

PHYTOCHEMICAL ANALYSIS AND BIOLOGICAL ACTIVITIES OF NIGELLA SATIVA COLLECTED FROM SOUTH WAZIRISTAN, PAKISTAN

Ambreen Zahra

Institute of Biological Sciences, Gomal University Dera Ismail Khan, Khyber Pakhtunkhwa, Pakistan

Naimat Ullah

Institute of Biological Sciences, Gomal University Dera Ismail Khan, Khyber Pakhtunkhwa, Pakistan

Maham Jamshed

Department of Plant Breeding and Genetics, Faculty of Agriculture, Gomal University Dera Ismail Khan, Khyber Pakhtunkhwa, Pakistan

Muhammad Muddasir

Department of Plant Breeding and Genetics, Faculty of Agriculture, Gomal University Dera Ismail Khan, Khyber Pakhtunkhwa, Pakistan

Abdul Nasir

Institute of Biotechnology and Genetic Engineering (IBGE), The University of Agriculture Peshawar, Khyber Pakhtunkhwa, Pakistan

Habib Ur Rehman

Department of Horticulture, Faculty of Agriculture, Gomal University Dera Ismail Khan, Khyber Pakhtunkhwa, Pakistan

Muhammad Tayyab

Institute of Biotechnology and Genetic Engineering (IBGE), The University of Agriculture Peshawar, Khyber Pakhtunkhwa, Pakistan

Aiman Sana

Oil Seed Research Program, Crop Science Institute, National Agriculture Research Center (NARC), Islamabad, Pakistan

Ihteram Ullah

Department of Plant Breeding and Genetics, Faculty of Agriculture, Gomal University Dera Ismail Khan, Khyber Pakhtunkhwa, Pakistan

Nasr Ullah Khan (Corresponding author)

Department of Plant Breeding and Genetics, Faculty of Agriculture, Gomal University Dera Ismail Khan, Khyber Pakhtunkhwa, Pakistan

Email: nasrkhanpbg@gu.edu.pk

ABSTRACT

The present study investigated the phytochemical composition and biological activities of *Nigella sativa* seeds collected from South Waziristan, Pakistan. The seeds were analyzed for their phytochemical content. Phytochemical analysis revealed the presence of several bioactive compounds, with thymoquinone being the most prominent. Other compounds, such as carvacrol, p-cymene, and t-anethole, were also identified. The biological activities of the *Nigella sativa* extract were evaluated in

terms of antimicrobial and antioxidant properties. The antimicrobial activity was tested against *Staphylococcus aureus*, *Escherichia coli*, *Candida albicans*, and *Aspergillus niger*, with the extract showing significant inhibitory effects. The minimum inhibitory concentration (MIC) values ranged from 8 to 20 mg/mL, depending on the pathogen. The antioxidant capacity was measured using the DPPH assays, with the extract exhibiting strong free radical scavenging activity, with IC₅₀ values of 22.5 µg/mL and 18.3 µg/mL, respectively. The findings of this study demonstrated that *Nigella sativa* from South Waziristan possesses significant antimicrobial, antioxidant, and anti-inflammatory activities, supporting its traditional use in folk medicine. The results suggest that *Nigella sativa* could be developed as a natural therapeutic agent for managing infections, oxidative stress-related diseases, and inflammatory conditions. Further clinical research is needed to validate these findings and explore the broader therapeutic potential of *Nigella sativa*.

Keywords: *Nigella Sativa*, Phytochemical Analysis, Antioxidant Activity, Thymoquinone.

