Research Article

Pathogenicity and Management of *Xanthomonas citri* pv. *malvacearum,* the causal organism of Bacterial Blight of Cotton in Dera Ghazi Khan

Muhammad Ali[1], Saliha Nazar[1], Awais Hanif[1], Abdul Hannan[1], Sagheer Atta[1]*
[1]*Department of Plant Pathology, Ghazi University, Dera Ghazi Khan, Pakistan*
Correspondence: satta@gudgk.edu.pk

ABSTRACT

Bacterial blight of cotton, caused by *Xanthomonas citri* pv. *malvacearum* (Xcm) is a major threat to cotton production in Pakistan. The pathogenicity of Xcm was evaluated against different cotton varieties to assess the susceptibility of local cotton varieties against the bacterial pathogen. Furthermore, multiple management strategies were performed in the field conditions to find out the suitable management strategy against the bacterial blight in Dera Ghazi Khan. The bacterium was isolated from affected cotton leaves, which produced yellow, mucoid colonies on nutrient agar media, confirming its identity as *Xanthomonas* using physical features and Gram staining. Three cotton varieties (BS-313, FH-333, and FH-Lalazar) were tested for pathogenicity, and the results showed that all were susceptible. FH-Lalazar had the highest disease severity (70.91%), followed by BS-313 (61.49%) and FH-333 (58.04%). Neem (*Azadirachta indica*) and moringa (*Moringa oleifera*) extracts were applied at a 15% concentration as Botanical extracts, and both decreased the severity of disease in comparison to the untreated plants. Neem extract was more effective (lowering the disease severity of the variety FH-333 to 49.22%) as compared to moringa extract. Kasugamycin and copper oxychloride decreased the severity to 10.45% in FH-333, demonstrating the superiority of chemical control. Tebuconazole and trifloxystrobin also reduced the disease severity, but not as much as kasugamycin. Statistical analysis (ANOVA and LSD test, $p < 0.05$) confirmed that the differences among treatments were significant, with chemical treatments showing the highest efficacy in reducing disease severity. These findings indicate that none of the tested cultivars exhibited resistance, and botanical extracts showed promise as environmentally friendly substitutes, although chemical treatments remain the most dependable option. For the sustainable management of bacterial blight in cotton under local conditions, an integrated strategy combining plant extracts, resistant varieties, and selective chemical use is advised.

Keywords: Cotton, bacterial blight, *Xanthomonas citri* pv. *malvacearum*, neem extract, moringa extract, kasugamycin, copper oxychloride, tebuconazole.

INTRODUCTION

Cotton (*Gossypium hirsutum* L.) is a flowering plant that belongs to the genus *Gossypium* of the family *Malvaceae*. It is also often called the "White gold of Pakistan" and the "King of fibers." Cotton crops thrive in tropical and subtropical regions in both the Old and New Worlds (Yadav et al., 2024). Approximately 26% of farmers grow cotton, and it occupies more than 15% of Pakistan's entire cultivated area (CGIAR, 2020).

In Pakistan, cotton covered approximately 2.4 million hectares in 2023/24, giving 7.3 million bales (≈1.6 million metric tons) with a yield of 665 kg/ha. Cotton is expected to cover 2.0 million hectares in 2024/25, yielding 5.2 million bales (≈1.13 million metric tons) and a yield of 621 kg/ha (USDA, 2025). Besides its status as a plant species, cotton is also one of the largest crops in global agriculture. Cotton, which is a leading traded natural fiber, earns a livelihood for more than 100 million rural families worldwide, particularly in developing countries (ICAC, 2023).

37.4% of Pakistan's workforce is employed in the agriculture sector, which contributes 24% to the country's GDP (Economic Survey of Pakistan 2023–24). Cotton is thus an important contributor to the livelihoods of the rural areas as well as the stability of the national economy, alongside being a necessary export commodity.

Article History
Received: September 9, 2025, Accepted: November 03, 2025, Published: December 2, 2025.

Cotton bacterial blight caused by the bacterium *Xanthomonas citri* pv. *malvacearum* was initially reported in Alabama, USA, in 1891 (Jalloul et al., 2015). While from Pakistan, the bacterial blight disease of cotton was reported in 1986 (Manna et al., 2024). The losses of cotton yield due to bacterial blight caused by Xcm can be between 1% and 27%. Yield losses have been reported to reach up to 80% in some severe cases (Manna et al., 2024). In cases of severe infestation, losses may exceed 50%, especially when the bacterium is thriving under hot and humid conditions (Mijatović et al., 2021). It has been proven that the disease severely limits the fibre production in the United States (Senchina et al., 2003), India (Verma, 1986), and Africa (Follin et al., 1988). It is currently distributed throughout all the world's cotton-producing areas (Hillocks, 1992; Zomorodian and Rudolph, 1993).

