

INVESTIGATION ON PREVALENCE OF FUSARIUM WILT OF SUGARCANE IN DISTRICT TANDO ALLAHYAR AND ITS *IN VITRO* MANAGEMENT

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ABSTRACT

Sugarcane is a major cash crop and plays an important role in the economy of many countries, including Pakistan. It is one of the most vital and globally significant crops which provide nearly 85% of the sugar consumed worldwide. The high amount of water and sugars (such as glucose, fructose, sucrose, and raffinose) present in sugarcane Juice, it also contains various vitamins (B1, B2, B3, B5, B6, B7, C and D) and minerals. Its per yield in Pakistan is quite low as compared to other countries of the world due to various factors, including diseases. Sugarcane crops suffer approximately 100 diseases worldwide, diseases can cause significant losses in sugarcane yield and quality, leading to economic losses for farmers and the industry. Among the diseases the fusarium wilt stands out as a significant concern for sugarcane farmers. Keeping in view the importance and losses caused by fusarium wilt of sugarcane, the present studies were conducted on the investigation and prevalence of fusarium wilt of sugarcane in District Tando Allahyar and its *in vitro* management. A survey was carried out from major sugarcane growing areas Missan Wadi, village Haji Muhammad Ibrahim Dars, Village Masoo Bozdar, Village Dad Khan Jarwar and Village Qazi Noor Muhammad Laghari. The highest disease incidence % was found at Missan Wadi 36% followed by Haji Muhammad Ibrahim Dars 28%. The minimum disease incidence % was found at Qazi Noor Muhammad Laghari 18%. The sampling was carried out from each location during survey. The tissue isolation technique was performed for each diseased sample. The isolated fungus was identified based on their morphological characteristics. The isolated fungus *Fusarium Sacchari* was managed by using different chemicals and plant extracts *in vitro* conditions by food poison technique. Highly significant difference was obtained between the tested treatments. The fungicides such as Pyraclostrobin, Clothianidin and Difenconazole were found highly effective at 15 ppm. while the Azoxystrobin and Oxathiapiprolin fungicides were found less effective. Among the tested plant extracts the Bitter apple leaves (*Citrullus colocynthis*) and Giant calotrope (*Calotropis gigantea*) were found highly effective whereas, the Moringa (*Moringa oleifera*) and Thorn apple leaves (*Datura stramonium*) were found moderately effective. The Cinnamon (*Cinnamomum verum*) was found lowest effective. The overall results shows that the chemicals are highly effective as compare to plant extracts. Based on the present findings it is concluded that Ready super (Pyraclostrobin) fungicide and Bitter apple leaves (*Citrullus colocynthis*) extracts are the most effective product, so they are highly recommended for the management of fusarium wilt of Sugarcane.

Key-words: sugarcane, fungal disease, Fusarium wilt, Tando Allah Yar, plant extracts.

INTRODUCTION

Sugarcane (*Saccharum officinarum* L.) is a globally significant crop, supplying approximately 85% of the world's sugar (FAOSTAT, 2023). In recent years, its cultivation has also expanded into the bioenergy sector, with energy canes being developed for biofuel and electricity production (Corcoran *et al.*, 2020). Sugarcane farming holds substantial agricultural, economic, environmental, and social value due to its wide range of applications and byproducts, which contribute to profitable diversification (Manzoor *et al.*, 2023). Historically, sugarcane has played a vital role in the socioeconomic advancement of human societies and is currently cultivated in more than 105 countries.

Traditionally, sugarcane has been utilized for the production of jaggery, brown sugar, and refined sugar (Raza *et al.*, 2023). In recent times, the focus of crop improvement has shifted from solely enhancing sucrose content to increasing biomass and fermentable sugars for renewable energy applications. Among its industrial uses, sugarcane bagasse—a fibrous byproduct—is gaining prominence as a key feedstock for bioenergy and biofuel production. In Pakistan, approximately 3.5 million metric tons of bagasse are consumed annually, with about 30% recovery from sugar mills, where it is used to generate steam energy (Wani *et al.*, 2023).