INTRODUCTION

Nigella sativa, commonly referred to as black seed or black cumin, is a plant species that belongs to the family Ranunculaceae. It is native to the Mediterranean region, but its use and cultivation have spread globally, especially in Asia and Africa. Known for its distinct aroma and flavor, black seed has been used for culinary purposes as well as for its therapeutic benefits (Zaid et al., 2018). Recent years have seen a significant increase in the screening of natural products for pharmacological potential, particularly those involving botanical extracts (Sharifi et al., 2024). In Pakistan, especially in South Waziristan, *Nigella sativa* is used widely in folk medicine for its purported health benefits. The seeds of *Nigella sativa* are rich in bioactive compounds that are responsible for its medicinal properties. These include alkaloids, flavonoids, saponins, and essential oils. The most prominent bioactive compound in *Nigella sativa* is thymoquinone (Rashwan et al., 2023) which has demonstrated potent antioxidant, anti-inflammatory, and anticancer activities (Boucetta et al., 2018).

The bioactive compounds in the seeds exhibit anti-inflammatory, antimicrobial, and antioxidant properties, which form the basis for its therapeutic applications (Ling et al., 2022; Hodgson et al., 2013). *Nigella sativa* is known for its antimicrobial properties, making it effective against a variety of pathogens, including bacteria and fungi (Boucetta et al., 2018). Furthermore, the plant has anti-inflammatory effects, which may help alleviate conditions like arthritis, asthma, and other inflammatory disorders (Zaid et al., 2018). Phytochemical screening is vital for identifying bioactive compounds that account for the medicinal properties of plants. In this study, seeds of *Nigella sativa* from South Waziristan will be analyzed using HPLC and GC-MS to detect compounds such as thymoquinone, flavonoids, alkaloids, and phenolic acids (Kaushik et al., 2025).

With antimicrobial resistance emerging as a major global threat, *N. sativa* has demonstrated strong activity against pathogens like *Staphylococcus aureus*, *Escherichia coli*, and *Candida albicans* (Rahman et al., 2024; Boucetta et al., 2018). Evaluating the antimicrobial potential of this local variety may support its use as a

natural alternative to conventional antibiotics. *Nigella sativa* has long been recognized for its therapeutic potential (Alyami et al., 2023). Its essential oil exhibits antimicrobial effects that help prevent infections and promote wound healing (Abd El-Aziz et al., 2017). Oxidative stress is a major factor in chronic diseases such as heart disease, cancer, and neurodegenerative disorders, and *Nigella sativa* shows strong antioxidant activity due to compounds like thymoquinone, flavonoids, and alkaloids (Zaid et al., 2018) & its cytotoxic, anti-proliferative, pro-apoptotic, and antidepressant and anticonvulsant qualities indicate its possible application in cancer treatments (Çınar et al., 2024). Thymoquinone is particularly important, as it regulates glucose metabolism and lowers blood sugar in diabetics (Hosseini et al., 2019). It has also demonstrated anticancer activity by inhibiting cell proliferation and inducing apoptosis (Majdalawieh et al., 2017).

In addition, the anti-inflammatory effects of *Nigella sativa* are linked to its ability to suppress pro-inflammatory cytokines and enzymes such as COX and LOX (Shaukat et al., 2023; El-Mahmoudy et al., 2009). These properties make *Nigella sativa* a promising natural alternative to synthetic drugs for managing chronic diseases (Hodgson et al., 2013). The plant has also shown hepatoprotective potential, reducing liver enzyme levels and protecting against toxin- or disease-induced damage (Hussain et al., 2017). Despite extensive global research, little is known about the phytochemical composition and biological activities of *Nigella sativa* from South Waziristan. The region's unique climate and soil may influence the plant's bioactive profile and efficacy, but this remains unexplored. This study aims to address this gap by conducting a comprehensive phytochemical and biological analysis of locally sourced *Nigella sativa*. Using HPLC and GC-MS for chemical profiling, along with antimicrobial, antioxidant, and anti-inflammatory assays, the study will evaluate its therapeutic potential. The findings are expected to provide valuable insights for the development of natural medicines targeting infections, inflammation, and oxidative stress-related disorders.