The bacterium Xanthomonas is one of *the* world's ten most significant plant pathogenic bacteria (Mansfield et al., 2012). It has previously been implicated as the incitant of cotton bacterial blight by *Pseudomonas malvacearum*, Bacterium *malvacearum*, X. campestris pv. *malvacearum*, and X. *axonopodis pv. malvacearum* (Schaad et al., 2006, 2007). The motile, rod-shaped bacterium measuring 0.7–0.9 µm in width and 1–1.2 in length can be grown singly or in pairs. The aerobic, gram-negative, non-spore-forming, non-acidifying cells are being formed. They possess a single polar flagellum that is 1–1.2 × 0.7–0.9 µm in diameter. On nutrient agar, Xcm forms yellow, convex, mucoid colonies (Showmaker et al, 2017)

Various methods are available for the control of cotton bacterial blight. The most widely used method is chemical control because of its extensive market availability and relatively low price. Nevertheless, since the bacterium is responsible for a systemic infection, the efficacy of poisonous chemicals in inhibiting the disease is restricted (Hussain and Tahir, 1993; Khan, 1995). Moreover, excessive use of chemicals can be harmful to the environment and human health. Though initially they were not very effective, researchers have also explored biological means, including the deployment of plant extracts (Mukhtar and Chohan, 1999). Plant extracts have gained prominence in recent times as eco-friendly alternatives to chemical bactericides.

Pakistan's biggest cash crop, cotton (*Gossypium hirsutum* L.), contributes substantially to the nation's GDP, rural economy, and foreign exchange reserves. Yet Xcm blight caused bacterial blight continues to be a threat to productivity, causing yield loss ranging up to 40–50% in conducive situations (Manna et al., 2024). Since it harms both farmer profitability and the country's cotton production, the re-emergence of the disease in cotton-producing regions such as Dera Ghazi Khan warrants prompt concern. While conventional chemical control continues to be popular, its downside highlights the importance of sustainable options. Plant extracts, such as those from *Azadirachta indica* and *Moringa olifer*, provide farmers with cheaper, readily available, and environmentally friendly options. Therefore, it is important to examine the pathogenicity of this bacterium on local cotton varieties and evaluate safer and more effective management options, particularly for Pakistani farming communities that have limited resource bases.

MATERIALS AND METHODS

Experimental Site and Design

The experiment was conducted at the Research Area of the Department of Plant Pathology, Ghazi University, Dera Ghazi Khan, Pakistan, during 2025 cotton season. Three varieties were used in a randomized complete block design (RCBD). To assess disease development and management tactics, three cotton varieties, BS-313, FH-333, and FH-Lalazar, were grown using standard field conditions.

Pathogen Isolation and Identification

In the field, leaves with lesions and angular leaf spots, two common signs of bacterial blight, were collected. After rinsing with sterile distilled water and surface sterilizing with 1% sodium hypochlorite for a minute, and then rinsing in autoclaved distilled water, the infected tissues were aseptically transferred onto nutrient agar medium. Bacterial colonies were purified by repeated streaking after plates were incubated for three to five days at 28 ± 2 °C. The convex, mucoid, and yellow morphology of the isolated colonies allowed for their identification. A Gram stain test was also conducted, confirming that the bacterium was Gram-negative.

Pathogenicity Test

A fresh inoculum was made using pure cultures of Xcm that had been cultured on nutrient agar for 48 hours. Sterile distilled water was used to suspend bacterial colonies, and the concentration was set at around 10^8 CFU/mL. Fifty to sixty days after seeding, healthy plants of the three cotton varieties (FH-333, BS-313, and FH-Lalazar) were carefully chosen and verified to be free of signs of bacterial blight before inoculation.

There were two inoculation techniques used. The leaf clipping method involved making tiny cuts at the leaf margins with sterile scissors dipped in a bacterial suspension, and the spray method involved using a handheld sprayer to evenly apply the bacterial suspension to the abaxial leaf surfaces until runoff occurred. Sterile distilled water was used to treat the control plants. Symptomatic leaves were collected and re-isolated on nutrient agar to confirm Koch's postulates.

Disease Assessment

The disease incidence was calculated using the formula given below.

$$\text{Disease Incidence (\%)} = \left(\frac{\text{Number of infected plant}}{\text{Total number of plant observed}}\right) \times 100$$

Fifteen days after inoculation, cotton plants were carefully examined for the characteristic symptoms of bacterial blight, including angular leaf spots, water-soaked lesions, blight patches, and necrosis. To quantify the intensity of the disease, disease severity was recorded according to the 0–10 Brinkerhoff disease rating scale (Brinkerhoff, 1977). In every experimental plot, twenty plants were randomly chosen, and on every plant, three fully expanded leaves from the middle part of the plant canopy were scored. This method helped ensure that the data were representative of field disease pressure overall and minimized bias in the evaluation. Visual disease scoring was done by comparing the progression of lesion development and leaf area infected with the descriptions in the Brinkerhoff scale.

