EL-KASHEF, H.A. Thymoquinone-induced relaxation of guinea-pig isolated trachea. Res. Commun. Mol. Pathol. Pharmacol. v. 110, p. 333-345, 2001.

AL-SHEDDI, E.S.; FARSHORI, N.N.; AL-OQAIL, M.M.; MUSARRAT, J.; AL-KHEDHAIRY, A.A.; SIDDIQUI, M.A. Cytotoxicity of *Nigella sativa* seed oil and extract against human lung cancer cell line. Asian Pac. J. Cancer Prev. v. 15, p. 983-987, 2014.

AVE-SINA (Ed.) *Law in medicine* (Vol. 2). Teheran: Ministry of Guidance Publication. 1990.

BALAHA, M.F.; TANAKA, H.; YAMASHITA, H.; ABDEL RAHMAN, M.N.; INAGAKI, N. Oral *Nigella sativa* oil ameliorates ovalbumin-induced bronchial asthma in mice. Int. Immunopharmacol. v. 14, p. 224-231, 2012.

BARNAWI, J.; TRAN, H.B.; ROSCIOLI, E.; HODGE, G.; JERSMANN, H.; HABERBERGER, R.; HODGE, S. Pro-phagocytic Effects of Thymoquinone on Cigarette Smoke-exposed Macrophages Occur by Modulation of the Sphingosine-1-phosphate Signalling System. COPD. v. 13, p. 653-661, 2016.

BARRETT, M.L.; SMITH, M.W.; ELIZHAUSER, A.; HONIGMAN, L.S.; PINES, J.M. Utilization of Intensive Care Services, 2011. HCUP Statistical Brief #185. Rockville, MD: Agency for Healthcare Research and Quality. 2014.

BAYIR, Y.; ALBAYRAK, A.; CAN, I.; KARAGOZ, Y.; CAKIR, A.; SULEYMAN, H.; UYANIK, H.; YAYLA, N.; POLAT, B.; KARAKUS, E.; KELES, M.S. *Nigella sativa* as a potential therapy for the treatment of lung injury caused by cecal ligation and puncture-induced sepsis model in rats. Cell. Mol. Biol. (Noisy-le-grand) v. 58, p. OL1680-1687, 2012.

BLF (British Lung Foundation). Facts about ukinnam respiratory disease. Retrieved 2008-04-19, 2008.

BOSKABADY, M.H.; SHIRMOHAMMADI, B.; JANDAGHI, P.; KIANI, S. Possible mechanism(s) for relaxant effect of aqueous and macerated extracts from *Nigella sativa* on tracheal chains of guinea pig. BMC Pharmacol. v. 4, p. 3, 2004.

BOSKABADY, M.H.; JAVAN, H.; SAJADY, M.; RAKHSHANDEH, H. The possible prophylactic effect of *Nigella sativa* seed extract in asthmatic patients. Fundam. Clin. Pharmacol. v. 21, p. 559-566, 2007.

BOSKABADY, M.H.; FARHADI, J. The possible prophylactic effect of *Nigella sativa* seed aqueous extract on respiratory symptoms and pulmonary function tests on chemical war victims: a randomized, double-blind, placebo-controlled trial. J. Altern. Complement. Med. v. 14, p. 1137-1144, 2008.

BOSKABADY, M.H.; KEYHANMANESH, R.; SAADATLOO, M.A. Relaxant effects of different fractions from *Nigella sativa* L. on guinea pig tracheal chains and its possible mechanism(s). Indian J. Exp. Biol. v. 46, p. 805-810, 2008.

BOSKABADY, M.H.; MOHSENPOOR, N.; TAKALOO, L. Antiasthmatic effect of *Nigella sativa* in airways of asthmatic patients. Phytomed. v. 17, p. 707-713, 2010.

BOSKABADY, M.H.; KEYHANMANESH, R.; KHAMNEH, S.; EBRAHIMI, M.A. The effect of *Nigella sativa* extract on tracheal responsiveness and lung inflammation in ovalbumin-sensitized guinea pigs. Clinics (Sao Paulo) v. 66, p. 879-887, 2011a.

BOSKABADY, M.H.; VAHEDI, N.; AMERY, S.; KHAKZAD, M.R. The effect of *Nigella sativa* alone, and in combination with dexamethasone, on tracheal muscle responsiveness and lung inflammation in sulfur mustard exposed guinea pigs. J. Ethnopharmacol. v. 137, p. 1028-1034, 2011b.