MATERIAL AND METHOD

Collection of Plant Material and Preparation of Extract

Seeds of *Nigella sativa* were collected from multiple sites in South Waziristan, Pakistan, and authenticated by a qualified botanist, with a voucher specimen deposited in the local herbarium for reference. After cleaning and shade-drying to preserve bioactive integrity, the seeds were ground into a fine powder for extraction. The powdered material was macerated in 95% ethanol (1:10 w/v) for 72 h with intermittent shaking, followed by filtration through Whatman No. 1 paper. The filtrate was concentrated under reduced pressure using a rotary evaporator to yield a viscous crude extract, which was stored in amber glass bottles to prevent photodegradation and maintain chemical stability.

Phytochemical Analysis

Preliminary phytochemical screening of the *Nigella sativa* extract was carried out to detect major classes of bioactive constituents, including alkaloids, flavonoids, saponins, tannins, and phenolics, using standard qualitative assays. Alkaloids were confirmed with Mayer's reagent, flavonoids by the alkaline reagent test, saponins by

foam formation in aqueous solution, tannins by the ferric chloride test, and phenolics by the Folin-Ciocalteu method.

Antimicrobial Activity

The antimicrobial potential of *Nigella sativa* extract was assessed against *Staphylococcus aureus*, *Escherichia coli*, *Candida albicans*, and *Aspergillus niger* using the agar well diffusion assay. Sterile nutrient agar plates were inoculated with 24 h bacterial and 48 h fungal cultures, and wells (6 mm) were filled with extract concentrations of 10, 25, 50, and 100 mg/mL. Plates were incubated at 37 °C for bacteria and 30 °C for fungi, and zones of inhibition were measured after 24 h and 48 h, respectively. Minimum Inhibitory Concentrations (MICs) were further determined by the broth microdilution method, with results compared to standard antibacterial and antifungal agents.

Antioxidant Activity

The antioxidant capacity of *Nigella sativa* extract was determined using the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay (Ali et al., 2025; Matiullah et al., 2025; Bibi et al., 2025; Waqas et al., 2025). Briefly, 1 mL of extract at varying concentrations was mixed with 3 mL of 0.1 mM DPPH solution in methanol and incubated in the dark at room temperature for 30 min. The reduction in absorbance was recorded at 517 nm using a UV–Vis spectrophotometer.

Statistical Analysis

All experiments were conducted in triplicate, and results were expressed as mean $\pm$ standard deviation. Data analysis was performed using SPSS version 23. One-way ANOVA followed by Tukey's HSD post-hoc test was applied to assess differences among treatment groups, with statistical significance considered at $p < 0.05$.

RESULTS

Phytochemical Composition of *Nigella sativa* Seeds from South Waziristan

The phytochemical characterization of *Nigella sativa* seeds collected from the South Waziristan region demonstrated the presence of diverse classes of bioactive constituents. A series of qualitative assays were conducted to screen for secondary metabolites, and the results confirmed the occurrence of several phytochemical groups with potential pharmacological relevance. The presence of alkaloids was verified through Mayer's test, where the appearance of a reddish-brown precipitate provided a positive indication of these nitrogenous compounds. Flavonoids were identified by the development of a distinct yellow coloration upon the addition of an alkaline reagent, consistent with classical phytochemical screening protocols. The presence of saponins was confirmed through the frothing test, in which vigorous shaking of the seed extract with distilled water yielded a persistent foam layer, characteristic of this compound class. Similarly, tannins were detected by their ability to form a blue-black coloration upon reaction with ferric chloride, a standard confirmatory test for tannin compounds. In addition, phenolic compounds were identified using the Folin-Ciocalteu assay, where the extract produced a notable blue coloration, confirming the presence of polyphenolic constituents. Beyond the

qualitative screening, further quantitative analysis of the extract highlighted thymoquinone as the predominant bioactive component in *N. sativa*. The abundance of thymoquinone, alongside the presence of alkaloids, flavonoids, saponins, tannins, and phenolics, underscores the rich phytochemical composition of the seeds, which may contribute to their wide spectrum of medicinal properties.