Table 1. Rating scale for disease screening of bacterial blight of cotton (Brinkerhoff, 1977)

Grade	Symptom Description	Level
0	No macroscopic symptoms	Immune (I)
1-3	Round, dry pinhead-sized lesions developed	Resistant (R)
4-6	Lesions turned into dry, angular lesions	Tolerant (T)
7-9	Lesions turned into water-soaked spot	Susceptible (S)
10	Spot turned to large, angular water-soaked lesions on leaf veins	Highly Susceptible (HS)

$$\text{Disease Severity (\%)} = \left(\frac{\sum Ratings}{\text{Total number of leaves observed} \times \text{Maximum disease rating}}\right) \times 100$$

Statistical Analysis

The collected data on disease severity and incidence were subjected to statistical analysis using Analysis of Variance (ANOVA) under a randomized complete block design (RCBD). Treatment means were compared using the Least Significant Difference (LSD) test at a 5% probability level ($p < 0.05$) to determine significant differences among treatments. Statistical computations were performed using SPSS version 26.0 (IBM Corp., Armonk, NY) or equivalent software.

Management Practices

Botanical Extracts

Moringa oleifera (Moringa tree) (T_1) and *Azadirachta indica* (Neem tree) (T_2) were used to prepare botanical extracts since their leaves contain naturally occurring bioactive compounds. The major active ingredients present in *M. oleifera* are saponins (Abalaka et al., 2012), while the major bioactive compound present in *A. indica* is nimbine (Lokanadhan et al., 2012). 15% aqueous leaf extracts of the two plants were prepared and used to assess their effectiveness against Xcm in this research.

Healthy plants' disease-free leaves were collected for processing, washed extensively with tap water to clean surface, and then shade-dried for approximately 48 hours to ensure total drying. Drying preserved the bioactive compounds and inhibited microbial contamination. The dehydrated leaves were pulverized into a fine powder through the use of an electric grinder. To prepare a 15% aqueous extract, 15 g of leaf powder of every plant material was individually mixed with 100 mL of sterile distilled water. Clear extracts were achieved by filtering the mixtures through a double layer of muslin cloth after proper mixing to release soluble content. Since prolonged storage may diminish the antibacterial activity of the extracts, they were invariably prepared freshly before use to ensure optimal activity.

A hand-held sprayer was used to spray the extracts in two different treatments. To reduce evaporation and increase leaf absorption, spraying was done early in the morning under quiet conditions. For even coverage of the whole plant canopy, for each treatment, the cotton leaves were sprayed both on the adaxial (top) and abaxial (bottom) sides until run-off occurred. This application method allowed the extracts to be in close contact with the leaf surfaces, where infection by Xcm commonly initiates. The botanical extract treatments used for this experiment are given in Table 2.

Table 2. Botanical extracts used for the management of bacterial blight of cotton

Treatments	Common Name	Botanical Name	Key Bioactive Constituents	Plant Part Used	Concentration
T1	Moringa	*Moringa oleifera*	Saponins (Abalaka et al., 2012)	Leaves	15%
T2	Neem	*Azadirachta indica*	Nimbine (Lokanadhan et al., 2012)	Leaves	15%

Chemical Control

The inhibitory capacity of two chemical formulations on bacterial blight of cotton was determined. These included Kasomil 47 WP (T3), which was made up of copper oxychloride and kasugamycin and was sprayed at the recommended rate of 250 g in 100 L of water. The second one was Triplet 75 WG (T4), which was applied at 65 g per 100 L of water and had a mixture of Tebuconazole and Trifloxystrobin. In order to ensure product integrity and consistency in treatments, the both chemicals were purchased from Petron Group, Pakistan.

With the use of a precision balance, required quantities of chemicals were weighed prior to being gradually mixed with fresh water and agitated to avoid clumping and ensure even distribution in an attempt to produce spray solutions. Due to possible decreases in chemical stability and field effectiveness with prolonged storage, each solution was prepared fresh on the day of application.

A hand sprayer was used to uniformly spray the solution over the cotton canopy. In order to maximize contact between the active constituents and potential sites of infection, the top (adaxial) and bottom (abaxial) surfaces of the leaves were thoroughly covered until runoff. Applications were made early morning when the temperature was moderate, the relative humidity was high, and wind velocity was minimal. Such conditions were used to avoid spray drift, reduce evaporation, and increase droplet retention.

The employment of clean sources of water, accurate measurement of the dosage, and consistent spraying pressure were some of the routine safety precautions when spraying chemicals. Caution was also used to avoid overapplication, which could lead to soil buildup or phytotoxicity.

Table 3. Chemical formulations used for the management of bacterial blight of cotton

Treatments	Formulation	Active Ingredient	Concentration / 100 L	Source
T3	Kasomil 47 WP	Kasugamycin + Copper oxychloride	250 g	Petron Group, Pakistan
T4	Triplet 75 WG	Tebuconazole + Trifloxystrobin	65 g	Petron Group, Pakistan

RESULTS

Identification of Pathogen

Following three to five days of incubation at 28 ± 2 °C, the colonies that were obtained on nutrient agar had a mucoid texture, convex elevation, and yellow pigmentation, all of which are known morphological characteristics of *Xanthomonas* species. The isolate's purity was confirmed by the uniform colonies with the same traits that were produced by repeated streaking. After Gram staining, microscopic analysis showed that the bacterium was Gram-negative and appeared as tiny, rod-shaped cells devoid of spores. These characteristics align with the accepted definition of Xcm. Colony morphology and Gram reaction together offered unmistakable proof of the pathogen's identity, which was subsequently applied to tests of cotton varieties' pathogenicity. Xcm was the pathogen identified by the staining reaction and morphological characteristics taken together.

