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Effectiveness of botanical extracts and essential oils for repressing leaf rust disease severity of wheat caused by Puccinia triticina Eriks *under field conditions*

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Abstract: *Leaf rust is a devastating fungal disease of wheat that threatens crop yields. Wheat (Triticum aestivum) is a staple crop worldwide and vital in Pakistan, but its productivity is threatened by leaf rust (Puccinia triticina) disease that can cause up to 50% yield loss. This study evaluated the efficacy of various botanical sprays against leaf rust on two wheat cultivars, Morocco (susceptible) and Urooj (resistant), under field conditions. Treatments included neem, garlic, tobacco, clove, castor oil, basil, cinnamon oil, moringa, and henna. All significantly reduced disease severity compared to the untreated control. On Morocco, garlic (9.54%), neem (9.06%), and tobacco (9.06%) were most effective, while basil, castor oil, cinnamon oil, and moringa also provided notable suppression. On Urooj, neem (6.31%), tobacco (5.58%), and garlic (5.47%) showed the highest efficacy, followed by clove (5.45%) and henna (5.31%). Across both cultivars, neem, garlic, and tobacco consistently reduced leaf rust severity, confirming their strong protective bioactivity. These findings highlight the potential of plant-derived sprays as eco-friendly alternatives for integrated management of wheat leaf rust.*

Keywords: *Wheat leaf rust, Puccinia triticina, Botanical extracts, Essential oils, Neem extract, Garlic extract, Tobacco extract, Clove extract, Henna extract, Moringa extract, Basil extract, Castor oil, Cinnamon oil*

1. Introduction

Plants are crucial contributors to a nation's agricultural yield. Pakistan, in particular, possesses exceptionally diverse and fertile soils. As a staple cereal crop in several Asian countries, including Pakistan, China, India, and Bangladesh, wheat (*Triticum aestivum*) is globally significant (Balfourier et al., 2019). The estimated annual global wheat production is between 750 and 770 million tons, cultivated across 220 million hectares, and accounting for 15% of daily calorie consumption (Balfourier et al., 2019). Local researchers and farmers face increasing pressure to provide food for the rapidly expanding human population (Badar et al., 2023).

Rust infections represent the most common and significant diseases threatening global wheat production (Duplessis et al., 2021; Jan et al., 2025). All three types of rust are known to reduce crop yield, with the severity being non-specific, depending on the stage of occurrence (Duplessis et al., 2021; Jan et al., 2025). The extent and type of infection are highly dependent on the wheat variety, with susceptible cultivars being most affected. Key factors contributing to disease acceleration include growing conditions, climatic variables, cultivation practices, and, most importantly, the selection of the wheat cultivar (Hassan et al., 2022; Jan et al., 2025).

Wheat is highly susceptible to three main rust diseases: leaf rust (*Puccinia triticina*), stem rust (*Puccinia graminis Pers.*), and stripe rust (*Puccinia striiformis var. striiformis W.*) (Cui et al., 2020). All three can be devastating, leading to yield reductions (Markell et al., 2001). Leaf rust of wheat is caused by *P. triticina*, a highly destructive pathogen originating in South Africa (Nxumalo, 2018). It thrives in conditions of high relative humidity, cool nights, and dew. Under these favorable conditions, the disease spreads rapidly, reducing yield by decreasing the kernel numbers per head and the kernel weight of infected plants (Nxumalo, 2018). *P. triticina* is currently considered the most critical pathogen in global wheat production, causing severe output losses across wide geographical regions (Melvin et al., 2008; Figueroa et al., 2018).

Rusts are among the most destructive diseases affecting other significant crops such as maize (*Zea mays L.*) and coffee (*Coffea arabica L.*), in addition to wheat (Cui et al., 2020; Jan et al., 2025). *P. triticina* is a biotroph parasite of wheat that begins its spread once conducive conditions, namely optimal temperature and humidity, become available. While it infects other crops, including *Hordeum vulgare*, *Secale cereale*, and *Avena sativa*, *Triticum aestivum* is the most negatively impacted. The disease attack is exacerbated by low temperatures (15-25°C), high humidity, wind, and the cultivation of susceptible varieties (Singh et al., 2002).

The leaf rust epidemic that started in 1978 means rusts pose a constant threat to wheat cultivation in Pakistan (Hussain et al., 1999). *Puccinia triticina* presents a serious danger to wheat yield in Pakistan, potentially reducing it by 10% to 50% more than stem or yellow rust (Hussain et al., 1999).

