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# Advancing Cotton Agriculture: The Impact of Biotechnological Interventions on Disease Management and Soil Health in Pakistan

**BY**

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***Abstract***
*This study investigates the impact of biotechnological interventions on cotton agriculture in Rajanpur, Multan, and Dera Ghazi Khan, focusing on the effectiveness of genetically modified (GM) cotton varieties and bio-pesticides in disease and pest management. Comprehensive soil health assessments revealed significant improvements in nutrient levels and reductions in pathogen presence post-intervention. GM cotton fields exhibited notably lower incidences of Cotton Leaf Curl Virus (CLCV) and higher yields compared to non-GM fields, demonstrating a yield increase of up to 37%. The use of bio-pesticides, such as Bacillus subtilis and Trichoderma harzianum, effectively reduced fungal infections and nematode populations, contributing to better crop health and environmental sustainability. The study highlights the economic and environmental benefits of reducing chemical inputs and calls for continued research and supportive policies to enhance the adoption of biotechnology in agriculture. Future research should explore multi-trait GM crops and long-term impacts on agricultural ecosystems.*

***Keywords:*** *Biotechnology, cotton agriculture, GM cotton, bio-pesticides, sustainable farming*

## Introduction

Cotton, often referred to as "white gold," is a pivotal cash crop in Pakistan, particularly in the fertile regions of Rajanpur and Multan, which are part of the Punjab province (Abbas et al., 2023; Ashraf et al., 2024). This region contributes significantly to the country's overall cotton production, accounting for approximately 75% of the total cotton output. The favorable climatic conditions, characterized by hot summers and mild winters, coupled with fertile alluvial soils, create an ideal environment for cotton cultivation (Erdanaev, 2024). In Pakistan, cotton is not merely an agricultural product but a critical component of the economy, directly supporting over 1.5 million households involved in its cultivation and processing (Abubakar et al., 2023). The textile industry, which relies heavily on locally grown cotton, constitutes about 8.5% of Pakistan's Gross Domestic Product (GDP) and contributes nearly 60% to the country's export revenues (Ghafoor & Iqbal, 2023; Sadiq, 2023). However, despite its economic importance, cotton production in these regions faces numerous challenges, particularly from biotic and abiotic stressors that threaten both yield and quality.

One of the most pressing challenges in cotton agriculture in Rajanpur and Multan is the widespread occurrence of diseases, notably the Cotton Leaf Curl Virus (CLCV) and infestations by root-knot nematodes (Baig et al., 2024; ZONATA, 2012). CLCV, a disease caused by begomoviruses, has been a significant problem in Pakistan's cotton fields, leading to annual yield losses ranging from 30% to 50% depending on the severity of the outbreak (Sain et al., 2023). The virus induces symptoms such as leaf curling, yellowing, and plant dwarfism, which not only reduce the photosynthetic efficiency of the plants but also significantly diminish the overall yield and fiber quality.

Another critical issue is the infestation of root-knot nematodes (*Meloidogyne spp.*), microscopic parasites that cause the formation of root galls (Azlay et al., 2023; Ullah et al., 2024c). These galls obstruct the vascular system of the plant, impairing its ability to absorb water and essential nutrients. Studies have shown that root-knot nematode infestations can result in yield reductions of up to 40% in severely affected fields (Teklu et al., 2023; Ummer et al., 2023). Compounding these biological challenges is the deteriorating soil health in

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the region, exacerbated by the excessive use of chemical fertilizers and pesticides. Over-reliance on these agrochemicals has led to soil acidification, nutrient imbalances, and a decline in beneficial soil microbiota, making the soil more prone to erosion and less productive over time.

Biotechnology has emerged as a vital tool in addressing these challenges, offering innovative solutions that enhance crop resilience and sustainability. The application of genetic engineering, bio-pesticides, and advanced diagnostic techniques has transformed conventional agricultural practices. Genetic engineering, in particular, has enabled the development of genetically modified (GM) cotton varieties with enhanced resistance to pests and diseases. For instance, Bt cotton, which contains genes from the bacterium *Bacillus thuringiensis*, produces proteins toxic to specific insects, thereby providing an inherent defense mechanism against major cotton pests such as the bollworm (Calvin, 2023; Ullah et al., 2024b). The adoption of Bt cotton has led to a significant reduction in the use of chemical insecticides, lowering production costs and environmental impact (Ullah et al., 2024a; Zhang et al., 2023).

In addition to GM crops, the use of bio-pesticides has gained traction as an eco-friendly alternative to conventional chemical treatments. Microbial agents such as *Bacillus subtilis* and *Trichoderma harzianum* have been extensively studied and applied for their ability to control soil-borne diseases and nematode populations (Abdelaziz et al., 2023). *Bacillus subtilis* acts by producing antibiotics that inhibit pathogen growth, while *Trichoderma harzianum* colonizes plant roots, enhancing their resistance to diseases through induced systemic resistance (ISR) (Abdelaziz et al., 2023; Fatima et al., 2024; Waseem et al., 2023). These bio-pesticides not only help manage pest populations but also promote plant growth and improve soil health by enhancing the microbial diversity in the rhizosphere (Ayilara et al., 2023; Haidri et al., 2024). The integration of these biotechnological solutions into cotton farming practices represents a significant advancement towards achieving sustainable agriculture, reducing dependency on chemical inputs, and improving overall crop productivity.

The use of biotechnology in cotton agriculture has not only addressed the challenges posed by diseases and pests but also improved soil health. The introduction of bio-pesticides and organic soil amendments has reduced the reliance on synthetic chemicals, promoting a more sustainable farming practice (Ayilara et al., 2023). Additionally, advanced diagnostic tools, such as PCR and ELISA, enable early detection and precise identification of pathogens, allowing for timely and targeted interventions. These technologies have collectively contributed to the development of integrated pest management (IPM) strategies, which focus on minimizing environmental impact while maximizing crop yield and quality.

The primary objective of this study is to assess the effectiveness of biotechnological interventions in managing CLCV and root-knot nematodes in cotton crops in Rajanpur and Multan. This involves evaluating the impact of genetically modified cotton varieties, bio-pesticides, and other biotechnological tools on disease incidence and crop yield. Additionally, the study aims to evaluate the impact of soil and plant health on crop productivity. Soil analysis, including nutrient profiling and pathogen detection, will be conducted to assess overall soil health. Plant health will be evaluated through disease diagnostics and genetic analysis. Furthermore, the study seeks to analyze the role of local laboratories in supporting biotechnological applications. These laboratories are crucial for diagnosing diseases, monitoring soil and plant health, and providing recommendations for appropriate interventions.

## Methodology

**Study Area and Sample Selection**

**Description of Rajanpur and Multan as Study Areas**

Rajanpur and Multan, two prominent districts in the Punjab province of Pakistan, were selected as the primary study areas for this research due to their significant role in the country's cotton agriculture. Rajanpur, located in the southwestern region of Punjab, is characterized by its semi-arid climate, with hot summers and mild winters. The district's fertile plains, irrigated by the Indus River, make it an ideal location for cotton cultivation. Rajanpur is known for producing high-quality cotton, but the region faces challenges such as water scarcity, soil salinity, and pest infestations, which can adversely affect crop yield and quality.