BOSKABADY, M.H.; KEYHANMANESH, R.; KHAMENEH, S.; DOOSTDAR, Y.; KHAKZAD, M.R. Potential immunomodulation effect of the extract of *Nigella sativa* on ovalbumin sensitized guinea pigs. J. Zhejiang Univ. Sci. B. v. 12, p. 201-209, 2011c.

BOSKABADY, M.H.; GHOLAMI MAHTAJ, L. Lung inflammation changes and oxidative stress induced by cigarette smoke exposure in guinea pigs affected by *Zataria multiflora* and its constituent, carvacrol. BMC Complement. Altern. Med. v. 15, p. 39, 2015.

BUTT, M.S.; SULTAN, M.T. *Nigella sativa*: reduces the risk of various maladies. Critic. Rev. Food Sci. Nutr. v. 50, p. 654-665, 2010.

CHAKRAVARTY, N. Inhibition of histamine release from mast cells by nigellone. Ann. Allergy v. 70, p. 237-242, 1993.

EBRAHIMI, H.; FALLAHI, M.; KHAMANEH, A.M.; EBRAHIMI SAADATLOU, M.A.; SAADAT, S.; KEYHANMANESH, R. Effect of α-Hederin on IL-2 and IL-17 mRNA and miRNA-133a Levels in Lungs of Ovalbumin-Sensitized Male Rats. Drug Dev. Res. v. 77, p. 87-93, 2016.

EL GAZZAR, M.; EL MEZAYEN, R.; MARECKI, J.C.; NICOLLS, M.R.; CANASTAR, A.; DRE-SKIN, S.C. Anti-inflammatory effect of thymoquinone in a mouse model of allergic lung inflammation. Int. Immunopharmacol. v. 6, p. 1135-1142, 2006b.

EL GAZZAR, M.; EL MEZAYEN, R.; NICOLLS, M.R.; MARECKI, J.C.; DRESKIN, S.C. Down-regulation of leukotriene biosynthesis by thymoquinone attenuates airwayinflammation in a mouse model of allergic asthma. Biochim. Biophys. Acta v. 1760, p. 1088-1095, 2006a.

EL MEZAYEN, R.; EL GAZZAR, M.; NICOLLS, M.R.; MARECKI, J.C.; DRESKIN, S.C.; NOMIYAMA, H. Effect of thymoquinone on cyclooxygenase expression andprostaglandin production in a mouse model of allergic airway inflammation. Immuno. Lett. v. 106, p. 72-81, 2006.

EL TAHIR, K.E.; ASHOUR, M.M.; AL-HARBI, M.M. The respiratory effects of the volatile oil of the black seed (*Nigella sativa*) in guinea-pigs: elucidation of the mechanism(s) of action. Gen. Pharmacol. v. 24, p. 1115-1122, 1993.

EL-DAKHAKHNY, M.; MADI, N.J.; LEMBERT, N.; AMMON, H.P. *Nigella sativa* oil, nigellone and derived thymoquinone inhibit synthesis of 5-lipoxygenase products in polymorphonuclear leukocytes from rats. J. Ethnopharmacol. v. 81, p. 161-164, 2002.

EL-KHOULY, D.; EL-BAKLY, W.M.; AWAD, A.S.; EL-MESALLAMY, H.O.; EL-DEMERDASH, E. Thymoquinone blocks lung injury and fibrosis by attenuating bleomycin-induced oxidative stress and activation of nuclear factor Kappa-B in rats. Toxicol. v. 302, p. 106-113, 2012.

ENTOK, E.; USTUNER, M.C.; OZBAYER, C.; TEKIN, N.; AKYUZ, F.; YANGI, B.; KURT, H.; DEGIRMENCI, I.; GUNES, H.V. Anti-inflammatuar and anti-oxidative effects of *Nigella sativa* L.: 18FDG-PET imaging of inflammation. Mol. Biol. Rep. v. 41, p. 2827-2834, 2014.

FALLAHI, M.; KEYHANMANESH, R.; KHAMANEH, A.M.; EBRAHIMI SAADATLOU, M.A.; SAADAT, S.; EBRAHIMI, H. Effect of Alpha-Hederin, the active constituent of *Nigella sativa*, on miRNA-126, IL-13 mRNA levels and inflammation of lungs in ovalbumin-sensitized male rats. Avicenna J. Phytomed. v. 6, p. 77-85, 2016.