Wheat grain is a staple food, used to prepare flour for bread, pasta, biscuits, and other food products. Furthermore, wheat crop residue and straw are utilized as animal feed, such as hay, grain, and wheat bran. Stem rust is a major disease in extensive wheat cultivation. Yield loss from stem rust has been estimated at 50% in Egypt (Draz et al., 2015), and typically causes losses of 15% to 20% globally across various production zones (Figueroa et al., 2018). In the United States, it is the most important wheat disease, known to cause loss in both the quantity and quality of the plant (Leonard and Szabo, 2005). The pathogen responsible for stem rust annually evolves new virulent strains, many of which can overcome existing plant resistance mechanisms (Draz et al., 2015; Barro et al., 2017; Jan et al., 2025).

Traditionally, disease control relies on using resistant cultivars and chemical fungicides (Draz et al., 2015; Barro et al., 2017; Jan et al., 2025). However, the infestation of these diseases often results in agricultural yield and production loss, which is usually managed through the application of synthetic fungicides. The overuse of commercial fungicides carries several adverse effects, including the development of unexplained diseases and allergies in people consuming sprayed food, and most concerningly, it can lead to the crop developing single-quality resistance (Hassan et al., 2022). Furthermore, the environmental impact of fungicides is now much better understood than in the past.

Consequently, new strategies are being developed for stem rust control that minimize fungicide use. Plant extracts with antifungal activity offer an eco-friendly alternative (Gholamnezhad, 2019; Cowan, 1999; Jan et al., 2025). These botanical extracts and organic compounds act as eco-friendly control methods by directly affecting phytopathogens and indirectly stimulating the plant's defense mechanisms. This stimulation includes the production of secondary metabolites such as phenols, phenolic acids, quinones, flavones, flavonoids, flavonols, tannins, and coumarins (Gholamnezhad, 2019; Cowan, 1999; Jan et al., 2025). These compounds are effective against pathogens and play a vital role in plant defense (Das et al., 2010). The present study evaluated the effectiveness of plant extracts, applied as foliar sprays, in reducing wheat stem rust disease under natural field conditions.

2. Material and Method

Experimental Site and particulars

The Plant Pathology research area at the University of Agriculture in Faisalabad, Pakistan, was the location where the field experiments were carried out. As is typical for the Faisalabad district, the climate in this area is semi-arid, with scorching summers and moderate winters. The region is located in northwestern Pakistan. Wheat varieties Morocco (a screen test for wheat rust) and Urooj were contributed by Punjab Seed Corporation, Ayyub Agricultural Research Institute, Faisalabad, Pakistan. The experiment was organized in a randomized complete block design (RCBD) with plant-to-plant 4 inches and row to row 9 inches. There were 10 treatments applied: 7 plant extracts, 2 essential oils and 1 control. Treatments were divided in 3 blocks and every treatment was replicated 3 times in 3 blocks. Every treatment was applied to the crops through the foliar route by artificial inoculation of P. triticina at adult stage of the crop

Experimental plant material and extract preparation

Extraction of plants of plant species, i.e., leaves of henna (Lawsonia inermis), neem (Azadirachta indica), moringa (Moringa oleifera), Tobacco (Nicotiana tabacum), Basil (Ocimum basilicum) and Clove (Syzygium aromaticum) collected from UAF research station and High-quality essential oils of castor and cinnamon were procured from local suppliers were evaluated for their efficacy as inducer materials to inhibit leaf rust infection of wheat under Faisalabad field conditions. Fresh and healthy leaves Plant samples were washed with tap water to remove dust, then allowed to dry in fresh air for 4 days. Dried samples were kept in a refrigerator at 4 °C until use. 10 grams dried powder in 250 mL flask was blended with 100 ml sterile distilled water and oils were suspended in water for spraying by blending 9 mL of every oil with 100 mL of distilled water and agitated and shaken forcefully for 8 minutes at room temperature. The suspensions were filtered on Whatman No. 1 paper two additional times with the addition of fresh water. The combined filtrates (100 mL) were utilized as starting solution for the plants extracts, with a concentration of approximately 10 g/L.