Pathogenicity Test

All three cotton cultivars exhibited symptoms of bacterial blight 7–10 days after inoculation, showing the virulence of the isolate. FH-333 presented lesions that progressed to form clear water-soaked spots, whereas BS-313 formed angular lesions that continued to develop into water-soaked areas characteristic of bacterial blight. Relatively, more severely affected was FH-Lalazar, which presented widespread leaf infection where lesions disseminated and formed several water-soaked spots. Conversely, control plants that had been administered sterile distilled water did not show any infection symptoms throughout the study. Gram staining once again confirmed the rod-shaped, Gram-negative nature of the pathogen after it had been re-isolated successfully from infected tissues, producing colonies of similar morphology to the original isolate. These findings satisfied Koch's postulates in the sense that they verified that the isolated bacterium was responsible for the disease that was seen. The reactions of different varieties to infection are listed in Table 4.

Table 4. Pathogenicity response of cotton varieties to Xcm

Variety	Symptoms observed (7–10 days)	Confirmation
BS-313	Angular lesions that turned into water-soaked areas	Positive
FH-333	Lesions developed into distinct water-soaked spots	Positive
FH-Lalazar	Extensive infection with numerous lesions turning into water-soaked spots	Positive
Control	No symptoms	Negative

Disease Assessment

Disease incidence and severity were taken 15 days after inoculation. Percentage disease severity, based on the 0–10 Brinkerhoff disease rating scale (Brinkerhoff, 1977), revealed that FH-Lalazar had the highest severity of disease (70.91%), which was followed by BS-313 (61.49%) and FH-333 (58.04%). The incidence of disease moved in a similar pattern, with FH-Lalazar having 78.57%, BS-313 72.56%, and FH-333 70.43%. Based on Brinkerhoff reaction scale, all three genotypes were Susceptible (S) and did not show resistance or tolerance at all. Though FH-333 had better performance compared to the other two cultivars, the overall result confirms that all genotypes tested are still susceptible to bacterial blight.

Table 5. Disease severity and incidence (%) of bacterial blight in cotton varieties (15 days after inoculation)

Variety	Disease severity (%)	Disease Incidence (%)	Reaction
BS-313	61.49	72.56	Susceptible (S)
FH-333	58.04	70.43	Susceptible (S)
FH-Lalazar	70.91	78.57	Susceptible (S)

Effect of Botanical extracts

Disease severity in the control without treatment was as high as 61.49% in BS-313, 58.04% in FH-333, and 70.91% in FH-Lalazar, which confirms that all three were susceptible in the field (Table 5). Application of neem extract (T1, 15%) lowered disease severity to 52.88% in BS-313, 49.22% in FH-333, and 62.40% in FH-Lalazar. Moringa extract (T2, 15%) also improved the plant health by reducing the disease severity to 57.19% in BS-313, 56.07% in FH-333, and 67.34% in FH-Lalazar.

Neem extract resulted in greater decreases than moringa in all varieties based on a comparison of treatments. Neem treatment lowered the severity of BS-313 and FH-333 by a mean of 9–10% compared to control, while moringa treatment lowered the severity by around 4–5%. Neem lowered severity more than 8% in the most susceptible variety, FH-Lalazar, while moringa lowered it by less than 4%. This trend indicates that neem extract was more suppressive of the disease, although the two treatments were both partially successful.

Table 6. Effect of botanical extracts on disease severity (%)

Variety	Untreated Control	T1 (Neem extract 15% concentration)	T2 (Moringa extract 15% concentration)
BS-313	61.49	52.88	57.19
FH-333	58.04	49.22	56.07
FH-Lalazar	70.91	62.40	67.34