Sugarcane is predominantly cultivated in tropical and subtropical regions, with Brazil, India, Thailand, China, and Pakistan collectively producing nearly 70% of the world's sugar. In Pakistan, sugarcane is grown on around

1.06 million hectares, with an average yield of 55 tons of cane and 5.5 tons of sugar per hectare. It is the second-largest cash crop, contributing about 3.6% to the national GDP. While domestic demand for refined sugar stands at approximately 4.5 million tons, the country's 85 sugar mills have an installed production capacity of 7.5 million tons, creating an export potential of 3.5 million tons (Qureshi *et al.*, 2020).

Globally, sugarcane is cultivated on 26.35 million hectares, with a total production of over 1.85 billion tons. In Pakistan, 88.65 million tons are produced from 1.26 million hectares (FAOSTAT, 2023). It is also one of the most productive crops in terms of biomass. The juice extracted from sugarcane is rich in various sugars—sucrose, glucose, fructose, and raffinose—as well as water (approximately 65%). It also contains essential vitamins (B1–B7, C, and D), minerals (e.g., potassium, magnesium, iron), organic acids, polyphenols, flavonoids, fatty acids, and carotenoids (Panigrahi *et al.*, 2021).

Sugarcane plays a crucial role in national economies by generating significant revenue through taxes and tariffs (Farooq *et al.*, 2019). Technological advancements have expanded its utility beyond sugar production to raw materials for various industries. However, despite being a robust crop, sugarcane is highly vulnerable to biotic stresses, particularly fungal pathogens. Over 100 fungal species are known to infect sugarcane, with diseases such as red rot, whip smut, sugarcane mosaic virus, pineapple disease, and Fusarium wilt posing major threats to its productivity (Rajput *et al.*, 2021). Disease-related yield losses can range from 10% to 50%, highlighting the importance of integrated disease management (Farber *et al.*, 2019).

Among these, *Fusarium* wilt ranks as one of the most devastating diseases after red rot. It significantly reduces yield in sugarcane-growing regions, including Bangladesh (Paul *et al.*, 2022). Yield losses can reach up to 10 tons per hectare, with infected fields displaying dry or dead canes post-harvest (Parthasarathy, 1972). Affected plants exhibit fewer tillers and show symptoms such as yellowing, drying, and eventual wilting of leaves, particularly in the crown area (Agnihotri and Singh, 1989; Viswanathan, 2013). Early-stage symptoms are often overlooked, but severe infections can lead to notable mortality in young seedlings.

The disease is caused by *Fusarium sacchari*, a soil- and set-borne pathogen identified through recent studies (Poongothai *et al.*, 2014a,b). Disease manifestation typically aligns with monsoon and post-monsoon seasons (Viswanathan, 2013). Because *F. sacchari* is an aggressive and persistent soil inhabitant, controlling the disease through cultural practices alone has proven inadequate (Sharma *et al.*, 2010). While fungicides offer quick and effective solutions, their cost and environmental impact necessitate alternative strategies (Moosa *et al.*, 2017). As a result, significant efforts have been directed toward the use of antifungal plant extracts, which offer sustainable, eco-friendly, and economically viable disease control options (Jaiman and Jain, 2010; Khalikar *et al.*, 2011; Rekha *et al.*, 2012; Abdul *et al.*, 2015).

Plant-derived compounds, rich in bioactive secondary metabolites, have shown promise in inhibiting fungal pathogens, including those causing wilt diseases in sugarcane (Sana *et al.*, 2017; Akhtar and Javai, 2018; Obongoya *et al.*, 2010; Rafiq *et al.*, 2021). Incorporating plant extracts into integrated disease management (IDM) frameworks offers a sustainable solution with minimal environmental impact.

Given sugarcane's critical role in food security, economic stability, and industrial growth, protecting it from devastating diseases such as Fusarium wilt is essential. In Pakistan, where sugarcane supports millions of livelihoods, effective disease management is crucial for maintaining crop productivity, ensuring a stable supply for both domestic and export markets, and promoting the long-term sustainability of agriculture (Shoaib *et al.*, 2018).