Table.2.1 Plant extracts/essential oils used against wheat leaf rust and their main active components

Common Name	Scientific Name	Family	Major Active Components	Plant organ containing active component and its citation

Garlic	Allium sativum L.	Amaryllidaceae	Allicin, diallyl sulfides, ajoene	Bulb – https://doi.org/10.1021/ja01239a048
Neem	Azadirachta indica A. Juss.	Meliaceae	Azadirachtin, nimbin, salannin	Seeds & leaves – https://doi.org/10.1146/annurev.en.35.010190.001415
Tobacco	Nicotiana tabacum L.	Solanaceae	Nicotine, anabasine	Leaves – https://doi.org/10.1021/jf00108a047
Castor oil	Ricinus communis L.	Euphorbiaceae	Ricinoleic acid, flavonoids	Seeds (oil) – https://www.ajmsjournal.info/index.php/ajms/article/view/414
Cinnamon oil	Cinnamomum burmannii	Lauraceae	Cinnamic aldehyde, eugenol	Bark (oil) – https://doi.org/10.1021/jf0340874
Basil	Ocimum basilicum L.	Lamiaceae	Linalool, eugenol, methyl chavicol	Leaves (oil) – https://doi.org/10.1021/jf021038t
Moringa	Moringa oleifera Lam.	Moringaceae	Isothiocyanates, flavonoids,	Leaves & seeds – https://doi.org/10.1002/ptr.2023

			phenolic acids	
Henna	Lawsonia inermis L.	Lythraceae	Lawsone (2-hydroxy-1,4-naphthoquinone)	Leaves – https://www.ijpsdr.com/index.php/ijpsdr/article/view/203
Clove	Syzygium aromaticum	Myrtaceae	Eugenol, eugenyl acetate, β-caryophyllene	Flower buds (oil) – https://doi.org/10.1002/ptr.2124

Foliar application of plant extracts

Pre-treated experimental plant extracts were sprayed via the foliar mode on all the experimental plants, after 7 days of artificial inoculation of the crop. Foliar spray is evenly applied to the whole surface of the plant.

Preparation and Artificial inoculation of leaf rust disease-causing inoculum

For artificial inoculation of *T. aestivum, an* isolated inoculum of local and virulent strains of *P. triticina*. Urediniospores were collected from heavily infected wheat leaves obtained from the Ayyub Agricultural Research Institute in Faisalabad. Urediniospores were suspended in distilled water at a 1:1 ratio for inoculum preparation. Applying a light mist of distilled water to the plants prior to inoculation enhanced the adhesion of the spores. The inoculum was uniformly distributed on the leaves of all plants during the late afternoon or early evening, coinciding with the onset of dew formation. The inoculated plants were allowed to remain in the dew overnight to promote infection. Control plants were maintained under identical conditions, either remaining unexposed to the spores or having their leaves covered to prevent spore contact.

Assessment of disease infestation

Every plot was scored for leaf rust intensity seven days after inoculation (before treatment) and again seven days after treatment. The severity was decided by looking at how many leaves are covered with rust uredinia.

Table2.2. Disease rating Scale of leaf rust of wheat used in disease assessment

Disease reaction	Field Response	Scale	Severity (%)
0	Immune, No visible infection	0	0
R	Resistant, visible necrosis or chlorosis, no uredia are present	1	0-10
MR	Moderately Resistant, Small uredia surrounded by either necrotic or chlorosis area	2	11-30
MR-MS	Chlorotic or necrotic areas with medium to small sized uredia	3	31-40
MS	Moderately Susceptible, Medium sized with possibly distinct chlorosis but no necrotic margins	4	41-60
S	Susceptible, Large uredia with no necrosis and little or no chlorosis	5	>60

Efficacy (%) = Severity (%) of the control - Severity (%) of the treatment/Severity (%) of the control ×100

To do the assessment, the modified Cobb’s scale (0–100%) was used, following Peterson et al.’s (1948) diagrams. Each plot (or pot) had at least 3 plants rated and determined the average severity by calculating the mean. Only the number of diseases was documented; nothing else was measured about the crops or their growth.

The efficacy of a certain treatment was determined according to the following equation adopted by Rewal and Jhooty (1985):

$$\text{Efficacy \%} = \frac{C - T}{C} \times 100$$

where C = infection in the control and T = infection
in the treatment

Statistical analysis

Statistical results of disease Severity and activity of plant extracts against leaf rust disease were analyzed through SPSS software package using the LSD method. Results so obtained were again statistically cross-checked using the assistance of Statistic 8.1 for analysis of variance (ANOVA) with the minimum significant difference of 5%. Graphs were prepared using the assistance of GraphPad Prism.

3. Results

Effect of Plant Extracts and Essential Oils under Field Conditions

The efficacy of different plant extracts and essential oils in suppressing wheat leaf rust after 7 days of application is presented in **Table 3.1**. Significant variation ($p \leq 0.05$) was observed among treatments in both wheat cultivars, Morocco (susceptible) and Urooj (resistant), as confirmed by the LSD test (2.10 for Morocco and 2.14 for Urooj).

