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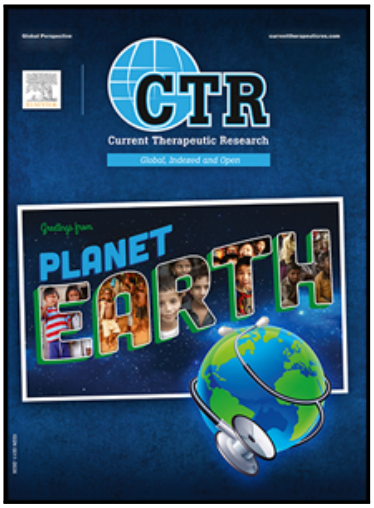


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# *Nigella sativa* L. as a Potential Phytotherapy for COVID-19: A Mini-Review of *In-silico* Studies

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## Abstract

### Background

Coronaviruses are responsible for several human diseases such as the pandemic infectious disease 2019 (COVID-19) which is caused by the Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2). *Nigella sativa* (NS) is a natural food supplement with a known safety profile that may provide a wealth of known antiviral compounds.

### Objective

To explore the studies supporting the NS potential for hitting SARS-CoV-2 targets.

### Methods

A literature search for scientific published or preprint *in-silico* studies between 1990 and 2020 in electronic databases (PubMed, Science Direct, Scopus, and Google Scholar) was performed for the terms *Nigella sativa*, black seed, coronavirus, SARS-CoV-2 and COVID-19.

**Results**

At least eight in-silico studies have shown that some compounds of NS, including Nigelledine, α-Hederin, Hederagenin, Thymohydroquinone, and Thymoquinone, had high to moderate affinity with SARS-CoV-2 enzymes and proteins. These compounds may potentially inhibit SARS-CoV-2 replication and attachment to host cell receptors.

**Conclusions**

These preliminary data propose NS as a potential phytotherapy candidate for COVID-19. Further preclinical experimental evidence is required followed by a phase 1 clinical trial.

**Keywords:** SARS-CoV-2; COVID-19; Coronavirus; *In-silico*; *Nigella sativa*

**Running Title:** *Nigella sativa* in COVID-19

## 1. Introduction

Coronaviruses (CoVs), enveloped RNA viruses, are characterized by spikes on their surface and belong to *Nidovirales* order [1]. They are responsible for a growing economic, social, and mortality burden in humans over the last decades. The spectrum of diseases associated with human CoVs range from the common cold to severe acute respiratory syndrome (SARS-CoV), and Middle East respiratory syndrome (MERS-CoV). Since December 2019, a newly discovered coronavirus (SARS-CoV-2) was the causative agent of the current pandemic of infectious disease (COVID-19). Unfortunately, there are no effective approved antiviral agents for these coronavirus strains [2,3].

Natural products provide a wealth of biologically active molecules with antiviral activity, and thus may have utility as potential therapeutic agents against coronavirus infections [4]. One of these products is Nigella sativa (NS), which has displayed several anti-viral properties [5].

NS is a well-known food supplement and medicinal plant in different cultures. The seeds of NS contain several active compounds in the classes of fixed oil, essential oil, saponins and alkaloids. In the literature, NS exhibited several pharmacological properties including anti-inflammatory, anti-microbial, and immunostimulatory activities [5,6].

The safety and efficacy of NS used for many human diseases has been established in several randomized clinical studies [7]. We also used NS oil in a randomized double-blind placebo-controlled trial on asthmatic patients with acceptable safety and efficacy profile [8]. Moreover, several meta-analyses have confirmed the beneficial effects and safety of NS on hyperlipidemia, type 2 diabetes, obesity, hypertension, and asthma [9–13]. Oral NS oil dosing of up to 5 grams daily for up to 12 weeks is believed to be possibly safe [14].