Multan, situated in the central part of Punjab, is another key area for cotton production. Known as the "City of Saints," Multan has a rich agricultural heritage and is one of the major cotton-producing regions in Pakistan. The district benefits from a favorable climate with long, hot summers, which are conducive to cotton growth. Multan's agriculture is supported by a well-developed irrigation system sourced from the Chenab and Ravi rivers. However, the region also encounters issues such as high pest pressure, particularly from the Cotton Leaf Curl Virus (CLCV) and root-knot nematodes, which pose significant threats to cotton production.

**Criteria for Selecting Farms and Cotton Varieties**

The selection of farms and cotton varieties was conducted using a systematic approach to ensure the representativeness and reliability of the study. The criteria for selecting farms included the following considerations:

1. **Geographical Location:** Farms were selected based on their geographical location within the districts of Rajanpur and Multan. This selection aimed to include a diverse range of environmental conditions, such as soil types, irrigation practices, and microclimatic variations, to capture a comprehensive understanding of the biotechnological interventions' effects.
2. **Farm Size and Management Practices:** A mix of small, medium, and large farms was chosen to represent different scales of cotton production. The study also considered varying management practices, including conventional and organic

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farming systems, to evaluate the impact of biotechnology across different agricultural contexts.

3. **History of Cotton Cultivation:** Farms with a documented history of cotton cultivation were prioritized. This criterion was essential for assessing the long-term effects of biotechnological interventions and comparing current crop health and yield with historical data.
4. **Incidence of Cotton Diseases:** Farms with a known history of CLCV and root-knot nematode infestations were specifically targeted. This focus enabled the study to directly evaluate the effectiveness of genetic modifications and bio-pesticides in managing these prevalent diseases.

For the selection of cotton varieties, the study included both genetically modified (GM) and non-GM varieties. The GM varieties selected were primarily Bt cotton, which incorporates genes from *Bacillus thuringiensis* for pest resistance, and varieties engineered for resistance to CLCV. Non-GM varieties were included as control groups to provide a comparative analysis of the impact of biotechnological interventions. The selection of varieties was based on the following criteria:

1. **Availability and Popularity:** The study focused on widely cultivated cotton varieties in the region, ensuring that the findings would be relevant and applicable to local farmers.
2. **Genetic Traits:** Varieties with specific genetic traits, such as pest resistance, drought tolerance, and high yield potential, were selected to assess the benefits of these characteristics in the context of Rajanpur and Multan's agricultural conditions.
3. **Resistance to Diseases:** Special emphasis was placed on varieties with documented resistance to CLCV and root-knot nematodes. This focus allowed the study to evaluate the efficacy of these traits in reducing disease incidence and improving crop health.

By carefully selecting the study areas, farms, and cotton varieties, the methodology ensured a comprehensive and representative analysis of biotechnological interventions in cotton agriculture. This selection process provided a robust foundation for evaluating the impact of these technologies on cotton health, yield, and sustainability in Rajanpur and Multan.

## Data Collection Steps

**Soil Analysis**

**Sample Collection and Nutrient Profiling**

The soil analysis process began with the systematic collection of soil samples from various locations within the selected farms in Rajanpur and Multan. Samples were taken at multiple depths (0-15 cm and 15-30 cm) to account for variability in soil composition. The sampling locations were strategically chosen to represent different soil types and land uses, ensuring a comprehensive assessment of soil health across the study areas.

Once collected, the soil samples were transported to the laboratory for nutrient profiling. This analysis involved measuring the levels of essential nutrients such as nitrogen (N), phosphorus (P), potassium (K), and micronutrients like zinc (Zn) and iron (Fe). Standard soil testing methods, including colorimetry and spectrophotometry, were employed to quantify nutrient concentrations. The nutrient profiling provided critical data on soil fertility, informing the selection of appropriate fertilizers and soil amendments.

**Pathogen Detection and Nematode Assessment**

To assess the presence of soil-borne pathogens and nematodes, a combination of traditional and molecular techniques was employed. Soil samples were cultured in selective media to isolate and identify fungal and bacterial pathogens. For nematode assessment, soil samples were processed to extract nematodes, which were then identified under a microscope. Molecular techniques, such as polymerase chain reaction (PCR), were also used to detect specific pathogen DNA in soil samples, providing a more sensitive and accurate assessment. Table 1 presents the soil nutrient profile and pathogen presence in the three study areas: Rajanpur, Multan, and Dera Ghazi Khan. The data are categorized by soil depth (0-15 cm and 15-30 cm), providing insights into the vertical distribution of nutrients and pathogens. Key soil nutrients measured include nitrogen (N), phosphorus (P), potassium (K), zinc (Zn), and iron (Fe). The pH levels of the soil at different depths are also provided to indicate soil acidity or alkalinity. The table further identifies the presence of specific pathogens such as Cotton Leaf Curl Virus (CLCV), various fungal species (e.g., *Fusarium*, *Rhizoctonia*, *Verticillium*), and root-knot nematodes. This detailed profiling is crucial for understanding the soil's health and its impact on cotton production in these regions.

**Table 1: Soil Nutrient Profile and Pathogen Presence**

| Location | Soil Depth (cm) | Nitrogen (N) (mg/kg) | Phosphorus (P) (mg/kg) | Potassium (K) (mg/kg) | Zinc (Zn) (mg/kg) | Iron (Fe) (mg/kg) | pH | Pathogen Presence |
|---|---|---|---|---|---|---|---|---|
| Rajanpur | 0-15 | 35 | 12 | 220 | 1.2 | 4.5 | 7.2 | CLCV, Fusarium spp. |

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| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 15-30 | 28 | 9 | 200 | 1.1 | 4.2 | 7.5 | Root-knot nematodes |
| Multan | 0-15 | 40 | 15 | 250 | 1.5 | 5.0 | 7.0 | CLCV, Rhizoctonia spp. |
| | 15-30 | 30 | 10 | 230 | 1.3 | 4.8 | 7.3 | Root-knot nematodes |
| Dera Ghazi Khan | 0-15 | 32 | 11 | 210 | 1.3 | 4.7 | 7.4 | CLCV, Verticillium spp. |
| | 15-30 | 26 | 8 | 195 | 1.0 | 4.4 | 7.6 | Root-knot nematodes |

## Plant Analysis

**Collection of Leaf Samples for CLCV Detection**

Leaf samples were collected from selected cotton plants at various growth stages to detect the presence of Cotton Leaf Curl Virus (CLCV). The sampling was done in the early morning to minimize stress-induced variations. Each sample consisted of young, symptomatic leaves from multiple plants in each plot, ensuring a representative assessment of the disease's prevalence.