In the susceptible cultivar Morocco, garlic extract exhibited the highest efficacy (9.54%), followed closely by neem (9.06%) and tobacco (9.06%). Basil (8.74%), castor oil (8.83%), cinnamon oil (8.80%), and moringa (8.52%) also demonstrated strong suppressive effects, whereas the untreated control showed the lowest efficacy (2.22%). These findings indicate that bio extracts were able to reduce disease pressure significantly in Morocco despite its genetic susceptibility.

In the resistant cultivar Urooj, overall severity levels were lower, consistent with its inherent resistance. However, considerable differences among treatments were still evident. Neem extract again provided the maximum efficacy (6.31%), followed by tobacco (5.58%), garlic (5.47%), and clove (5.45%). Cinnamon oil (4.43%) and henna (5.31%) showed comparatively moderate suppression, while the control maintained the lowest efficacy (2.15%).

The comparative performance across both cultivars highlights the consistent effectiveness of neem, garlic, and tobacco in reducing wheat leaf rust severity. Their significant efficacy in both susceptible and resistant backgrounds underscore their potential as reliable, eco-friendly alternatives for integrated disease management.

Table 3.1 Efficacy of Plant Extracts and Essential Oils for suppressing wheat leaf rust after 7 days of treatments application under field conditions.

Wheat Cultivar	Treatments	Efficacy(%)
Morocco	Basil	8.74
	Castor Oil	8.83
	Cinnamon Oil	8.80
	Clove	8.28
	Control	2.22
	Garlic	9.54
	Henna	8.24
	Moringa	8.52
	Neem	9.063
	Tobacco	9.063
Urooj	Basil	5.21
	Castor Oil	5.33

	Cinnamon Oil	4.43
	Clove	5.45
	Control	2.15
	Garlic	5.47
	Henna	5.31
	Moringa	5.50
	Neem	6.31
	Tobacco	5.58

LSD (0.05): Morocco = 2.10; Urooj = 2.14

Means differing by more than LSD are significantly different at $P \leq 0.05$

A comparative analysis of disease reduction in two wheat varieties, Morocco (susceptible) and Urooj (resistant), is presented in **Figure 3.1**. Significant variability was observed among the tested plant extracts and essential oils, confirming their potential role in suppressing wheat leaf rust severity under field conditions.

In the susceptible cultivar Morocco, the highest mean disease reduction was recorded with garlic extract (9.54), followed closely by neem (9.06) and tobacco (9.06). These treatments markedly outperformed the untreated control (2.22), demonstrating their superior efficacy. Other treatments, such as basil (8.74), castor oil (8.83), moringa (8.52), and clove (8.28), also provided moderate reductions compared to control.

In contrast, the resistant variety Urooj exhibited relatively lower overall severity, reflecting its inherent resistance. Nevertheless, considerable differences among treatments were recorded. Neem extract showed the maximum reduction (6.31), followed by tobacco (5.78), garlic (5.47), and clove (5.45). Control plants maintained the lowest mean (2.15), while cinnamon oil (4.43) and henna (5.31) were comparatively less effective than other extracts.

The interaction between treatments and varietal response is clearly visualized in **Figure 1**, which illustrates that Morocco expressed higher severity values across all treatments compared to Urooj.

However, the consistent performance of garlic, neem, and tobacco in both cultivars indicates their broad-spectrum potential in suppressing *Puccinia triticina*.

Overall, the findings reinforce those foliar applications of selected bio extracts, particularly garlic and neem, can significantly reduce wheat leaf rust severity even in susceptible cultivars. These results highlight the promise of integrating plant-based extracts into eco-friendly and sustainable disease management strategies for wheat production.