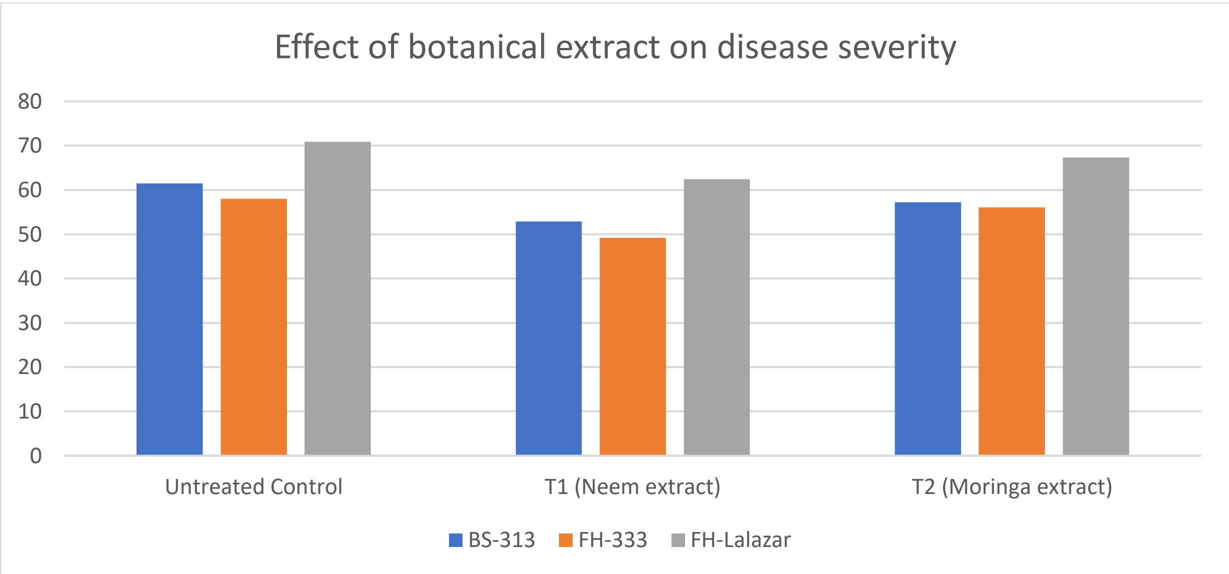

Figure 1. Effect of botanical extracts (Neem and Moringa) on bacterial blight severity in cotton varieties

Chemical Control for Bacterial Blight of Cotton

All three varieties of cotton gave a significant reduction in the severity of the disease compared to the untreated plants. Use of Tebuconazole + trifloxystrobin and Kasugamycin + copper oxychloride reduced the disease severity on BS-313 from 61.49% to 12.30% and 21.52%, respectively. Comparable results were obtained in FH-333, where disease severity reduced from 58.04% in the control that was not treated to 10.45% with Kasugamycin + copper oxychloride and 20.60% with Tebuconazole + trifloxystrobin. After chemical treatments, FH-Lalazar, which was the most infected variety in the natural environment (70.91% in the control), also showed a great improvement, with severity falling to 14.89% under Kasugamycin + copper oxychloride and 24.11% under Tebuconazole + trifloxystrobin. Tebuconazole + trifloxystrobin was uniformly less effective than Kasugamycin + copper oxychloride in all the varieties, as indicated by the relative performance of the two chemicals. Kasugamycin + copper oxychloride treatment suppressed disease in BS-313 and FH-333 by nearly 80%, while Tebuconazole + trifloxystrobin suppressed disease by merely about 60–65%. The same trend was observed in the highly susceptible line FH-Lalazar, where Tebuconazole + trifloxystrobin resulted in a lesser decrease in severity compared to the control, but Kasugamycin + copper oxychloride decreased it by over three-fourths (Table 7).

Table 7. Effect of chemical treatments on disease severity (%)

Variety	**Untreated Control**	**Kasugamycin + copper oxychloride**	**Tebuconazole + trifloxystrobin**
BS-313	61.49	12.30	21.52
FH-333	58.04	10.45	20.60
FH-Lalazar	70.91	14.89	24.11

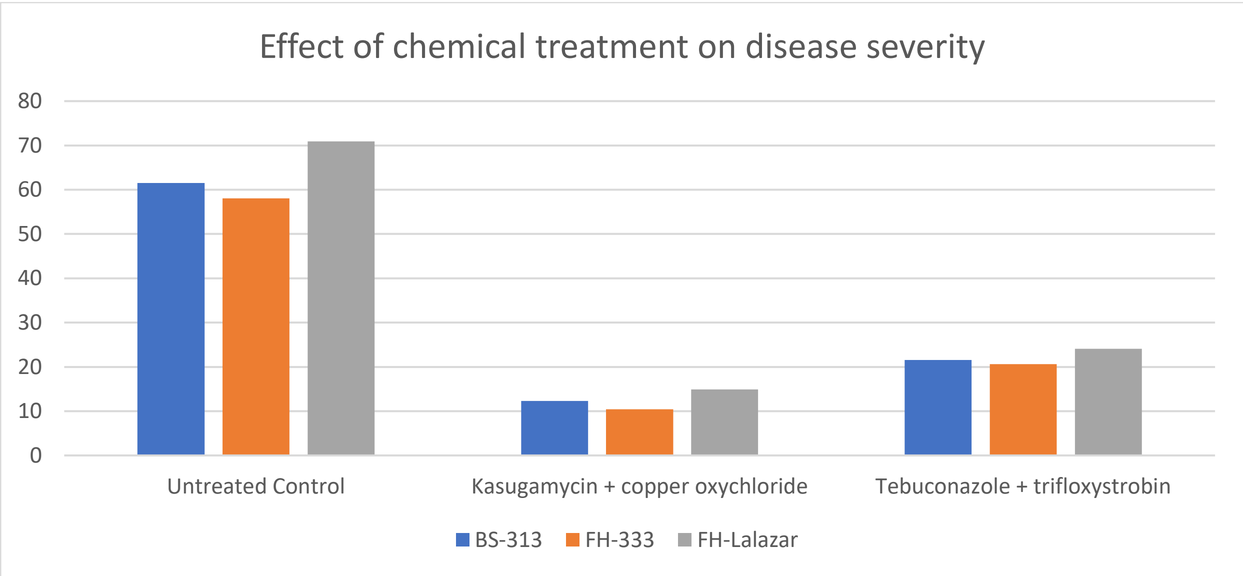

Figure 2. Effect of chemical treatments (Kasugamycin + Copper oxychloride and Tebuconazole + Trifloxystrobin) on bacterial blight severity in cotton varieties