In *in vitro* studies, the antiviral activities of NS on different viruses were documented in the literature [5]. NS oil suppresses the viral load of Murine cytomegalovirus in infected mice to an undetectable level [15]. NS honey was found to inhibit HIV-1 replication [16]. NS had virucidal activity against herpes simplex and hepatitis A virus infections [16]. NS decreased the coronavirus load in infected HeLa cells with stimulated interleukin-8 secretion and down-regulation of Transient Receptor Potential genes expression such as TRPM6, TRPA1, TRPC4 and TRPM7 [17]. Hepatitis C virus (HCV) replication was inhibited by NS [18]. NS inhibited the growth of influenza virus H5N1 in *in vitro* [19].

In a human clinical study, patients with HCV infection showed significant improvement in HCV viral load after 3 months of NS treatment [20]. A case report of treatment with NS for 6 months showed a sustained seroreversion in a 46-year-old HIV patient and was also reported in an additional 6 HIV cases [21,22].

In recent years, *in-silico* molecular docking studies on natural products enable computational screening approaches for assessing their therapeutic potential. These studies utilize bioinformatics techniques and can be used to discover how candidate drugs cause therapeutic activity by predicting interactions between drugs and proteins, and analysis of impact on biological pathways and functions[23].

The aim of this mini literature review is to explore any published manuscript or preprint on *in-silico* studies of the specific anti-coronavirus potential of NS.

## 2. Methods

A literature search for scientific published manuscripts or preprint *in-silico* studies found in electronic databases (PubMed, Science Direct, Scopus, and Google Scholar) was performed using the terms *Nigella sativa*, black seed, coronavirus, SARS-CoV-2 and COVID-19. Studies were searched for electronically between the years 1990 and 2020.

## 3. Results

In this mini literature review, there were at least eight *in-silico* studies that explored the effects of NS compounds on SARS-CoV-2. A summary of those studies is presented in Table 1. However, there have been no reported clinical trials on NS in human coronavirus cases at this time.

Molecular docking of compounds from NS and some antiviral drugs was performed to determine their binding affinity with SARS-CoV-2 related molecular targets such as 6LU7, 2GTB, 6Y2E, ACE2 and HSPA5. The binding of some natural compounds might prevent the adhesion of coronavirus to host epithelial cells. Nigelledine, an alkaloid in NS, docked with 6LU7 active sites showed an energy complex score close to chloroquine and better than hydroxychloroquine and favipiravir. α-Hederin, a saponin in NS, docked with 2GTB active sites showed an energy score better than chloroquine, hydroxychloroquine and favipiravir [24]. Thymoquinone, the main essential oil constituent of NS, had a binding affinity with 6LU7, ACE2 and HSPA5 active sites with a score less than hydroxychloroquine in 6LU7 and ACE2 [25,26]. Also, Hederagenin, a saponin in NS, docked with 6LU7, 6Y2E, ACE2 and GRP78 active sites showed a binding score less than Saquinavir in 6LU7 and 6Y2E [27,28]. Thymohydroquinone showed moderate docking energy with SARS−CoV−2 Main-protease (6LU7), endoribonucleoase (Nsp15/NendoU), ADP−ribose−1″−phosphatase (ADRP), RNA−dependent RNA polymerase (RdRp), the binding domain of the SARS−CoV−2 spike protein (rS), and human ACE2 [29]. Nigellidine showed high

binding affinity SARS−CoV−2 enzymes and proteins such as N- terminus-protenase (QHD43415_3), main-protease (6LU7), non-structural protein 2 (NSP2), spike-glycoprotein(6vsb), and nucleocapsid (QHD43423). Nigellidine had high binding energy with human receptors, inflammatory signal molecules and other proteins such as human IL1R (1itb), TNFR1 (1ncf), TNFR2 (3alq) [30].

Therefore, the natural compounds found in NS such as nigellidine, α-Hederin, hederagenin, thymohydroquinone and thymoquinone were potentially active compounds that might inhibit coronavirus. Preclinical evidence is required to determine the activity of NS against coronavirus. If proven activity resulted from preclinical investigations, a clinical phase 1 trial of NS in patients with COVID-19 is suggested to explore its clinical activity.