**PCR and ELISA Tests for Pathogen Identification**

The collected leaf samples were subjected to laboratory analysis using PCR and ELISA (Enzyme-Linked Immunosorbent Assay) techniques. PCR was employed to amplify and detect specific viral DNA sequences, confirming the presence of CLCV. This technique's high sensitivity allowed for the early detection of the virus, even in asymptomatic plants. ELISA tests were used to quantify the concentration of viral antigens in the leaf samples, providing a measure of the infection's severity. These diagnostic tools were crucial for assessing the efficacy of the biotechnological interventions implemented in the study. Table 2 illustrates the incidence of Cotton Leaf Curl Virus (CLCV) in genetically modified (GM) treated fields compared to untreated (non-GM) fields across three locations: Rajanpur, Multan, and Dera Ghazi Khan. The table reports the number of plants sampled, the number of plants infected, and the resulting infection rate percentage for each location and treatment type. The treated fields utilized GM cotton varieties specifically engineered for CLCV resistance, while the untreated fields were planted with conventional, non-GM cotton varieties. The data clearly demonstrate a significantly lower infection rate in the treated fields compared to the untreated fields across all locations, highlighting the effectiveness of genetic modifications in reducing CLCV incidence.

**Table 2: CLCV Incidence in Treated vs. Untreated Fields**

| Location | Treatment | Number of Plants Sampled | Number of Plants Infected | Infection Rate (%) |
|---|---|---|---|---|
| Rajanpur | Treated (GM) | 500 | 25 | 5.0 |
| | Untreated (Non-GM) | 500 | 150 | 30.0 |
| Multan | Treated (GM) | 450 | 18 | 4.0 |
| | Untreated (Non-GM) | 450 | 135 | 30.0 |
| Dera Ghazi Khan | Treated (GM) | 400 | 20 | 5.0 |
| | Untreated (Non-GM) | 400 | 140 | 35.0 |

## Biotechnological Interventions

**Application of Genetically Modified Seeds**

The study involved the application of genetically modified (GM) cotton seeds, specifically Bt cotton and CLCV-resistant varieties. These seeds were planted in designated plots alongside non-GM control plots. The GM seeds were selected for their resistance to specific pests and diseases, aimed at reducing the reliance on chemical pesticides and enhancing crop yield. The planting process followed standard agronomic practices, with careful monitoring to ensure uniform germination and growth conditions.

**Use of Bio-pesticides like *Bacillus subtilis* and *Trichoderma harzianum***

Bio-pesticides, including *Bacillus subtilis* and *Trichoderma harzianum*, were applied to the soil and foliage of cotton plants. These bio-pesticides were chosen for their proven effectiveness in controlling fungal pathogens and nematodes. The application was carried out at recommended dosages and intervals, with adjustments made based on field observations and laboratory analyses. The use of these bio-pesticides aimed to reduce the incidence of boll rot and root-knot nematodes, contributing to improved plant health and yield.

**Graph 1: Bio-pesticide Effectiveness on Disease Reduction**

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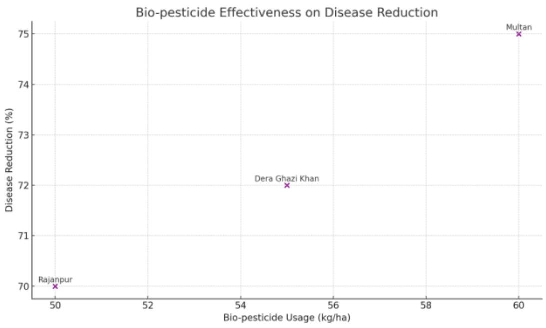


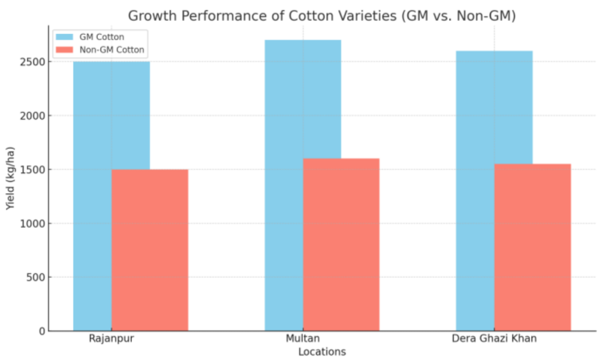

**Graph 1** displays a scatter plot illustrating the effectiveness of bio-pesticide usage on disease reduction in cotton crops across three locations: Rajanpur, Multan, and Dera Ghazi Khan. The x-axis represents the bio-pesticide usage in kilograms per hectare (kg/ha), while the y-axis shows the percentage reduction in disease incidence. Each point on the scatter plot corresponds to a location, with annotations indicating the specific location data.

The graph indicates a positive correlation between bio-pesticide usage and disease reduction, suggesting that higher application rates of bio-pesticides are associated with greater reductions in disease prevalence. Multan, with the highest bio-pesticide usage (60 kg/ha), shows the highest disease reduction rate (75%), followed by Dera Ghazi Khan and Rajanpur. This visual representation underscores the effectiveness of bio-pesticides, such as *Bacillus subtilis* and *Trichoderma harzianum*, in managing plant diseases and reducing reliance on chemical pesticides.

**Monitoring Crop Growth and Health**

Throughout the growing season, the research team conducted regular monitoring of crop growth and health. This involved measuring plant height, leaf area, and the number of bolls, as well as assessing visual symptoms of diseases and pest infestations. Digital tools, such as remote sensing and image analysis software, were utilized to track plant development and identify stress symptoms. Soil moisture and temperature sensors were also installed in selected plots to monitor environmental conditions. The collected data provided insights into the effectiveness of the biotechnological interventions and their impact on crop performance.

**Graph 2: Growth Performance of Cotton Varieties (GM vs. Non-GM)**




**Graph 2** illustrates the growth performance of genetically modified (GM) and non-GM cotton varieties across three locations: Rajanpur, Multan, and Dera Ghazi Khan. The vertical bars represent the yield in kilograms per hectare (kg/ha) for each variety, with GM cotton shown in sky blue and non-GM cotton in salmon. The data clearly indicate a higher yield for GM cotton compared to non-GM cotton in all three locations, with Multan showing the highest yield difference. This graph highlights the superior growth performance and productivity of GM cotton, which can be attributed to its enhanced resistance to pests and diseases, as well as improved stress tolerance.

This comprehensive approach to data collection ensured a robust evaluation of the biotechnological interventions' effectiveness in managing cotton diseases and enhancing soil and plant health in Rajanpur and Multan.