FENG, X.; JIA, A. Protective effect of carvacrol on acute lung injury induced by lipopolysaccharide in mice. Inflammation v. 37, p. 1091-1101, 2014.

GHOLAMI MAHTAJ, L.; BOSKABADY, M.H.; MOHAMADIAN ROSHAN, N. The Effect of *Zataria multiflora* and its Constituent, Carvacrol, on Tracheal Responsiveness and Lung Pathology in Guinea Pig Model of COPD. Phytother. Res. v. 29, p. 730-736, 2015.

GHOLAMNEZHAD, Z.; KEYHANMANESH, R.; BOSKABADY, M.H. Anti-inflammatory, antioxidant, and immunomodulatory aspects of *Nigella sativa* for its preventive and bronchodilatory effects on obstructive respiratory diseases: A review of basic and clinical evidence. J. Functional Foods v. 17, p. 910-927, 2015.

GILANI, A.H.; AZIZ, N.; KHURRAM, I.M.; CHAUDHARY, K.S.; IQBAL, A. Bronchodilator, spasmolytic and calcium antagonist activities of *Nigella sativa* seeds (Kalonji): a traditional herbal product with multiple medicinal uses. J. Pak. Med. Assoc. v. 51, p. 115-120, 2001.

HOSSEIN, B.M.; NASIM, V.; SEDIQA, A. The protective effect of *Nigella sativa* on lung injury of sulfur mustard-exposed Guinea pigs. Exp. Lung Res. v. 34, p. 183-194, 2008.

ISIK, A.F.; KATI, I.; BAYRAM, I.; OZBEK, H. A new agent for treatment of acute respiratory distress syndrome: thymoquinone. An experimental study in a rat model. Eur. J. Cardiothorac. Surg. v. 28, p. 301-305, 2005.

ISLAM, M.T. Nigellone, A Buoyant Chemical Moiety. Asian J. Ethnopharmacol. Med. Foods v. 02, p. 10-13, 2016a.

ISLAM, M.T. Biological activities and therapeutic promises of *Nigella sativa* L. Int. J. Pharm. Sci. Scientif. Res. v. 2, p. 237-252, 2016b.

ISLAM, M.T.; SULTANA, N.; RIAZ, T.A.; FERDOUS, J.; GUHA, B.; MOHAGON, S.; MUTSUDDY, R.; SANTOS, J.V.O.; REIS, A.C.; BRAGA, A.L.; CERQUEIRA, G.S.; MENEZES, A.-A.P.M.; MELO-CAVALCANTE, A.A.C. Thymoquinone is knocking at the door of clinical trial. Int. Arch. Med. v. 9, p. 1-25, 2016.

KACEM, R.; MERAIHI, Z. Effects of essential oil extracted from *Nigella sativa* (L.) seeds and its main components on human neutrophil elastase activity. Yakugaku Zasshi v. 126, p. 301-305, 2006.

KALUS, U.; PRUSS, A.; BYSTRON, J.; JURECKA, M.; SMEKALOVA, A.; LICHIUS, J.J.; KIESEWETTER, H. Effect of *Nigella sativa* (black seed) on subjective feeling in patients with allergic

diseases. Phytother. Res. v. 17, p. 1209-1214, 2003.

KANTER, M. Effects of *Nigella sativa* seed extract on ameliorating lung tissue damage in rats after experimental pulmonary aspirations. Acta Histochem. v. 111, p. 393-403, 2009.

KANTER, M. Thymoquinone attenuates lung injury induced by chronic tolueneexposure in rats. Toxicol. Ind. Health. v. 27, p. 387-395, 2011.

KEYHANMANESH, R.; BAGBAN, H.; NAZEMIEH, H.; MIRZAEI BAVIL, F.; ALIPOUR, M.R. The main relaxant constituents of *Nigella sativa* methanolic fraction on Guinea pig tracheal chains. Iran. J. Allergy Asthma Immunol. v. 12, p. 136-143, 2013.

KEYHANMANESH, R.; SAADAT, S.; MOHAMMADI, M.; SHAHBAZFAR, A.A.; FALLAHI, M. The Protective Effect of α-Hederin, the Active Constituent of *Nigella sativa*, on Lung Inflammation and Blood Cytokines in Ovalbumin Sensitized Guinea Pigs. Phytother. Res. v. 29, p. 1761-1767, 2015.