MATERIALS AND METHODS

Observation of disease prevalence of sugarcane wilt at different locations of District Tando Allahyar

Different locations Viz; Missan Wadi, village Haji Mohammad Ibrahim Dars, Village Masoo Bozdar, Village Dad Khan Jarwar and Village Qazi Noor Muhammad Laghari in district Tando Allahyar of Sindh were surveyed by zigzag method to record the sugarcane wilt disease incidence %. The samples showing the wilt symptoms were also collected from each location in paper envelope and brought in the mycology laboratory of Plant Disease Research Institute Agriculture research Centre Tando Jam. During survey the disease incidence % was determined of 50 plants randomly from each location by using the following mathematical disease incidence (%) formula.

$$\text{Disease incidence (\%)} = \frac{\text{Number of diseased plants}}{\text{Total number of observed plants}} \times 100$$

Isolation and identification of the causal fungus

Using the Potato Dextrose Agar medium and the tissue isolation method outlined by (Agrios *et al.*, 2005), the Fusarium wilt pathogen was isolated. During this procedure, infected sugarcane setts were divided into tiny pieces

that were between 0.5 to 2.0 cm in after washing these parts under tap water they were surface sterilized using a 0.1% mercuric chloride solution. To get rid of any remaining mercuric chloride, the parts were thoroughly washed twice with sterilized distilled water after sterilization. Five of the fragments were then put on petri plates with PDA that had been sterilized. For seven days, the plates were incubated at $25^{\circ}\text{C} \pm 2^{\circ}\text{C}$ in order to aid in the target pathogens recovery. Using common identification keys, the isolated fungus was then recognized based on its morphological features (Booth *et al.*, 1971; Nelson *et al.*, 1983).

Pure culture of the causal fungus

The fungus that was isolated from five distinct places was kept alive on 90 mm petri plates and Potato Dextrose Agar (PDA) slants. $25^{\circ}\text{C} \pm 2^{\circ}\text{C}$ was shown to be the ideal temperature for development. The cultures were kept at $15\text{-}20^{\circ}\text{C}$ and underwent monthly subculturing to guarantee their ongoing viability. Furthermore, for additional research, subcultures were maintained on PDA containing sterilize petri plates (Fig. 1).

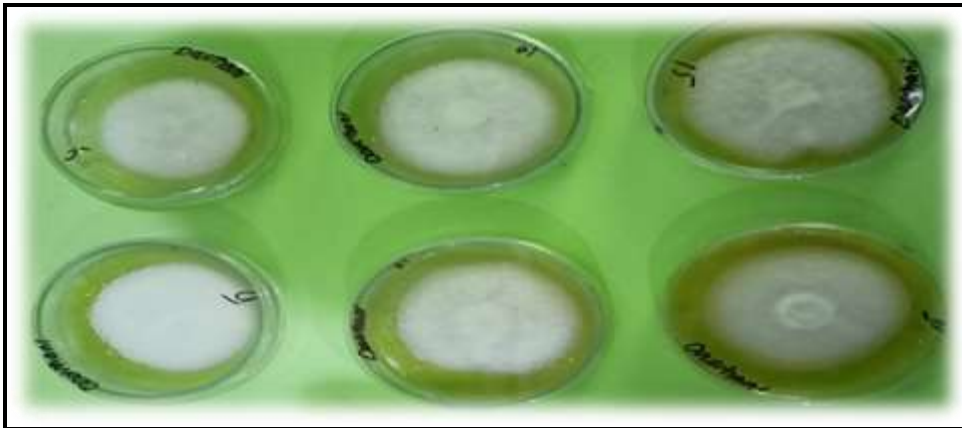


Fig. 1. Pure culture of *Fusarium Sacchari* on PDA plates.

Pathogenicity test of Fusarium wilt disease of sugarcane

A soil inoculation procedure was used to determine the pathogenicity of the isolated fungus. To achieve sufficient fungal growth, a pure culture of the fungus was initially produced on Potato Dextrose Agar (PDA) plates and incubated at $27^{\circ}\text{C} \pm 2^{\circ}\text{C}$ for eight days. After that, containers were filled with sterilized soil and sugarcane setts were planted in them. In these pots, (Fig. 2) a conidial suspension was added to the soil at a concentration of $1 \times 10^6 \text{ mL}^{-1}$. After that, the pots were put in a greenhouse that was kept at $30^{\circ}\text{C} \pm 2^{\circ}\text{C}$. (Sen and Kapoor, 1975), reported that disease signs were observed and documented after the fungus was artificially inoculated.