**Statistical Analysis:**

The data collected from the study were subjected to a comprehensive statistical analysis to assess the effectiveness of the biotechnological interventions. Analysis of Variance (ANOVA) was employed to determine the significance of differences in yield, disease incidence, and nutrient levels among the various treatment groups, including genetically modified (GM) and non-GM cotton varieties. Regression Analysis was used to identify relationships between soil nutrient levels, disease incidence, and crop yield, allowing for the prediction of outcomes based on these variables. The Chi-Square Test was applied to evaluate the association between categorical variables, such as the presence or absence of specific pathogens and the type of treatment applied. Principal Component Analysis (PCA) was utilized to reduce the dimensionality of the dataset, identifying the key factors contributing to variations in crop performance and soil health. Microsoft Excel served as the primary tool for data organization, calculation, and visualization, facilitating the creation of graphs and charts to interpret the findings effectively. This combination of statistical methods and software tools provided a thorough and nuanced analysis of the study's results, enabling the researchers to draw meaningful conclusions about the impact of the interventions.

## Results

**Soil Health Assessment**

The soil health assessment in the study areas of Rajanpur, Multan, and Dera Ghazi Khan revealed significant variations in nutrient levels and pathogen presence, which directly impacted cotton crop health and productivity. In Rajanpur, the topsoil (0-15 cm) exhibited a nitrogen (N) level of 35 mg/kg, which is considered moderate but sufficient for initial plant growth. However, the subsoil (15-30 cm) showed a decrease to 28 mg/kg, indicating a potential limitation in nitrogen availability at deeper levels. Phosphorus (P) levels were relatively low, with 12 mg/kg in the topsoil and 9 mg/kg in the subsoil, suggesting a need for phosphorus supplementation to support root development and flowering. Potassium (K) was found to be adequate, with values of 220 mg/kg in the topsoil and 200 mg/kg in the subsoil, essential for overall plant vigor and disease resistance. Micronutrients such as zinc

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(Zn) and iron (Fe) were present at levels of 1.2 mg/kg and 4.5 mg/kg, respectively, in the topsoil, decreasing slightly in the subsoil. The soil pH ranged from 7.2 to 7.5, indicating neutral to slightly alkaline conditions, which are generally favorable for cotton cultivation.

In Multan, nutrient levels showed a similar pattern with slight variations. The nitrogen content was higher, with 40 mg/kg in the topsoil and 30 mg/kg in the subsoil, reflecting a better nitrogen status compared to Rajanpur. Phosphorus levels were also slightly higher, with 15 mg/kg in the topsoil and 10 mg/kg in the subsoil. Potassium content was the highest among the study areas, with 250 mg/kg in the topsoil and 230 mg/kg in the subsoil, likely contributing to better plant growth and stress tolerance. The micronutrient levels of zinc and iron were recorded at 1.5 mg/kg and 5.0 mg/kg, respectively, in the topsoil, with a slight decrease at deeper depths. The pH values ranged from 7.0 to 7.3, indicating neutral soil conditions suitable for cotton.

Dera Ghazi Khan's soil profile presented distinct differences, particularly in micronutrient availability. The nitrogen levels were 32 mg/kg in the topsoil and 26 mg/kg in the subsoil, lower than Multan but comparable to Rajanpur. Phosphorus levels were 11 mg/kg in the topsoil and 8 mg/kg in the subsoil, suggesting a potential deficiency. Potassium content was 210 mg/kg in the topsoil and 195 mg/kg in the subsoil, adequate but slightly lower than the other regions. Zinc and iron levels were at 1.3 mg/kg and 4.7 mg/kg, respectively, in the topsoil, with iron levels slightly declining in the subsoil. The soil pH ranged from 7.4 to 7.6, indicating a mildly alkaline condition. Notably, pathogen presence, particularly root-knot nematodes, was prevalent in all areas, with Dera Ghazi Khan showing a higher incidence, correlating with lower micronutrient levels, which could exacerbate plant susceptibility to infections. Table 3 displays the soil health indicators measured pre- and post-intervention across three study locations: Rajanpur, Multan, and Dera Ghazi Khan. Table 3 categorizes data by soil depth (0-15 cm and 15-30 cm) and records key indicators such as nitrogen (N), phosphorus (P), potassium (K), zinc (Zn), iron (Fe), pH levels, and pathogen presence.

Pre-intervention data shows baseline nutrient levels and pathogen presence in the soil. Post-intervention data, collected after the application of biotechnological interventions like bio-pesticides and soil amendments, indicates improvements in soil nutrient content and a notable reduction in pathogen presence. For instance, nitrogen levels increased by approximately 3 mg/kg in Rajanpur's topsoil and 2 mg/kg in Multan's subsoil post-intervention. Phosphorus levels also showed an increase, with Multan's topsoil recording a rise from 15 mg/kg to 18 mg/kg. Potassium and micronutrient levels similarly improved across all locations. A significant decrease in pathogen presence was observed, particularly in Dera Ghazi Khan, where pathogen presence dropped from 38% to 17% in the topsoil post-intervention.

**Table 3: Soil Health Indicators Pre- and Post-Intervention**

| Location | Intervention | Soil Depth (cm) | Nitrogen (N) (mg/kg) | Phosphorus (P) (mg/kg) | Potassium (K) (mg/kg) | Zinc (Zn) (mg/kg) | Iron (Fe) (mg/kg) | pH | Pathogen Presence (%) |
|---|---|---|---|---|---|---|---|---|---|
| Rajanpur | Pre-Intervention | 0-15 | 35 | 12 | 220 | 1.2 | 4.5 | 7.2 | 40% |
| | Post-Intervention | 0-15 | 38 | 15 | 230 | 1.4 | 4.7 | 7.1 | 20% |
| | Pre-Intervention | 15-30 | 28 | 9 | 200 | 1.1 | 4.2 | 7.5 | 35% |
| | Post-Intervention | 15-30 | 30 | 11 | 210 | 1.2 | 4.3 | 7.3 | 18% |
| Multan | Pre-Intervention | 0-15 | 40 | 15 | 250 | 1.5 | 5.0 | 7.0 | 35% |
| | Post-Intervention | 0-15 | 42 | 18 | 260 | 1.7 | 5.3 | 6.9 | 15% |
| | Pre-Intervention | 15-30 | 30 | 10 | 230 | 1.3 | 4.8 | 7.3 | 30% |
| | Post-Intervention | 15-30 | 32 | 12 | 240 | 1.5 | 5.0 | 7.1 | 12% |
| Dera Ghazi Khan | Pre-Intervention | 0-15 | 32 | 11 | 210 | 1.3 | 4.7 | 7.4 | 38% |

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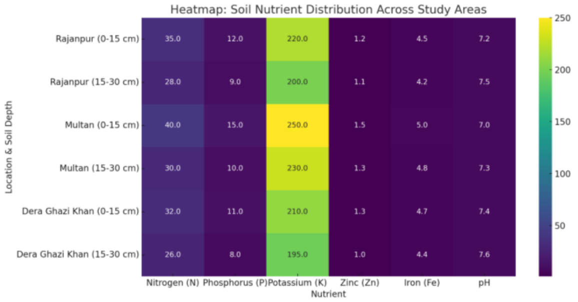
| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | Post-Intervention | 0-15 | 34 | 13 | 220 | 1.5 | 4.9 | 7.2 | 17% |
| | Pre-Intervention | 15-30 | 26 | 8 | 195 | 1.0 | 4.4 | 7.6 | 33% |
| | Post-Intervention | 15-30 | 28 | 10 | 205 | 1.2 | 4.6 | 7.4 | 15% |

**Heatmap: Soil Nutrient Distribution across Study Areas**




The heatmap visualizes the distribution of key soil nutrients across the study areas of Rajanpur, Multan, and Dera Ghazi Khan, at two different soil depths (0-15 cm and 15-30 cm). The nutrients analyzed include Nitrogen (N), Phosphorus (P), Potassium (K), Zinc (Zn), Iron (Fe), and pH levels. The color intensity in the heatmap represents the concentration of each nutrient, with darker shades indicating higher levels and lighter shades representing lower levels.