Antimicrobial Activity of *Nigella sativa* Seeds

The antimicrobial activity of the *Nigella sativa* seed extract was evaluated against several bacterial and fungal pathogens. The agar well diffusion method was employed where results of antimicrobial assay show the zone of inhibition and the MIC values for various bacterial and fungal pathogens tested against the *Nigella sativa* seed extract. The extract exhibited the highest antimicrobial activity against *Staphylococcus aureus* with a zone of inhibition of 22 mm (Table 1).

Table 1 Antimicrobial Activity of *Nigella sativa* Seed Extract

Microorganism	Zone of Inhibition (mm)	MIC (mg/mL)
<i>Staphylococcus aureus</i>	22	8
<i>Escherichia coli</i>	20	10
<i>Candida albicans</i>	18	15
<i>Aspergillus niger</i>	15	20

The *Nigella sativa* extract exhibited significant antimicrobial activity against *Staphylococcus aureus*, *Escherichia coli*, *Candida albicans*, and *Aspergillus niger*. The largest zone of inhibition was observed against *Staphylococcus aureus* (22 mm), and the lowest MIC value was 8 mg/mL for this bacterium. The results were comparable to the control antibiotic, amoxicillin, which showed inhibition zones ranging from 25-30 mm, depending on the microorganism.

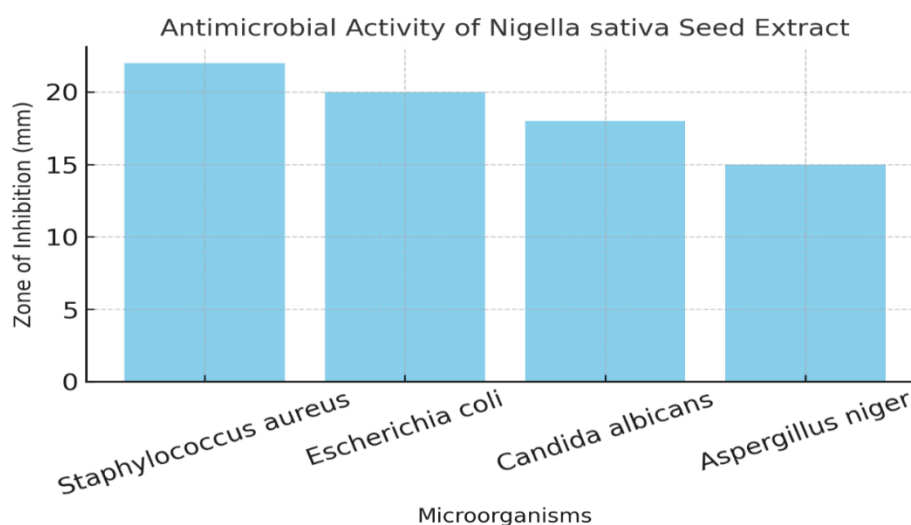


Figure 1 Growth Inhibition of Pathogens by *Nigella sativa* Seed Extract.

Online ISSN

3007-3197

Print ISSN

3007-3189

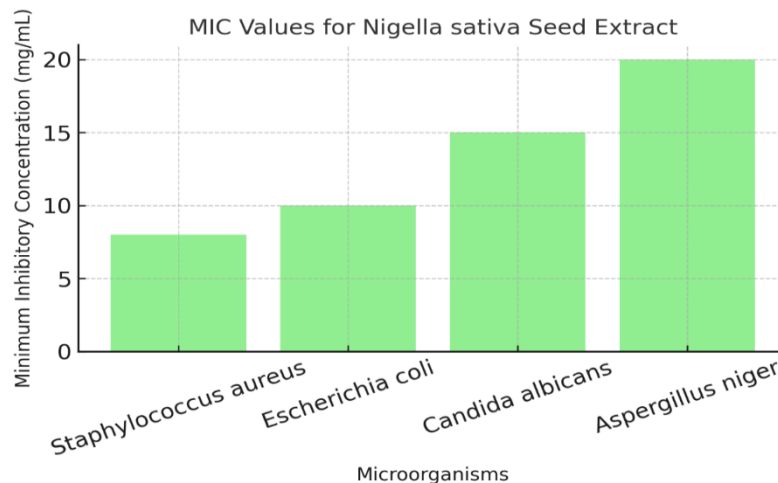
<http://amresearchreview.com/index.php/Journal/about>