Statistical Validation of Treatment Effects

One-way ANOVA revealed a highly significant difference ($p < 0.001$) among treatments in reducing bacterial blight severity across all cotton varieties. LSD comparison ($p < 0.05$) confirmed that Kasugamycin + Copper oxychloride exhibited the lowest mean disease severity (12.55%), followed by Tebuconazole + Trifloxystrobin (22.07%). Neem and moringa extracts (54.83% and 60.20%, respectively) also reduced severity compared to the untreated control (63.48%), though the reductions were less pronounced.

These statistical findings validate that chemical treatments were significantly more effective than botanical treatments and controls.

Table 8. ANOVA for disease severity among treatments

Source of Variation	df	Mean Square	F-value	p-value
Treatments	4	2304.26	32.74	<0.001***
Error	10	70.42	–	–
Total	14	–	–	–

The results of the Analysis of Variance (ANOVA) performed on the disease severity data recorded under different management treatments (Table 8). This analysis was carried out to determine whether the treatments applied had a statistically meaningful impact on reducing bacterial blight severity in cotton. The ANOVA showed that the treatment effect was highly significant ($p < 0.001$), indicating that the observed differences among the treatments were not due to random variation but were the result of true differences in treatment performance. This confirms that the application of various management strategies had different levels of effectiveness in controlling the disease.

Table 9. LSD mean comparison of treatments ($p < 0.05$)

Treatment	Mean Severity (%)	Group	Mean Disease Severity (%) ± SD
Kasugamycin + Cu Oxychloride	12.55	a	13.09 ± 3.88
Tebuconazole + Trifloxystrobin	22.07	b	21.90 ± 3.90
Neem Extract (15%)	54.83	c	55.17 ± 4.31
Moringa Extract (15%)	60.20	c	60.53 ± 4.50
Control	63.48	d	63.46 ± 3.71

The comparison of mean disease severity values along with their associated standard deviations (SD) for each treatmentallows a better understanding of the variability and consistency of treatment responses (Table 9). Additionally, the Least Significant Difference (LSD) test was used to statistically separate the treatments into distinct groups, where treatments with different letters differ significantly in their effect. This helps in identifying which treatments offered the greatest reduction in disease severity and which ones performed similarly or differently from each other. The chemical treatments formed distinct groups, reflecting their superior disease suppression compared to botanical extracts and the untreated control (Table 9).

DISCUSSION

The present research highlights the consistent issue of cotton bacterial blight caused by Xcm in the agroclimatic conditions of Dera Ghazi Khan. With the help of morphological characteristics and Gram staining, the pathogen was identified and isolated successfully. This verifies earlier findings that described *Xanthomonas* spp. to be rod-shaped, gram-negative bacteria producing yellow, mucoid colonies on nutrient agar (Schaad et al., 2006, 2007). Cotton bacterial blight was first documented in 1891 in the USA (Jellous et al., 2015) and later reported from

Pakistan in 1986 (Mana et al., 2024). The disease has been one of the most harmful barriers to cotton yields worldwide for many decades. Since bacterial blight has the potential to reduce yields by up to 27-80% under optimal weather, its economic impact has long been emphasized (Manna et al., 2024).

The study's varietal screening revealed that BS-313, FH-333, and FH-Lalazar were all vulnerable to bacterial blight, with varying degrees of disease manifestation. None of the genotypes demonstrated resistance in the field, with FH-Lalazar displaying the highest disease severity, followed by BS-313 and FH-333. These findings are consistent with previous studies demonstrating that most of Pakistan's commercially cultivated cotton types continue to be vulnerable to bacterial blight (Hillocks, 1992; Akhtar et al., 2010). Although the differences in severity were small, this variability reflects host-pathogen interaction variations and may also be influenced by the presence of virulent races such as Race 18, which has been widely reported in Pakistan (Hussain, 1984). The absence of complete resistance in extensively grown cultivars highlights the need for continued breeding and screening programs to transfer durable resistance genes to high-yielding backgrounds.

In contrast with the untreated check, botanical extracts with plant extracts revealed promising results in alleviating the severity of bacterial blight. Both the extracts of neem (*Azadirachta indica*) and moringa (*Moringa oleifera*) at a 15% concentration reduced disease levels, with neem performing better than moringa at all three varieties. These results validate plant compounds' antimicrobial potential to fight bacterial blight pathogens. Azadirachtin and nimbin are two active compounds present in neem leaves with well-documented antibacterial activity (Lokanadhan et al., 2012). Likewise, the flavonoids, phenolics, and saponins present in moringa leaves are known to inhibit bacterial growth (Rashid et al., 2016). While they cautioned that efficacy differed by crop and extract, similar findings were described by Mukhtar and Chohan (1999), where different extracts from plants were effective against *Xanthomonas* spp. The notable role of plant extracts in reducing the level of disease in cotton and other crops has also been confirmed by more up-to-date studies (Mashamaite et al., 2022; Adusei et al., 2022). The uniform efficacy of plant treatments on all varieties renders them attractive as environmentally friendly options, even though they were less effective, for farmers with restricted resources, despite their inferior performance compared to chemical bactericides.