## 4. Conclusions

This mini literature review documented the inhibitory effects of NS compounds against SARS-CoV-2 in several molecular docking studies. However, to our knowledge, there is no reported clinical trial of NS in human coronavirus cases. Therefore, we propose NS as a potential phytotherapy candidate in preclinical investigations followed by clinical investigations in the treatment of coronavirus diseases such as COVID-19. Also, further in-silico investigation on other natural products from traditional medicines is suggested to apply them in the treatment of COVID-19.

## 5. References


1. Fehr AR, Perlman S. Coronaviruses: An overview of their replication and pathogenesis. In: *Coronaviruses: Methods and Protocols*. Vol 1282. Springer New York; 2015:1-23. doi:10.1007/978-1-4939-2438-7_1

2. World Health Organization. Coronavirus. https://www.who.int/health-topics/coronavirus#tab=tab_1. Published 2020. Accessed May 12, 2020.

3. Shereen MA, Khan S, Kazmi A, Bashir N, Siddique R. COVID-19 infection: Origin, transmission, and characteristics of human coronaviruses. *J Adv Res*. 2020;24:91-98. doi:10.1016/j.jare.2020.03.005

4. Lin LT, Hsu WC, Lin CC. Antiviral natural products and herbal medicines. *J Tradit Complement Med*. 2014;4(1):24-35. doi:10.4103/2225-4110.124335

5. Molla S, Abul M, Azad K, et al. A REVIEW ON ANTIVIRAL EFFECTS OF NIGELLA SATIVA L. *pharmacologyonline*. 2019;2:47-53. http://pharmacologyonline.silae.it. Accessed March 18, 2020.

6. Ahmad A, Husain A, Mujeeb M, et al. A review on therapeutic potential of Nigella sativa: A miracle herb. *Asian Pac J Trop Biomed*. 2013;3(5):337-352. doi:10.1016/S2221-1691(13)60075-1

7. Tavakkoli A, Mahdian V, Razavi BM, Hosseinzadeh H. Review on clinical trials of black seed (Nigella sativa ) and its active constituent, thymoquinone. *J Pharmacopuncture*. 2017;20(3):179-193. doi:10.3831/KPI.2017.20.021

8. Koshak A, Wei L, Koshak E, et al. Nigella sativa Supplementation Improves Asthma

Control and Biomarkers: A Randomized, Double-Blind, Placebo-Controlled Trial. *Phyther Res*. 2017;31(3). doi:10.1002/ptr.5761

9. He T, Xu X. The influency of Nigella sativa for asthma control: A meta-analysis. *Am J Emerg Med*. November 2019. doi:10.1016/j.ajem.2019.11.036

10. Sahebkar A, Soranna D, Liu X, et al. A systematic review and meta-analysis of randomized controlled trials investigating the effects of supplementation with Nigella sativa (black seed) on blood pressure. *J Hypertens*. 2016;34(11):2127-2135. doi:10.1097/HJH.0000000000001049

11. Namazi N, Larijani B, Ayati MH, Abdollahi M. The effects of Nigella sativa L. on obesity: A systematic review and meta-analysis. *J Ethnopharmacol*. 2018;219:173-181. doi:10.1016/j.jep.2018.03.001

12. Daryabeygi-Khotbehsara R, Golzarand M, Ghaffari MP, Djafarian K. Nigella sativa improves glucose homeostasis and serum lipids in type 2 diabetes: A systematic review and meta-analysis. *Complement Ther Med*. 2017;35:6-13. doi:10.1016/j.ctim.2017.08.016

13. Sahebkar A, Beccuti G, Simental-Mendía LE, Nobili V, Bo S. Nigella sativa (black seed) effects on plasma lipid concentrations in humans: A systematic review and meta-analysis of randomized placebo-controlled trials. *Pharmacol Res*. 2016;106:37-50. doi:10.1016/j.phrs.2016.02.008

14. Therapeutic Research Center. the Natural Medicines Research Collaboration. https://naturalmedicines.therapeuticresearch.com/. Published 2020. Accessed April 14, 2020.