Fig.. 2. Pathogenicity test of the isolated fungus *Fusarium sacchari* causing Fusarium wilt of sugarcane.

Evaluation of different fungicides on the mycelial colony growth *Fusarium sacchari*

Using the food poisoning approach, the effectiveness of many fungicides, such as Ready Super

(Pyraclostrobin), Dividend Star (Difenoconazole), Dynasty CST (Azoxystrobin), Parlor (Clothianidin), and Orandis opti (Oxathiapiprolin) was assessed *in vitro*. The detail of fungicides tested in the experiment is mentioned in (Table. 1).

Three replications of the fungicide test were conducted at 5, 10 and 15 ppm concentrations. A sterile cork borer (5mm in diameter) was used to separate the test pathogen from a culture plate that was 8–10 days old, and it was then positioned in the middle of a PDA plate that had the fungicide, treatment applied to it in order to evaluate the effectiveness. After that, the plates were incubated at $27^{\circ}\text{C} \pm 2^{\circ}\text{C}$. For comparison, control plates devoid of the test fungus were kept. On the back of the petri plates, two lines that were mutually perpendicular and intersected in the middle were drawn to represent the fungus's colony expansion. After every 24 hours, the growth and inhibition % were measured in millimeters until the control plates were completely covered with fungal growth. The typical growth inhibition % calculation was used to determine each fungicide's effectiveness:

$$I = \frac{C - T}{C} \times 100$$

I= Growth Inhibition % of test pathogen

C= The control plate radial mycelial colony growth (mm)

T= The treatments radial colony growth (mm)

Table 1. The fungicides that tested against fusarium wilt of sugarcane under *in vitro* condition.

S. No	Trade Name	Chemical Name	Sources Name
1.	Ready Super	Pyraclostrobin	Rudolf Group
2.	Dividend Star	Defenoconazole	Syngenta
3.	Dynasty CST	Azoxystrobin	Syngenta
4.	Parlor	Clothianidin	Greenlet
5.	Orandis Opti	Oxathiapiprolin	Syngenta

Evaluation of different plant extracts on the mycelial colony growth *Fusarium sacchari*

The leaves were carried from a variety of plants, such as Giant Calotropis (*Calotrope gigantea*), Cinnamon (*Cinnamomum verum*), Thorn apple leaves (*Datura Stramonium*), Moringa (*Moringa olefera*), and Bitter Apple leaves (*Citrullus colocynthis*), were collected from trust worthy sources (Table 2, Fig.. 3). The cinnamon (*C. verum*), was bought from Tando Allahyar local market. First, tap water was used to wash the leaves, and then sterilized water was used. Cinnamon and all of the chosen plant leaves were allowed to air dry at room temperature in the shade. Using a pestle and mortar, the leaves were pounded into a fine powder when they had completely dried. A 100% plant extract solution was created by mixing one gram of each powdered extract with one milliliter of distilled water in a 1:1 (w/v) ratio and filtering the mixture through muslin fabric. To avoid contamination, the extracts were put in flasks, sealed with cotton, and heated to 100°C for ten minutes (Madavi and Singh, 2005). From the previously sterilized stock solution, concentrations of 5, 10 and 15 % were made and added to different conical flasks holding sterilized PDA medium. In an inoculation chamber, the PDA plates (90mm) were poured in an aseptic manner. Using a sterile cork borer (5mm), pathogen samples were extracted from culture plates that were 8 to 10 days old and inserted into the middle of PDA plates. The plates were subsequently incubated at $27^{\circ}\text{C} \pm 2^{\circ}\text{C}$. As controls, Petri dishes were used but without the test fungus. On the underside of the petri plates, two lines that were mutually perpendicular and intersected in the middle were drawn to represent the test fungus's radial colony development. Millimeter-based measurements of colony growth and inhibition % were made every 24 hours until the untreated plates were completely covered. The growth inhibition % of each test extracts was determined by using previously mentioned growth inhibition % formula.

Table 2. The application of different plant extracts against Fusarium wilt of sugarcane under *in vitro* conditions.