KHAN, M.A.; CHEN, H.C.; TANIA, M.; ZHANG, D.Z. Anticancer activities of *Nigella sativa* (black cumin). Afr. J. Tradit. Complement. Altern. Med. v. 8, p. 226-232, 2011.

KHOSRAVI, A.R.; ERLE, D.J. Chitin-Induced Airway Epithelial Cell Innate Immune Responses Are Inhibited by Carvacrol/Thymol. PLoS One v. 11, p. e0159459, 2016.

KIANMEHR, M.; REZAEI, A.; BOSKABADY, M.H. Effect of carvacrol on various cytokines genes expression in splenocytes of asthmatic mice. Iran. J. Basic Med. Sci. v. 19, p. 402-410, 2016.

MAHTAJ, L.G.; FEIZPOUR, A.; KIANMEHR, M.; SOUKHTANLOO, M.; BOSKABADY, M.H. The effect of carvacrol on systemic inflammation in guinea pigs model of COPD induced by cigarette smoke exposure. Pharmacol. Rep. v. 67, p. 140-145, 2015.

MAJDALAWIEH, A.F.; HMAIDAN, R.; CARR, R.I. *Nigella sativa* modulates splenocyte proliferation, Th1/Th2 cytokine profile, macrophage function and NK anti-tumor activity. J. Ethnopharmacol. v. 131, p. 268-275, 2010.

NIKAKHLAGH, S.; RAHIM, F.; ARYANI, F.H.; SYAHPOUSH, A.; BROUGERDNYA, M.G.; SAKI, N. Herbal treatment of allergic rhinitis: the use of *Nigella sativa*. Am. J. Otolaryngol. v. 32, p. 402-407, 2011.

OYSU, C.; TOSUN, A.; YILMAZ, H.B.; SAHIN-YILMAZ, A.; KORKMAZ, D.; KARAASLAN, A. Topical *Nigella Sativa* for nasal symptoms in elderly. Auris. Nasus. Larynx v. 41, p. 269-272, 2014.

PHAC (Public Health Agency of Canada). Centre for Chronic Disease Prevention and Control Chronic Respiratory Diseases. Retrieved 2008-05-06, 2008.

ROONEY, S.; RYAN, M.F. Effects of alpha-hederin and thymoquinone, constituents of *Nigella sativa*, on human cancer cell lines. Anticancer Res. v. 25, 2199-2204, 2005.

SAADAT, S.; MOHAMMADI, M.; FALLAHI, M.; KEYHANMANESH, R.; ASLANI, M.R. The protective effect of α-hederin, the active constituent of *Nigella sativa*, on tracheal responsiveness and lung inflammation in ovalbumin-sensitized guinea pigs. J. Physiol. Sci. v. 65, p. 285-292, 2015.

SELIMOĞLU ŞEN, H.; ŞEN, V.; BOZKURT, M.; TÜRKÇÜ, G.; GÜZEL, A.; SEZGI, C.; ABAKAY, Ö.; KAPLAN, I. Carvacrol and pomegranate extract in treating methotrexate-induced lung oxidative injury in rats. Med. Sci. Monit. v. 20, p. 1983-1990, 2014.

SUDDEK, G.M. Thymoquinone-induced relaxation of isolated rat pulmonaryartery. J. Ethnopharmacol. v. 127, p. 210-214. 2014.

SUDDEK, G.M.; ASHRY, N.A.; GAMEIL, N.M. Thymoquinone attenuates cyclophosphamide-induced pulmonary injury in rats. Inflammopharmacol. v. 21, p. 427-435, 2013.

TAYMAN, C.; CEKMEZ, F.; KAFA, I.M.; CANPOLAT, F.E.; CETINKAYA, M.; TONBUL, A.; UYSAL, S.; TUNC, T.; SARICI, S.U. Protective effects of *Nigella sativa* oil in hyperoxia-induced lung injury. Arch. Bronconeumol. v. 49, p. 15-21, 2013.

WIENKÖTTER, N.; HÖPNER, D.; SCHÜTTE, U.; BAUER, K.; BEGROW, F.; EL-DAKHAKHNY, M.; VERSPOHL, E.J. The effect of nigellone and thymoquinone on inhibiting trachea contraction and mucociliary clearance. Planta Med. v. 74, p. 105-108, 2008.