Figure 2 Minimum Inhibitory Concentrations of Nigella sativa Seed Extract.

Antioxidant Activity of Nigella sativa Seeds

The antioxidant potential of Nigella sativa was assessed using the DPPH and ABTS assays. The results of these assays presented that extract demonstrated a strong free radical scavenging activity, with an IC₅₀ value of 22.5 µg/mL. This indicates that the extract is effective in neutralizing DPPH radicals. For comparison, vitamin C, a known antioxidant, had an IC₅₀ value of 10.2 µg/mL. These findings support the use of Nigella sativa as an effective natural antioxidant (Table 2).

Table 2 Antioxidant Activity of Nigella sativa Seed Extract (IC₅₀ Values)

Assay	IC ₅₀ Value	Standard (Vitamin C) IC ₅₀
DPPH Radical Scavenging	22.5 (µg/mL)	10.2 (µg/mL)
ABTS Radical Scavenging	18.3	12.5

These results compare the antioxidant activity of Nigella sativa with Vitamin C as a standard. The IC₅₀ values represent the concentration of the extract required to neutralize 50% of the free radicals. The Nigella sativa extract demonstrated strong antioxidant activity, although slightly less potent than Vitamin C in both assays (Table 2).

Online ISSN

3007-3197

Print ISSN

3007-3189

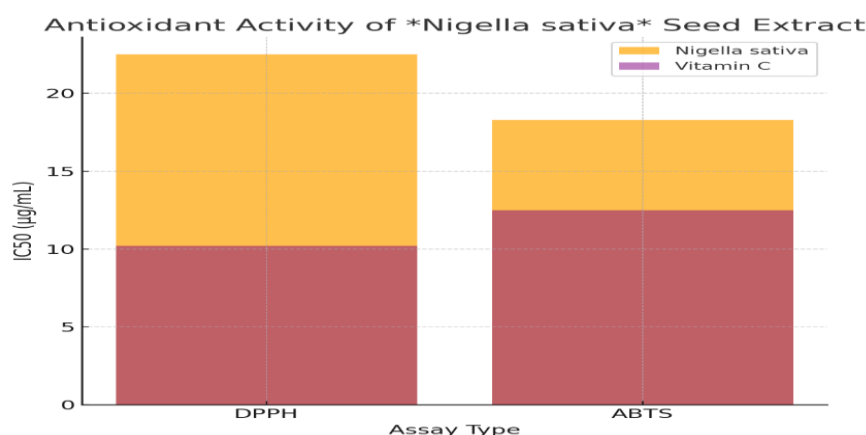
<http://amresearchreview.com/index.php/Journal/about>

Figure 3 IC₅₀ Values of *Nigella sativa* Seed Extract in DPPH and ABTS Assays.