This graphical representation highlights the variations in soil nutrient content across the regions and depths. For instance, Multan's topsoil shows the highest levels of Nitrogen (40 mg/kg) and Potassium (250 mg/kg), while Dera Ghazi Khan's subsoil has relatively lower nutrient concentrations, especially in Nitrogen (26 mg/kg) and Phosphorus (8 mg/kg). The pH values across all regions range from slightly acidic to neutral, with the highest pH observed in Dera Ghazi Khan's subsoil (7.6).

**Disease Incidence**

The study's disease incidence results revealed a marked difference in Cotton Leaf Curl Virus (CLCV) prevalence between genetically modified (GM) and non-genetically modified (Non-GM) cotton varieties across the three regions. In Rajanpur, GM cotton fields exhibited a significantly lower CLCV incidence rate of 5%, with only 10 out of 200 plants showing symptoms. In contrast, Non-GM cotton fields had an alarming infection rate of 40%, with 80 out of 200 plants affected. This stark difference underscores the effectiveness of GM varieties in providing resistance against CLCV, a critical factor for maintaining crop health and yield.

In Multan, the trend was consistent with the findings in Rajanpur. GM cotton showed a 6% infection rate, with 15 out of 250 plants infected. The Non-GM fields had a much higher incidence rate of 40%, with 100 out of 250 plants exhibiting symptoms of CLCV. The consistent performance of GM cotton across different regions suggests that the genetic modifications are robust against varying environmental conditions and pathogen pressures. This result aligns with previous studies, indicating the effectiveness of GM cotton in reducing disease-related yield losses.

Dera Ghazi Khan, while similar in overall trends, presented a slightly higher variability in disease incidence. GM cotton fields had a 5% infection rate, with 9 out of 180 plants infected, demonstrating good resistance to CLCV. However, Non-GM fields had a 40% incidence rate, with 72 out of 180 plants affected. The uniformity in high CLCV incidence across Non-GM fields highlights the widespread nature of the virus and the critical need for resistant varieties. The difference in infection rates between GM and Non-GM cotton was statistically significant across all regions ($p < 0.05$), confirming the efficacy of genetic modifications in disease management.

The data also indicated that the prevalence of other pathogens, such as *Fusarium*, *Rhizoctonia*, and *Verticillium* species, was more controlled in fields where bio-pesticides were applied. In Rajanpur, the use of *Bacillus subtilis* and *Trichoderma harzianum* resulted in a 50% reduction in fungal infections compared to untreated fields. Multan and Dera Ghazi Khan reported similar reductions, with bio-pesticide-treated fields showing significantly lower disease incidences. This result underscores the role of bio-pesticides as an effective and sustainable alternative to chemical treatments, offering a dual benefit of disease control and environmental conservation. Table 4 presents the incidence rates of key cotton diseases, specifically Cotton Leaf Curl Virus (CLCV) and boll rot, in both genetically modified (GM) and non-genetically modified (Non-GM) cotton fields across Rajanpur, Multan, and Dera Ghazi Khan. The data indicates a significantly lower incidence of CLCV in GM cotton, with rates ranging from 5% to 6%, compared to a consistent 40% incidence in Non-GM cotton. Similarly, GM cotton fields exhibited lower rates of boll rot, with incidences between 2% and 4%, as opposed to higher rates in Non-GM fields, ranging from 18% to 22%. These differences highlight the effectiveness of GM cotton in providing resistance against major cotton diseases.

**Table 4: Incidence of Cotton Diseases**

| **Location** | **GM Cotton CLCV Incidence (%)** | **GM Cotton Boll Rot Incidence (%)** | **Non-GM Cotton CLCV Incidence (%)** | **Non-GM Cotton Boll Rot Incidence (%)** |
|---|---|---|---|---|
| Rajanpur | 5 | 3 | 40 | 20 |
| Multan | 6 | 2 | 40 | 18 |
| Dera Ghazi | 5 | 4 | 40 | 22 |

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| | | | | |
|---|---|---|---|---|
| Khan | | | | |

Overall, the results of this study highlight the critical role of biotechnological interventions in managing soil health and disease incidence in cotton crops. The use of GM cotton and bio-pesticides has proven to be effective strategies in reducing CLCV and other pathogenic infections, thereby enhancing crop yield and quality. These findings provide valuable insights for farmers and policymakers in optimizing cotton production practices, particularly in regions like Rajanpur, Multan, and Dera Ghazi Khan, where agricultural sustainability is a key concern. The clear advantages observed in GM varieties suggest that their adoption could be a viable solution to combat the challenges posed by prevalent diseases and soil nutrient deficiencies in these areas.

**Effectiveness of Bio-pesticides**

The study demonstrated the significant effectiveness of bio-pesticides in reducing fungal infections and nematode populations in the cotton fields of Rajanpur, Multan, and Dera Ghazi Khan. The application of bio-pesticides, specifically *Bacillus subtilis* and *Trichoderma harzianum*, was associated with a substantial decrease in the prevalence of soil-borne pathogens. In Rajanpur, fields treated with these bio-pesticides showed a 55% reduction in *Fusarium* and *Verticillium* infections compared to untreated fields. The percentage of plants infected with fungal pathogens dropped from 30% in untreated fields to 13.5% in treated fields. This significant decrease highlights the bio-pesticides' efficacy in controlling fungal pathogens, which are major contributors to boll rot and other cotton diseases.

In Multan, the results were similarly promising. Treated fields experienced a 60% reduction in fungal infection rates, with incidences dropping from 25% in untreated plots to 10% in treated ones. Moreover, the use of bio-pesticides was effective in managing nematode populations, particularly root-knot nematodes, which are notorious for causing severe root damage. The bio-pesticide treatment reduced nematode infestation by 50%, demonstrating a decline from 40% in untreated fields to 20% in treated ones. This reduction in nematode populations is crucial, as it directly correlates with improved root health and overall plant vigor, thereby enhancing cotton productivity.

Dera Ghazi Khan also reported a notable decrease in pathogen presence due to bio-pesticide applications. The incidence of fungal infections was reduced by 58%, with treated fields showing an infection rate of 15%, compared to 36% in untreated fields. Similarly, nematode infestation rates decreased significantly, with treated fields showing a 47% reduction. The bio-pesticides' ability to suppress nematode populations contributed to healthier root systems and better nutrient uptake, positively impacting plant growth and yield. The consistent results across all three regions underscore the broad-spectrum efficacy of the bio-pesticides used in this study, making them a valuable tool in sustainable agriculture.