The application of chemical management methods here indicated that synthetic treatments performed better than botanical extracts when it came to reducing the severity of bacterial blight. These findings were statistically supported by ANOVA and LSD tests ($p < 0.05$), confirming significant differences among treatments and verifying that chemical control showed the highest efficacy in reducing disease severity. Tebuconazole + trifloxystrobin and kasugamycin + copper oxychloride both substantially reduced the occurrence of the disease compared to the control. The dual effect of copper oxychloride's contact protection with kasugamycin's dual effect as an aminoglycoside antibiotic that inhibits bacterial protein synthesis could be the rationale behind the superior control of kasugamycin + copper oxychloride. These results are in accordance with previous studies proving the high efficacy of copper-based fungicides and antibiotics, such as streptomycin and oxytetracycline, against bacterial cotton diseases (Hussain & Tahir, 1993; Khan, 1995). Yet, due to their possibilities of environmental pollution, pathogen resistance, and phytotoxicity, their application must be executed with utmost caution.

Everything considered, this research underscores the importance of integrating chemical and biological methods for the long-term management of cotton bacterial blight. Although chemical treatments remain the best in inhibiting diseases immediately, their limitations necessitate safe, eco-friendly alternatives such as neem and moringa extracts. The most likely way forward is through integrated management methods combining biological extracts, host resistance, and judicious chemical applications. Long-term bacterial blight management requires an integrated approach that reduces the occurrence of resistant populations of pathogens, keeps costs down, and maintains environmental safety, as emphasized by previous researchers (Verma, 1986; Mansfield et al., 2012).

CONCLUSION

Based on this study, bacterial blight of cotton caused by Xcm remains a major production-limiting factor in Dera Ghazi Khan. All three tested cotton varieties—BS-313, FH-333, and FH-Lalazar were found to be susceptible, with FH-Lalazar exhibiting the highest disease severity. Among the biological treatments, neem (*Azadirachta indica*) and moringa (*Moringa oleifera*) leaf extracts at a 15% concentration effectively reduced disease incidence, with neem performing comparatively better. Chemical treatments provided stronger disease suppression, with kasugamycin + copper oxychloride showing the greatest efficacy, followed by tebuconazole + trifloxystrobin.

These findings indicate that while chemical options remain the most effective for short-term control, botanical extracts offer promising, environmentally safer alternatives. Long-term and sustainable management of bacterial blight should adopt an integrated approach combining resistant cultivars, botanical extracts, and judicious chemical use. Future research should focus on screening additional plant-derived extracts for antibacterial efficacy and developing or integrating resistant cotton genotypes through breeding programs to achieve durable resistance under local agroclimatic conditions.

ACKNOWLEDGEMENT

The authors gratefully acknowledge the financial support for this research provided by the Higher Education Commission (HEC) of Pakistan under the National Research Program for Universities (NRPU) through project number 17001.

REFERENCES

Abalaka, M. E., Daniyan, S. Y., Oyeleke, S. B., & Adeyemo, S. O. (2012). The antibacterial evaluation of *Moringa oleifera* leaf extracts on selected bacterial pathogens. *Journal of Microbiology Research*, *2*(2), 1–4.

Adusei, S., Otchere, J. K., Oteng, P., Mensah, R. Q., & Tei-Mensah, E. (2022). Phytochemical analysis, antioxidant and metal chelating capacity of *Tetrapleura tetraptera*. *Heliyon*, *5*(11), e02865.

Brinkerhoff, L. A. (1977). Bacterial blight of cotton. *FAO Consultant Report Pak/73/026*. FAO, Rome.

CGIAR. (2020). *Cotton crop: A situational analysis of Pakistan*. CGIAR Research Program on Climate Change, Agriculture and Food Security (CCAFS). https://cgspace.cgiar.org/bitstreams/0925c0ef-f70e-438c-b365-89fc8cb5f1d5/download?utm_source=chatgpt.com

Follin, J. C., Girardot, B., Mangano, V., & Benitez, R. (1988). Nouveaux résultats sur le déterminisme génétique de la résistance foliaire totale du cotonnier (*Gossypium hirsutum* L.) à la bactériose *Xanthomonas campestris* pv. *malvacearum* (Smith) Dye, race 18 et 20. *Coton et Fibres Tropicales*, *43*, 167–175.

Government of Pakistan. (2023). *Economic Survey of Pakistan 2023–24*. Ministry of Finance, Government of Pakistan. https://www.finance.gov.pk/survey_2122.html

Hillocks, R. J. (1992). Bacterial blight. In R. J. Hillocks (Ed.), *Cotton diseases* (pp. 39–85). CAB International.