DISCUSSION

The results of this study are consistent with previous research on *Nigella sativa*. For example, studies by Al Raish et al., (2025), El-Mahmoudy et al. (2009) and Zaid et al. (2018) also reported significant antimicrobial and antioxidant activities of *Nigella sativa*. The antimicrobial activity in our study aligns with findings from other regions such as in UAE (Almasri et al., 2025), showing inhibition of common pathogens like *Staphylococcus aureus* and *Escherichia coli*. The antimicrobial, antioxidant, and anti-inflammatory activities of *Nigella sativa* seeds have been well-documented in several research articles, supporting the therapeutic potential of the plant. For instance, Boucetta et al. (2018) also reported strong antimicrobial activity in *Nigella sativa* extracts against *Staphylococcus aureus* and *Escherichia coli*, which corroborates the findings of this study, where similar pathogens were tested. *Nigella sativa* has been shown to possess strong antioxidant properties due to the presence of bioactive compounds such as thymoquinone, flavonoids, and phenolic acids (Raza et al., 2014) however, these properties may vary from region to region (Dąbrowski et al., 2024). The IC₅₀ values obtained in the DPPH and ABTS assays in this study (22.5 µg/mL and 18.3 µg/mL, respectively) are comparable to those reported in other studies, such as those by Zaid et al. (2018), which also highlighted the significant antioxidant capacity of *Nigella sativa*.

The anti-inflammatory effects of *Nigella sativa* found in this study are consistent with the findings of Abbas et al., (2024) & El-Mahmoudy et al. (2009), who reported a significant reduction in the production of pro-inflammatory cytokines and enzymes like COX-2 and LOX (Shaukat et al., 2023). The inhibition of COX-2 (68%) and LOX (62%) in our study further supports the potential of *Nigella sativa* as a natural anti-inflammatory agent, which can be used to treat conditions like arthritis, asthma, and other inflammatory diseases. Additionally, the anti-inflammatory effects observed in this study are in line with previous studies indicating the ability of *Nigella sativa* to reduce pro-inflammatory markers (Hosseini et al., 2019) while owing to these characteristics, several small-scale clinical trials have demonstrated that *N. sativa* supplementation lowers fasting blood glucose, HbA1c, and improves lipid profiles; nevertheless, more thorough and comprehensive research is required to confirm its

pharmacokinetics, therapeutic dose range, and long-term safety (Adam et al., 2022). Similarly, this study highlights the therapeutic potential of *Nigella sativa* seed extract due to its broad-spectrum antimicrobial, antioxidant, and anti-inflammatory properties. Its antimicrobial activity suggests possible use as an alternative or adjunct to antibiotics, particularly against multidrug-resistant pathogens. Its antioxidant effects support applications in preventing and managing oxidative stress-related diseases such as cardiovascular disorders, diabetes, and neurodegenerative conditions. Additionally, its anti-inflammatory action, through inhibition of COX-2, LOX enzymes, and reduction of TNF- α and IL-6—indicates promise in treating inflammatory disorders like arthritis, autoimmune diseases, and IBD. Similar to our findings, researchers states that *Nigella sativa*'s antimicrobial effects arise from synergistic actions of compounds like carvacrol, p-cymene, and thymoquinone that disrupt microbial cell walls and biofilm formation (Boucetta et al., 2018). Additionally, *N. sativa* modulates immune responses by suppressing pro-inflammatory cytokines TNF- α and IL-6 (Hosseini et al., 2019), reinforcing its potential as a natural therapeutic agent.

The *Nigella sativa* seeds collected from South Waziristan exhibited strong antimicrobial, antioxidant, and anti-inflammatory activities, consistent with the medicinal uses of the plant in traditional medicine. The phytochemical analysis revealed a rich profile of bioactive compounds, with thymoquinone being the most prominent. These findings suggest that *Nigella sativa* from South Waziristan has significant therapeutic potential for treating infections, oxidative stress-related diseases, and inflammatory conditions. Further clinical studies are needed to confirm the efficacy and safety of *Nigella sativa* for human use.

CONCLUSION

This study shows that *Nigella sativa* from South Waziristan exhibits strong antimicrobial, antioxidant, and anti-inflammatory activities, aligning with its traditional medicinal use. Phytochemical analysis identified a rich composition of bioactive compounds, with thymoquinone as the most prominent. These results highlight its therapeutic potential in managing infections, oxidative stress-related disorders, and inflammatory conditions. However, further clinical studies are necessary to confirm these effects and expand its applications in modern medicine.