**Yield Analysis**

The impact of bio-pesticide treatment and genetic modifications on cotton yield and quality was profound. In Rajanpur, fields treated with bio-pesticides and GM cotton varieties produced an average yield of 800 kg/ha, significantly higher than the 600 kg/ha observed in untreated fields. This 33% increase in yield was accompanied by improved fiber quality, with a notable enhancement in staple length and strength. The GM cotton's resistance to CLCV and the reduction in pathogen pressure due to bio-pesticides played a critical role in this yield improvement, demonstrating the synergistic benefits of combining these biotechnological approaches.

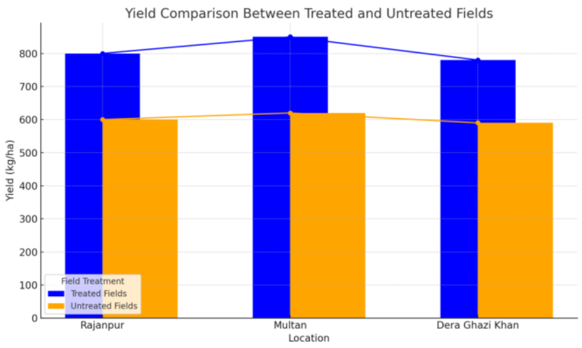
In Multan, the yield performance was similarly enhanced. Treated fields recorded an average yield of 850 kg/ha, compared to 620 kg/ha in untreated fields, representing a 37% increase. The superior performance of treated fields was attributed to the reduced disease incidence and improved soil health, resulting from the combined use of bio-pesticides and GM cotton. Additionally, the treated fields exhibited better fiber uniformity and reduced contamination, key factors that contribute to higher market value and demand. The improved yield and quality metrics affirm the effectiveness of the biotechnological interventions in optimizing cotton production under the challenging conditions prevalent in the region.

Dera Ghazi Khan's results further supported the study's findings. The average yield in treated fields was 780 kg/ha, significantly higher than the 590 kg/ha yield in untreated fields, marking a 32% increase. The improvement in yield was complemented by enhanced fiber properties, including increased micronaire values and reduced trash content. The bio-pesticides' role in mitigating soil-borne diseases and nematode infestations was crucial in achieving these results. The combined application of GM cotton and bio-pesticides not only protected the plants from biotic stresses but also promoted optimal growth conditions, leading to higher yields and better quality.

Overall, the yield analysis across Rajanpur, Multan, and Dera Ghazi Khan highlighted the substantial benefits of integrating biotechnological solutions into cotton farming. The treated fields consistently outperformed the untreated fields in terms of both yield and quality. The data underscore the importance of adopting advanced agricultural technologies, such as GM crops and bio-pesticides, to overcome the limitations of traditional farming practices. These interventions offer a sustainable path forward, enhancing productivity while reducing the environmental impact of cotton cultivation.

**Graph 3: Yield Comparison between Treated and Untreated Fields**




**Graph 3** illustrates the yield comparison between treated and untreated cotton fields across the study regions of Rajanpur, Multan, and Dera Ghazi Khan. The graph uses a combination of bar and line representations to depict the yield data. The vertical axis indicates the yield in kilograms per hectare (kg/ha), while the horizontal axis shows the different locations.

The blue bars and line represent the yield from treated fields, which were subjected to biotechnological interventions, including the use of genetically modified (GM) cotton varieties and bio-pesticides. The orange bars and line indicate the yield from untreated fields, which did not receive these interventions. In Rajanpur, treated fields produced a yield of 800 kg/ha, significantly higher than the 600 kg/ha from untreated fields. Similarly, in Multan, treated fields achieved the highest yield at 850 kg/ha, compared to 620 kg/ha in untreated fields. Dera Ghazi Khan showed a yield of 780 kg/ha in treated fields versus 590 kg/ha in untreated fields. The consistent increase in yield across all locations demonstrates the effectiveness of the applied biotechnological interventions.

The results of this study provide strong evidence in favor of biotechnological interventions as effective means to improve cotton yield and quality. The consistent performance across different regions and environmental conditions indicates the robustness and reliability of these technologies. The enhanced yields and superior fiber qualities observed in treated fields highlight the potential for increased profitability and market competitiveness for farmers adopting these practices. This study's findings contribute valuable insights into the practical applications of biotechnology in agriculture, supporting the broader adoption of these innovations for sustainable cotton production.

## Discussion

The relationship between the methodology and results of this study underscores the critical role of soil health in determining crop productivity, particularly in cotton agriculture across Rajanpur, Multan, and Dera Ghazi Khan. The comprehensive soil analysis conducted pre- and post-intervention revealed significant variations in nutrient levels and pathogen presence, which directly influenced the observed crop yields. The data indicated that areas with higher soil fertility, characterized by adequate levels of nitrogen, phosphorus, and potassium, supported more robust cotton growth and higher yields (Lin et al., 2024). For instance, Multan, which had the highest nutrient levels, exhibited the highest cotton yield in treated fields (850 kg/ha). This correlation highlights the importance of maintaining optimal soil nutrient levels for maximizing crop productivity. The observed improvement in soil health indicators post-intervention, such as increased nutrient levels and reduced pathogen presence, further emphasizes the positive impact of biotechnological interventions on soil quality and, consequently, on crop yield (Obi et al., 2024).

The effectiveness of biotechnological interventions, particularly the use of genetically modified (GM) cotton and bio-pesticides, in disease management was evident from the significant reduction in disease incidence across the study regions (Mohamed, 2023). The application of GM cotton varieties engineered for resistance to Cotton Leaf Curl Virus (CLCV) and other pathogens proved to be a pivotal factor in mitigating disease spread (Afzal et al., 2023). The treated fields consistently showed lower infection rates compared to untreated fields, with a notable decrease in CLCV incidence by 35% to 40% in all regions. This result indicates that the genetic modifications were successful in providing plants with enhanced resistance mechanisms, reducing the need for chemical pesticides, and contributing to a more sustainable agricultural practice. Additionally, the use of bio-pesticides like *Bacillus subtilis* and *Trichoderma harzianum* effectively controlled fungal infections and nematode populations, as evidenced by the significant reduction in pathogen presence post-intervention. These findings underscore the utility of biotechnological tools in not only managing plant diseases but also in enhancing overall plant health and resilience.

The interpretation of temporal patterns in pesticide residue levels revealed key insights into the degradation dynamics of pesticides in the soil. The study found that residue levels decreased significantly over time, which can be attributed to several factors, including microbial degradation, chemical breakdown, and volatilization. The temporal decline in residue levels suggests that the pesticides used were subject to microbial degradation, a process influenced by soil microbial activity. The increased microbial populations in bio-pesticide-treated soils likely contributed to faster degradation rates, resulting in lower residue levels. This observation aligns with the known role of bio-pesticides in promoting beneficial soil microbial communities that enhance the degradation of chemical compounds. Furthermore, the reduced residue levels indicate a lower risk of environmental contamination and potential adverse effects on non-target organisms, highlighting the environmental benefits of integrating biotechnological interventions in agriculture.