15. Salem ML, Hossain MS. Protective effect of black seed oil from Nigella sativa against murine cytomegalovirus infection. *Int J Immunopharmacol*. 2000;22(9):729-740. doi:10.1016/S0192-0561(00)00036-9

16. Barakat AB, Shoman SA, Dina N, Alfarouk OR. Antiviral activity and mode of action of Dianthus caryophyllus L. and Lupinus termes L. seed extracts against in vitro herpes simplex and hepatitis A viruses infection. *J Microbiol Antimicrob*. 2010;2(3):23-29. http://www.academicjournals.org/JMA. Accessed April 24, 2020.

17. Ulasli M, Gurses SA, Bayraktar R, et al. The effects of Nigella sativa (Ns), Anthemis hyalina (Ah) and Citrus sinensis (Cs) extracts on the replication of coronavirus and the expression of TRP genes family. *Mol Biol Rep*. 2014;41(3):1703-1711. doi:10.1007/s11033-014-3019-7

18. Oyero OG, Toyama M, Mitsuhiro N, et al. Selective inhibition of hepatitis c virus replication by alpha-zam, a nigella sativa seed formulation. *African J Tradit Complement Altern Med*. 2016;13(6):144-148. doi:10.21010/ajtcam.v13i6.20

19. Dorra N, El-Berrawy M, Sallam S, Mahmoud R. Evaluation of Antiviral and Antioxidant Activity of Selected Herbal Extracts. *J High Inst Public Heal*. 2019;49(1):36-40. doi:10.21608/jhiph.2019.29464

20. Barakat EMF, El Wakeel LM, Hagag RS. Effects of Nigella sativa on outcome of hepatitis C in Egypt. *World J Gastroenterol*. 2013;19(16):2529-2536. doi:10.3748/wjg.v19.i16.2529

21. Onifade AA, Jewell AP, Adedeji WA. Nigella sativa concoction induced sustained seroreversion in HIV patient. *Afr J Tradit Complement Altern Med*. 2013;10(5):332-335.

22. Onifade AA, Jewell AP, Ajadi TA, Rahamon SK, Ogunrin OO. Effectiveness of a herbal remedy in six HIV patients in Nigeria. *J Herb Med*. 2013;3(3):99-103. doi:10.1016/j.hermed.2013.04.006

23. Romano JD, Tatonetti NP. Informatics and computational methods in natural product drug discovery: A review and perspectives. *Front Genet*. 2019;10(APR):368. doi:10.3389/fgene.2019.00368

24. Bouchentouf S, Noureddine M. Identification of Compounds from Nigella Sativa as New Potential Inhibitors of 2019 Novel Coronasvirus (Covid-19): Molecular Docking Study Elucidation of neurodegenerative pathologies processes by molecular modeling View project Molecular operating enviro. ChemRxiv. doi:10.26434/chemrxiv.12055716.v1

25. Sekiou O, Ismail B, Zihad B, Abdelhak D. In-Silico Identification of Potent Inhibitors of COVID-19 Main Protease (Mpro) and Angiotensin Converting Enzyme 2 (ACE2) from Natural Products: Quercetin, Hispidulin, and Cirsimaritin Exhibited Better Potential Inhibition than Hydroxy-Chloroquine Against. *chemRxiv*. April 2020. doi:10.26434/chemrxiv.12181404.v1

26. Elfiky AA. Natural products may interfere with SARS-CoV-2 attachment to the host cell. *J Biomol Struct Dyn*. April 2020:1-16. doi:10.1080/07391102.2020.1761881

27. Sampangi-ramaiah MH, Vishwakarma R, Shaanker RU. Molecular docking analysis of selected natural products from plants for inhibition of SARS-CoV-2 main protease. *Curr Sci*. 2020;118(7):1087-1092. http://sts.bioe.uic.edu/castp/index.html?3igg. Accessed May 3, 2020.