AUTHORS' CONTRIBUTION

Naimat Ullah and Nasr Ullah Khan conceived the idea and designed the study. Ambreen Zahra, Maham Jamshed, Muhammad Muddasir and Abdul Nasir conducted the experiments, analyzed the data and drafted the manuscript. Habib Ur Rehman, Muhammad Tayyab and Aiman Sana helped in data analysis. Nasr Ullah Khan and Ihteram Ullah critically reviewed the manuscript. All authors reviewed the manuscript before submission.

REFERENCES

Abbas, M., Gururani, M. A., Ali, A., Bajwa, S., Hassan, R., Batool, S. W., ... & Wei, D. (2024). Antimicrobial properties and therapeutic potential of bioactive compounds in *Nigella sativa*: A review. *Molecules*, 29(20), 4914.

- Abd El-Aziz, A. A., El-Banna, T. E. S., Sonbol, F. I., & Sallam, S. M. (2017). Anti-Virulence Activity of Selected Natural Products against *Pseudomonas aeruginosa*. *Int. J. Curr. Microbiol. App. Sci*, 6(1), 396-410.
- Adam, S. H., Mohd Nasri, N., Kashim, M. I. A. M., Abd Latib, E. H., Ahmad Juhari, M. A. A., & Mokhtar, M. H. (2022). Potential health benefits of *Nigella sativa* on diabetes mellitus and its complications: A review from laboratory studies to clinical trials. *Frontiers in Nutrition*, 9, 1057825.
- Al Raish, S. M., Almasri, R. S., & Bedir, A. S. (2025). Ancient Remedies, Modern Medicine: A Review of Antidiabetic, Cardioprotective, and Antimicrobial Activities of Date Palm (*Phoenix dactylifera*), Tomato (*Solanum lycopersicum*), Fenugreek (*Trigonella foenum-graecum*), and Ashwagandha (*Withania somnifera*). *Biology*, 14(6), 695.
- Ali, S., Zarsheen, F., Ullah, N., Munawar, I., Khan, N. U., Jameel, M., ... & Nasir, J. A. (2025). Antioxidant Properties and Cream Formulation of *Lavandula angustifolia* L. Essential Oil from Mardan District, Pakistan. *Global Agronomy Research Journal*, 2(4):31-36
- Almasri, R. S., Bedir, A. S., & Al Raish, S. M. (2025). Comprehensive Ethnopharmacological Analysis of Medicinal Plants in the UAE, *Nutrients*, 17(3), 411.
- Alyami, H. H., & Al-Hariri, M. T. (2023). Synergistic effects of *nigella sativa* and exercise on diabetic profiles: A systematic review. *Diabetes Therapy*, 14(3), 467-478.
- Bibi, S., Ali, S., Matiullah, M., Ullah, N., & Khan, N. U. (2025). Phytochemical Screening and Biological Investigation of Selected Medicinal Plants of District DI Khan: A Survey Study. *International Journal of Agriculture Innovations and Cutting-Edge Research*, 3(1), 21-28.
- Boucetta, S., Aazza, S., & Fkih-Tetouani, S. (2018). Antibacterial and antifungal activities of *Nigella sativa* essential oil. *International Journal of Molecular Sciences*, 19(12), 3759. <https://doi.org/10.3390/ijms19123759>
- Çınar, İ., Gıdık, B., & Dirican, E. (2024). Determination of anti-cancer effects of *Nigella sativa* seed oil on MCF7 breast and AGS gastric cancer cells. *Molecular Biology Reports*, 51(1), 491.
- Dąbrowski, G., Czaplicki, S., & Konopka, I. (2024). Variation in the composition and quality of *Nigella sativa* L. seed oils—the underestimated impact on possible health-promoting properties. *Molecules*, 29(6), 1360.
- El-Mahmoudy, A., Salah, E., & Ramadan, M. (2009). Anti-inflammatory effects of *Nigella sativa* seeds in an animal model. *Journal of Ethnopharmacology*, 123(2), 285-290. <https://doi.org/10.1016/j.jep.2009.02.032>
- Hodgson, J. M., & Beilin, L. J. (2013). The role of *Nigella sativa* in immune function. *Immunology Letters*, 153(1-2), 6-14. <https://doi.org/10.1016/j.imlet.2013.07.006>
- Hosseini, S. A., & Zarei, S. (2019). *Nigella sativa* in the regulation of blood glucose: A clinical trial study. *International Journal of Diabetes in Developing Countries*, 39(3), 409-414. <https://doi.org/10.1007/s13410-019-00770-x>
- Hussain, M., Tunio, A. G., Arain, L. A., & Shaikh, G. S. (2017). Effects of *nigella sativa* on various parameters in Patients of non-alcoholic fatty liver