Hussain, T. (1984). Prevalence and distribution of *Xanthomonas campestris* pv. *malvacearum* races in Pakistan and their reaction on different cotton lines. *Tropical Pest Management*, *30*(2), 159–162.

Hussain, T., & Tahir, M. (1993). Chemical control of bacterial blight of cotton. *Pakistan Journal of Phytopathology*, *5*, 119–121.

International Cotton Advisory Committee (ICAC). (2023). *Cotton: World statistics and market outlook*. https://icac.org

Jalloul, A., Sayegh, M., Champion, A., & Nicole, M. (2015). Bacterial blight of cotton. *Phytopathologia Mediterranea*, *54*(1), 3–20.

Khan, M. A. (1995). *In vitro* sensitivity of *Xanthomonas campestris* pv. *malvacearum* to Agrimycin-100 and its effect on cotton plant in relation to symptom expression. *Pakistan Journal of Phytopathology*, *7*, 199–201.

Lokanadhan, S., Muthukrishnan, P., & Jeyaraman, S. (2012). Neem products and their agricultural applications. *Journal of Biopesticides*, *5*(Suppl.), 72–76.

Manna, S., Gawande, S. P., Meshram, M. D., Ingle, R., & Nagrale, D. (2024). Phenotypic and genetic characterization of *Xanthomonas citri* pv. *malvacearum* strains affecting upland cotton in Central India. *Indian Phytopathology*, *77*(1), 125–137.

Mansfield, J., Genin, S., Magori, S., Citovski, V., Anum, S. S., Ronald, P., Dow, M., Verdier, V., Beer, S. V., Machado, M. A., Toth, I., Salmond, G., & Foster, G. (2012). Top 10 plant pathogenic bacteria in molecular plant pathology. *Molecular Plant Pathology*, *13*(6), 614– 629.

Mashamaite, C. V., Ngcobo, B. L., Manyevere, A., Bertling, I., & Fawole, O. A. (2022). Assessing the usefulness of *Moringa oleifera* leaf extract as a biostimulant to supplement synthetic fertilizers: A review. *Plants*, *11*(17), 2214.

Mijatović, J., Severns, P. M., Kemerait, R. C., Walcott, R. R., & Kvitko, B. H. (2021). Patterns of seed-to-seedling transmission of *Xanthomonas citri* pv. *malvacearum*, the causal agent of cotton bacterial blight. *Phytopathology*, *111*(12), 2176–2184.

Mukhtar, T., & Chohan, R. A. (1999). Effect of leaf extracts of some plants on the growth of nematophagous fungus *Verticillium chlamydosporium*. In *Proceedings of the 2nd National Conference on Plant Pathology* (pp. 27–29). University of Agriculture, Faisalabad.

Schaad, N. W., Postnikova, E., Lacy, G., Sechler, A., Agarkova, I., Stromberg, P. E., Stromberg, V. K., & Vidaver, A. K. (2006). Emended classification of *Xanthomonas* pathogens on citrus. *Systematic and Applied Microbiology*, *29*(8), 690–695.

Schaad, N. W., Postnikova, E., Lacy, G., Sechler, A., Agarkova, I., Stromberg, P. E., Stromberg,

V. K., & Vidaver, A. K. (2007). List of new names and new combinations previously effectively, but not validly, published: Validation list no. 115. *International Journal of Systematic and Evolutionary Microbiology, 57*(10), 2459–2460.

Senchina, D. S., Alvarez, I., Cronn, R. C., Liu, B., Rong, J., Noyes, R. D., Paterson, A. H., Wing, R. A., Wilkins, T. A., & Wendel, J. F. (2003). Rate variation among nuclear genes and the age of polyploidy in *Gossypium. Molecular Biology and Evolution*, *20*(4), 633–643.

Showmaker, K. C., Jones, J. B., & Smith, S. A. (2017). The genome of the cotton bacterial blight pathogen *Xanthomonas citri* pv. *malvacearum* strain MSCT1. *Genome Announcements, 5*(28), e00622-17.

USDA Foreign Agricultural Service. (2025). *Cotton and products annual: Pakistan.* Global Agricultural Information Network (GAIN) Report No. PK2025-0002. Islamabad: U.S. Department of Agriculture.

Verma, J. P. (1986). *Bacterial blight of cotton.* CRC Press.

Yadav, M., Saxena, G., Verma, R. K., Asif, M. H., Singh, V. P., Sawant, S. V., & Singh, S. P. (2024). Genome-wide identification and expression analysis of autophagy-related genes (ATG) in *Gossypium* spp. reveals their crucial role in stress tolerance. *South African Journal of Botany, 167*, 82–93.

Zomorodian, A., & Rudolph, K. (1993). *Xanthomonas campestris* pv. *malvacearum*, cause of bacterial blight of cotton. In J. G. Swings & E. L. Civerolo (Eds.), *Xanthomonas* (pp. 19– 28). Chapman and Hall.zzz