28. Rajapaksa RMH, Perera BT, Nisansala MJ, Perera WPRT, Dissanayake KGC.

POTENTIAL OF INHIBITING THE RECEPTOR BINDING MECHANISM OF SARS-COV-2 USING PHYTOCHEMICAL EXTRACTS OF MEDICINAL HERB; MOLECULER DOCKING STUDY. *Glob J Eng Sci Res Manag*. 2020;7(4):51-61. doi:10.5281/zenodo.3766184

29. Silva JKR da, Figueiredo PLB, Byler KG, Setzer WN. Essential Oils as Antiviral Agents. Potential of Essential Oils to Treat SARS−CoV−2 Infection: An In−Silico Investigation. *Int J Mol Sci*. 2020;21(10):3426. doi:10.3390/ijms21103426

30. Maiti S, Banerjee A, Nazmeen A, Kanwar M, Das S. Active-site Molecular docking of Nigellidine to nucleocapsid / Nsp2 / Nsp3 / M Pro of COVID-19 and to human IL1R and TNFR1 / 2 may stop viral- growth / cytokine-flood , and the drug source Nigella sativa ( black cumin ) seeds show potent antioxidant role. Research Square. doi:10.21203/rs.3.rs-26464/v1

Table 1. A summary of effects of *Nigella sativa* compounds on SARS-CoV-2 targets.

| Reference | NS material | SARS−CoV−2 targets | Control | Effects |
|---|---|---|---|---|
| [16] | Thymoquinone | 6LU7 | NA | -Thymoquinone had a moderate binding affinity with 6LU7 |
| [17] | Nigellidine, α-Hederin | 6LU7, 2GTB | -Chloroquine<br>-HCQ<br>-Favipiravir | -Nigellidine and α-Hederin had the most binding affinity with 6LU7 and 2GTB<br>-Nigelledine was better than hydroxychloroquine and favipiravir<br>-α-Hederin better than chloroquine, hydroxychloroquine and favipiravir |
| [18] | Hederagenin | 6LU7, 6Y2E | Saquinavir | -Hederagenin had a high binding affinity with 6LU7 but less than Saquinavir and 6Y2E close to Saquinavir |
| [19] | Nigellidine | 6LU7, NSP2, 6vsb, QHD43415_3, QHD43423, IL1R, TNFR1, TNFR2 | NA | -Nigellidine had a high binding affinity with several SARS−CoV−2 and inflammatory molecular targets |
| [20] | Hederagenin | ACE2, GRP78 | NA | -Hederagenin had the highest binding affinity with ACE2 and GRP78 |
| [21] | Thymoquinone | 6LU7, ACE2 | HCQ | -Thymoquinone had a moderate binding affinity with 6LU7 and ACE2 1R42, but less than Hydroxy-Chloroquine |
| [22] | Thymoquinone | HSPA5 | NA | -Thymoquinone had a moderate binding affinity to HSPA5 |
| [23] | Thymohydro-quinone | 6LU7, Nsp15 / NendoU, ADRP, RdRp, rS, ACE2 | NA | -Thymohydroquinone had a moderate binding affinity with several SARS−CoV−2 molecular targets |

6LU7: main protease; 2GTB: main peptidase; ACE2: Angiotensin converting enzyme 2; HSPA5:Heat Shock Protein A5; HCQ: Hydroxychloroquine; Nsp15/NendoU: endoribonucleoase; ADRP: ADP−ribose−1″−phosphatase; RdRp: RNA−dependent RNA polymerase; rS: binding domain of SARS−CoV−2 spike protein; QHD43415_3: N-terminus-protenase; NSP2: Non-structural protein 2; 6vsb: Spike-glycoprotein; QHD43423: Nucleocapsid; IL1R: Interleukin 1 receptor; TNFR1: Tumor necrosis factor receptor 1; TNFR1: Tumor necrosis factor receptor 2.