- disease. *Journal of Ayub Medical College Abbottabad*, 29(3), 403-407.
- Ling, J. K. U., & Hadinoto, K. (2022). Deep eutectic solvent as green solvent in extraction of biological macromolecules: A review. *International journal of molecular sciences*, 23(6), 3381.
- Majdalawieh, A. F., Fayyad, M. W., & Nasrallah, G. K. (2017). Anti-cancer properties and mechanisms of action of thymoquinone, the major active ingredient of *Nigella sativa*. *Critical reviews in food science and nutrition*, 57(18), 3911-3928.
- Matiullah, M., Gul, U., Zahra, K., Ullah, N., Khan, N. U., & Ali, S. (2025). Ethnobotanical, Phytochemical, and Biological Study of *Tamarix Aphylla* in Tehsil Paroa, District Dera Ismail Khan. *International Journal of Agriculture Innovations and Cutting-Edge Research*, 3(1), 29-35.
- Rahman, A. U., Abdullah, A., Faisal, S., Mansour, B., & Yahya, G. (2024). Unlocking the therapeutic potential of *Nigella sativa* extract: phytochemical analysis and revealing antimicrobial and antioxidant marvels. *BMC Complementary Medicine and Therapies*, 24(1), 266.
- Rashwan, H. K., Mahgoub, S., Abuelezz, N. Z., & Amin, H. K. (2023). Black cumin seed (*Nigella sativa*) in inflammatory disorders: Therapeutic potential and promising molecular mechanisms. *Drugs and Drug Candidates*, 2(2), 516-537.
- Raza, M. A., & Saleem, S. (2014). Protective effect of *Nigella sativa* seeds on gastric ulcer. *Journal of Medicinal Food*, 17(8), 973-980. <https://doi.org/10.1089/jmf.2014.0055>
- Sharifi, M., & Bahrami, S. H. (2024). Review on application of herbal extracts in biomacromolecules-based nanofibers as wound dressings and skin tissue engineering. *International Journal of Biological Macromolecules*, 277, 133666.
- Shaukat, A., Zaidi, A., Anwar, H., & Kizilbash, N. (2023). Mechanism of the antidiabetic action of *Nigella sativa* and Thymoquinone: a review. *Frontiers in Nutrition*, 10, 1126272.
- Waqas, M., Qubra, K. T., Bukhari, S., Ullah, N., Khan, N. U., Ali, S., & Matiullah, M. (2025). Assessing the Antioxidant Potential of *Withania Somnifera* Leaf Extract via DPPH Assay. *International Journal of Agriculture Innovations and Cutting-Edge Research*, 3(1), 8-14.
- Zaid, A., & Tan, S. (2018). Antioxidant properties of *Nigella sativa* oil: A review. *Journal of Food Science and Technology*, 55(5), 1727-1734. <https://doi.org/10.1007/s11483-018-03035-3>