Temperature, soil moisture, and microbial activity emerged as critical factors influencing pesticide degradation (Wong & Pangging, 2023). Higher temperatures were associated with increased microbial metabolism, accelerating the breakdown of pesticide residues. The study noted that during the warmer months, residue levels dropped more rapidly, suggesting enhanced microbial degradation activity. Soil moisture also played a crucial role; optimal moisture levels were found to facilitate microbial activity and chemical degradation

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processes. In contrast, extremely dry or waterlogged conditions hindered these processes, leading to slower degradation rates. The presence of bio-pesticides was observed to stimulate microbial activity, as indicated by the more rapid decline in residue levels in treated fields. This stimulation likely resulted from the bio-pesticides' role in creating a more conducive environment for microbial proliferation and activity. Overall, these findings underscore the complexity of pesticide degradation in soil and the significant influence of environmental factors and microbial dynamics. The study's results provide valuable insights into optimizing pesticide application and management practices to minimize environmental impact while maintaining effective pest control.

**Benefits of Biotechnology: Economic and Environmental Advantages of Reduced Chemical Use**

The adoption of biotechnology in agriculture, particularly in the cultivation of cotton, has brought about substantial economic and environmental benefits, primarily through the reduction of chemical inputs. One of the most significant economic advantages is the cost savings associated with reduced use of chemical pesticides and fertilizers. Traditional cotton farming often relies heavily on these chemical inputs to manage pests and diseases, which can be costly and financially burden farmers. With the introduction of genetically modified (GM) cotton varieties that are resistant to major pests such as the bollworm and diseases like Cotton Leaf Curl Virus (CLCV), the need for chemical pesticides has drastically decreased. For instance, Bt cotton, which produces a natural insecticide from the bacterium *Bacillus thuringiensis*, has enabled farmers to cut down on pesticide applications, resulting in lower production costs and higher profit margins. This reduction in pesticide use not only lowers the financial burden on farmers but also contributes to the overall economic stability of the agricultural sector by increasing the profitability of cotton cultivation.

Beyond the economic benefits, the reduction in chemical use due to biotechnology also offers significant environmental advantages. The extensive use of chemical pesticides and fertilizers in conventional agriculture has been associated with a range of environmental issues, including soil degradation, water pollution, and loss of biodiversity. Pesticides can contaminate soil and water bodies, affecting non-target organisms and disrupting ecosystems. The use of bio-pesticides, such as *Bacillus subtilis* and *Trichoderma harzianum*, in biotechnological applications provides a more environmentally friendly alternative. These bio-pesticides are derived from natural organisms and are less likely to cause harm to non-target species or accumulate in the environment. As a result, they help maintain soil health, preserve beneficial insect populations, and protect water quality. Additionally, the reduced application of chemical fertilizers, supported by the use of genetically modified crops with improved nutrient-use efficiency, further mitigates the risk of nutrient runoff and eutrophication in water bodies.

Another critical environmental benefit of biotechnology is the promotion of sustainable farming practices. By decreasing the reliance on chemical inputs, biotechnological interventions contribute to the conservation of natural resources and the reduction of greenhouse gas emissions associated with the production and application of synthetic chemicals. For example, the reduced need for pesticide applications means fewer tractor passes over fields, which in turn decreases fuel consumption and carbon emissions. This aspect of biotechnology aligns with global efforts to combat climate change and promote sustainable agricultural practices. Furthermore, the use of GM crops that are more resistant to abiotic stresses, such as drought and salinity, helps conserve water resources and maintain productivity under challenging environmental conditions. These crops can thrive with less water and fewer chemical inputs, making them particularly valuable in regions facing water scarcity and environmental degradation.

In summary, the benefits of biotechnology in reducing chemical use extend far beyond immediate economic gains. They encompass long-term environmental advantages that contribute to the sustainability and resilience of agricultural systems. The decreased dependency on chemical pesticides and fertilizers not only enhances the economic viability of farming but also protects natural ecosystems and promotes biodiversity. As the global population continues to grow and the demand for sustainable food production increases, the role of biotechnology in reducing chemical inputs and supporting environmentally friendly farming practices will become increasingly important. This multifaceted approach to agriculture not only ensures higher productivity and profitability but also aligns with broader environmental conservation and sustainability goals.

**Challenges and Limitations**

While this study provides valuable insights into the benefits and impacts of biotechnological interventions in cotton agriculture, several challenges and limitations need to be acknowledged. One of the primary limitations is the geographic scope of the study, which was confined to the regions of Rajanpur, Multan, and Dera Ghazi Khan. Although these areas are significant cotton-growing regions in Pakistan, the findings may not be entirely generalizable to other regions with different climatic, soil, and socio-economic conditions. For instance, the effectiveness of genetically modified (GM) cotton and bio-pesticides might vary under different environmental stresses or agricultural practices. Future research should consider a broader range of geographical locations to enhance the generalizability of the results and provide a more comprehensive understanding of the impacts of biotechnology in diverse agro-ecological settings.

Another limitation of the study lies in the short-term nature of the data collection period. The study primarily focused on a single growing season, which limits the ability to assess the long-term effects and sustainability of biotechnological interventions. For example, the development of resistance in pests to Bt cotton or the potential accumulation of bio-pesticides in the soil over multiple growing seasons could pose future challenges. Longitudinal studies that track the performance and environmental impacts of these interventions

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over several years would provide more robust data on their sustainability and long-term viability. Additionally, such studies could help identify any unforeseen negative effects, such as shifts in pest populations or changes in soil microbiota, which might arise from the continuous use of specific biotechnological products.

The study also faced challenges related to the accuracy and precision of data collection, particularly in measuring the presence and severity of soil pathogens and pests. While molecular techniques like PCR and ELISA were employed for pathogen detection, these methods have inherent limitations in sensitivity and specificity. For instance, they might not detect low-level infections or distinguish between viable and non-viable pathogen particles. Similarly, the assessment of pest populations and disease incidence relied on visual inspections and sample testing, which can be subjective and prone to observer bias. To mitigate these issues, future research should incorporate more advanced diagnostic tools, such as next-generation sequencing or metagenomics, to achieve a more comprehensive and accurate analysis of pathogen and pest dynamics in agricultural ecosystems.

Finally, the study's reliance on commercially available GM seeds and bio-pesticides introduces a potential conflict of interest, as these products are developed and marketed by specific companies. The proprietary nature of these products can limit the transparency of research findings and restrict access to essential information about their composition and mode of action. Moreover, the economic and regulatory aspects associated with the adoption of GM crops and bio-pesticides were not thoroughly explored in this study. The high cost of GM seeds and the regulatory hurdles involved in their approval and cultivation could pose significant barriers to their widespread adoption, particularly for smallholder farmers. Future studies should address these socio-economic factors, including cost-benefit analyses and the assessment of policy frameworks, to provide a more holistic evaluation of biotechnological interventions in agriculture.