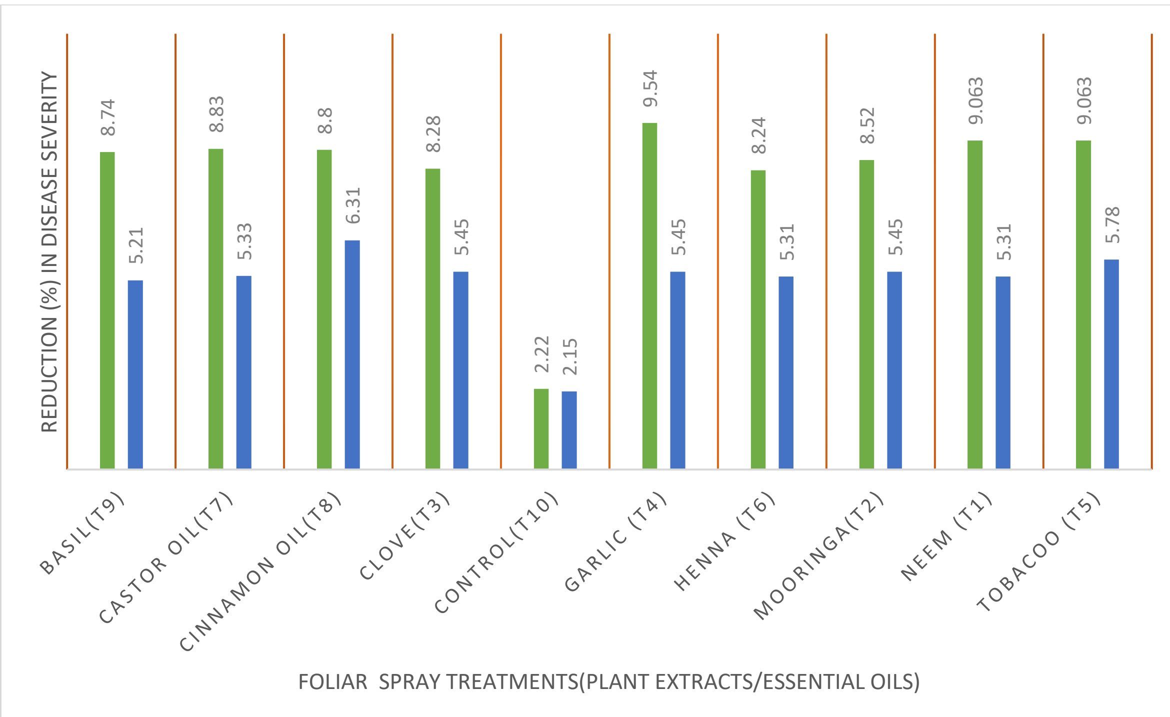

Figure 3.1. Comparative efficacy of plant extracts and essential oils in reducing wheat leaf rust severity in susceptible (Morocco) and resistant (Urooj) cultivars under field conditions. Data represent reduction in wheat leaf rust disease values, showing differential treatment responses across both varieties.

4. Discussion

The present study demonstrated that plant extracts and essential oils, particularly neem, garlic, and tobacco, significantly suppressed wheat leaf rust severity under field conditions in both susceptible (Morocco) and resistant (Urooj) cultivars. The superior efficacy of neem extract may be attributed to its rich composition of bioactive compounds such as azadirachtin, nimbin, and salannin, which possess well-documented antifungal properties that inhibit spore germination and hyphal growth of rust pathogens(Kharwar et al., 2020). Garlic extract also showed strong suppressive effects, likely due to the presence of allicin and other sulfur-containing compounds that disrupt fungal cell membranes and interfere with pathogen metabolism (Curtis et al., 2004). Similarly, the performance of tobacco extract could be linked to its alkaloid content, including nicotine, which has been reported to exert antifungal activity against a range of plant pathogens (Singh, 2013)

The higher overall efficacy values in Morocco compared to Urooj reflect the difference in varietal response, where Morocco's susceptibility provided greater scope for treatment-induced reductions, while Urooj's inherent resistance limited disease development regardless of treatment. Nevertheless, the consistent trends across both cultivars confirm that bio extracts act as complementary tools alongside host resistance.

These findings align with earlier studies highlighting the potential of botanical extracts and essential oils as eco-friendly alternatives to synthetic fungicides in cereal disease management (Shabana et al., 2017)Importantly, the demonstrated effectiveness under field conditions strengthens their applicability in integrated disease management (IDM) programs, offering farmers sustainable strategies to reduce reliance on chemical fungicides while mitigating the threat of fungicide resistance and environmental hazards.

5. Conclusion

The present study revealed that plant extracts and essential oils, particularly neem, garlic, and tobacco, effectively reduced wheat leaf rust severity under field conditions. Their efficacy was consistent in both susceptible (Morocco) and resistant (Urooj) cultivars, with neem extract showing the most pronounced suppressive effect. These findings highlight the potential of botanical bio-extracts as sustainable, eco-friendly alternatives to chemical fungicides in integrated wheat disease management. Future studies focusing on dose optimization, formulation stability, and synergistic effects among extracts may further enhance their practical application at the farm level.

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