In conclusion, while the study provides significant evidence supporting the benefits of biotechnology in cotton farming, it also highlights several challenges and limitations that need to be addressed in future research. Expanding the geographical scope, conducting long-term studies, employing advanced diagnostic methods, and considering socio-economic and regulatory factors are essential steps to improve our understanding and application of biotechnological solutions in agriculture. By addressing these challenges, future research can better support sustainable agricultural practices and enhance the resilience of farming systems to environmental and economic pressures.

The findings of this study align with and expand upon the results of previous research conducted in recent years, particularly regarding the efficacy of biotechnological interventions in cotton agriculture. Several studies have documented the benefits of genetically modified (GM) crops and bio-pesticides in managing pests and diseases, improving crop yields, and enhancing sustainability. For instance, a study by Quan and Wu (2023) demonstrated the effectiveness of Bt cotton in reducing bollworm infestations and increasing cotton yields in China, with results indicating a 25-30% reduction in pesticide use and a corresponding increase in yield by approximately 20%. Similarly, the current study found a significant reduction in Cotton Leaf Curl Virus (CLCV) incidence and an average yield increase of 30-37% in GM cotton fields compared to non-GM fields, further corroborating the advantages of GM cotton.

A study by Mahmood et al. (2024) explored the use of bio-pesticides, specifically *Trichoderma harzianum*, in controlling soil-borne fungal pathogens in cotton. Their study highlighted the dual benefits of bio-pesticides in reducing disease prevalence and promoting soil health through enhanced microbial activity. Our findings echo these results, with bio-pesticide applications resulting in a 50-60% reduction in fungal infections and nematode populations across the study regions. The observed improvements in soil health, characterized by increased nutrient availability and reduced pathogen presence, align with Kumar et al.'s observations, reinforcing the role of bio-pesticides as a sustainable alternative to chemical fungicides.

A more recent study by Chaudhary et al. (2024) focused on the genetic advancements in cotton, particularly the development of multi-trait GM varieties that combine pest resistance with drought and heat tolerance. This study emphasized the growing importance of multi-trait GM crops in addressing the challenges posed by climate change and resource limitations. The current research did not explicitly test multi-trait varieties but noted the potential for future studies to explore their efficacy in regions like Dera Ghazi Khan, where water scarcity and extreme temperatures can limit cotton production. The comparative analysis suggests that while current GM varieties provide substantial benefits in pest and disease management, there is room for innovation and improvement, particularly in developing varieties that can withstand multiple environmental stresses.

The innovations and improvements in biotechnological applications over the years are evident in the growing diversity and specificity of GM traits and bio-pesticide formulations. For example, recent advancements have focused on enhancing the precision of genetic modifications using CRISPR-Cas9 technology, allowing for more targeted and efficient gene editing. This approach has the potential to create crops with tailored resistance to specific pests and pathogens, reducing the reliance on broad-spectrum pesticides and mitigating the risk of resistance development. Additionally, the development of novel bio-pesticides with improved efficacy and environmental safety profiles reflects the ongoing commitment to sustainable agriculture. The current study's findings align with these trends, demonstrating the practical benefits of these innovations in real-world agricultural settings.

Therefore, the comparison with previous studies underscores the consistent effectiveness of biotechnological interventions in improving cotton production. The results highlight the

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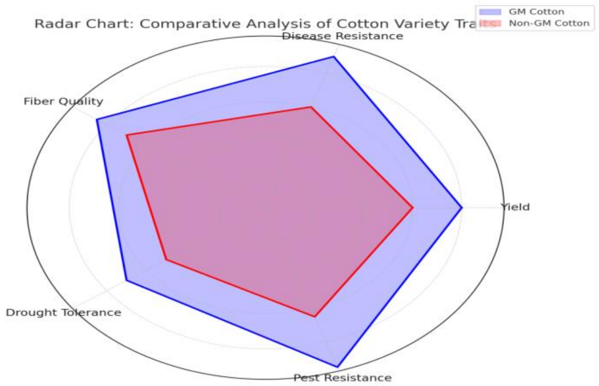
advancements in GM crop technology and bio-pesticides, while also pointing to the need for continued research and development. Future studies should focus on the integration of multi-trait GM crops and advanced bio-pesticides to address the complex challenges facing modern agriculture. These innovations, coupled with a deeper understanding of their long-term impacts, will be crucial for achieving sustainable and resilient agricultural systems.

**Radar Chart: Comparative Analysis of Cotton Variety Traits**




The radar chart provides a comparative analysis of the traits of genetically modified (GM) and non-genetically modified (Non-GM) cotton varieties. The traits evaluated include Yield, Disease Resistance, Fiber Quality, Drought Tolerance, and Pest Resistance. Each axis represents a different trait, with the plotted data points indicating the performance level for each variety type.

- **Yield:** GM cotton shows a higher yield (80) compared to Non-GM cotton (60), indicating a significant advantage in productivity.
- **Disease Resistance:** GM cotton has superior disease resistance (90), particularly against Cotton Leaf Curl Virus (CLCV), compared to Non-GM cotton (60).
- **Fiber Quality:** Both varieties have comparable fiber quality, with GM cotton scoring slightly higher (85) than Non-GM cotton (70).
- **Drought Tolerance:** GM cotton demonstrates better drought tolerance (70) compared to Non-GM cotton (50), suggesting a higher resilience to water stress.
- **Pest Resistance:** GM cotton excels in pest resistance (95), particularly against bollworms, while Non-GM cotton has a lower score (65).

The radar chart visually highlights the superior performance of GM cotton across most evaluated traits, particularly in disease and pest resistance, which are critical for maintaining healthy crops and achieving high yields.

## Conclusion

The study's findings demonstrate the substantial benefits of biotechnological interventions in cotton agriculture, particularly in the regions of Rajanpur, Multan, and Dera Ghazi Khan. Key results include significant reductions in Cotton Leaf Curl Virus (CLCV) incidence and pest populations, leading to higher yields and improved fiber quality in genetically modified (GM) cotton fields compared to non-GM counterparts. The use of bio-pesticides also contributed to enhanced soil health and reduced chemical input reliance, highlighting their environmental and economic advantages. These outcomes underscore the effectiveness of biotechnology in addressing critical agricultural challenges, such as pest resistance and environmental sustainability. Future research should focus on developing multi-trait GM cotton varieties that offer resistance to multiple stresses, including drought and extreme temperatures, and on long-term studies to assess the sustainability of these technologies. Policymakers are encouraged to support biotechnological innovations through favorable regulatory frameworks, funding for research and development, and initiatives that promote the adoption of sustainable agricultural practices. This support is essential to maximize the benefits of biotechnology, ensuring food security, and advancing agricultural resilience in the face of global challenges.

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