S.NO	English Name	Local Name	Scientific Name
1.	Giant calotrope	Akk	<i>Calotrope gigantea</i>
2.	Cinnamon	Darchini	<i>Cinnamomum verum</i>
3.	Thorn apple leaves	Datura	<i>Datura stramonium</i>
4.	Moringa	Suhanjara	<i>Moringa olefera</i>
5.	Bitter apple leaves	Tooh	<i>Citrullus colocynthis</i>

Data analysis

The experimental data were gathered and subjected to statistical analysis using Statistix 8.1 computer software

(Analytical Software, 2005). Analysis of variance (ANOVA) was conducted, and mean values were distinguished using the least significant difference (LSD) test. A significance level 0.5% was applied to all experimental data to adjust the probability value.



Fig. 3. Different plant extracts used for *in vitro* condition of Fusarium wilt causing fungus in sugarcane.

RESULTS

Prevalence of Fusarium wilts disease of sugarcane at different locations of District Tando Allahyar

During survey different locations were surveyed in the surrounding of district Tando Allahyar. The disease was recorded in all locations with the range of 18 to 36% disease incidence. The highest disease incidence % was recorded at Missan Wadi 36% followed by Haji Muhammad Ibrahim Dars 28%, Village Dad Khan Jarwar 22% and Village Masoo Bozdar 20% respectively. While minimum disease incidence 18% was recorded at Qazi Noor Muhammad Laghari (Fig. 4).

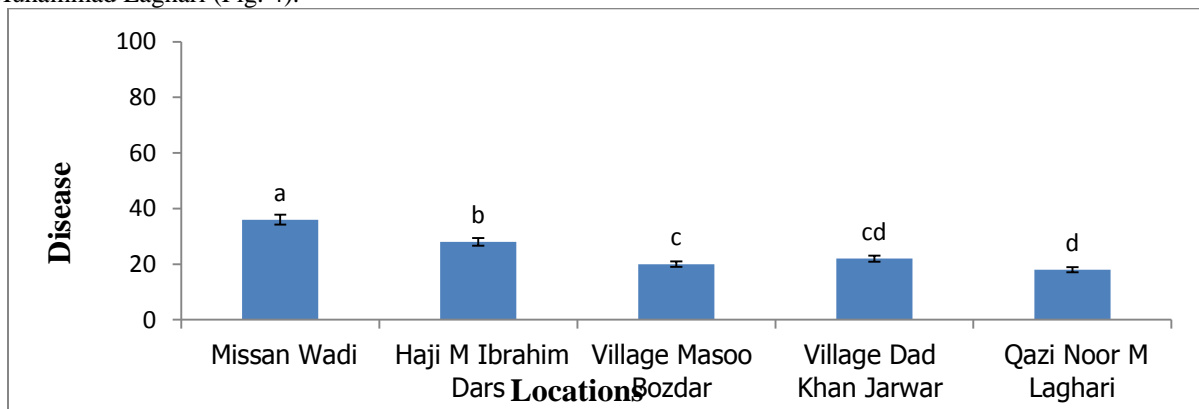


Fig. 4. Disease incidence % of Fusarium wilt of sugarcane at different locations of district Tando Allahyar.

Isolation and identification of sugarcane wilt causing pathogen

The pathogen that caused the infection was isolated using the tissue isolation technique. Fourteen days after inoculation, the profuse, aerial, pale white mycelium in the isolated colonies became purple. Numerous microconidia in false heads on ovoid, single-celled monophialides or polyphialides were seen, along with elongated macroconidia with a curved apical cell and a poorly developed basal cell with three septa. The isolated pathogen's morphological traits, conidia, and conidiophores matched those of *Fusarium sacchari*, the sugarcane wilt disease's causal agent (Fig. 5A,B,C).

Pathogenicity test

Using the soil inoculation approach, the pathogen suspension of *Fusarium sacchari* was introduced into pots with one month old sugarcane plants. The procedure was carried out in a greenhouse, as explained in the materials and methods section of this chapter. Every therapy has a control group. The findings showed that when the infection progressed, the leaves of infected plants first went yellow before becoming brown. The collar root area showed

signs of white mycelium development. In addition, the stem's base became dark and sank. The plant gradually dried and toppled within 15 days of inoculation (Fig. 6). The un-inoculated plants did not produce the symptoms. The re-isolation was also carried out from the inoculated sugarcane. The *Fusarium sacchari* was isolated and identified on the basis of their morphological characteristics.

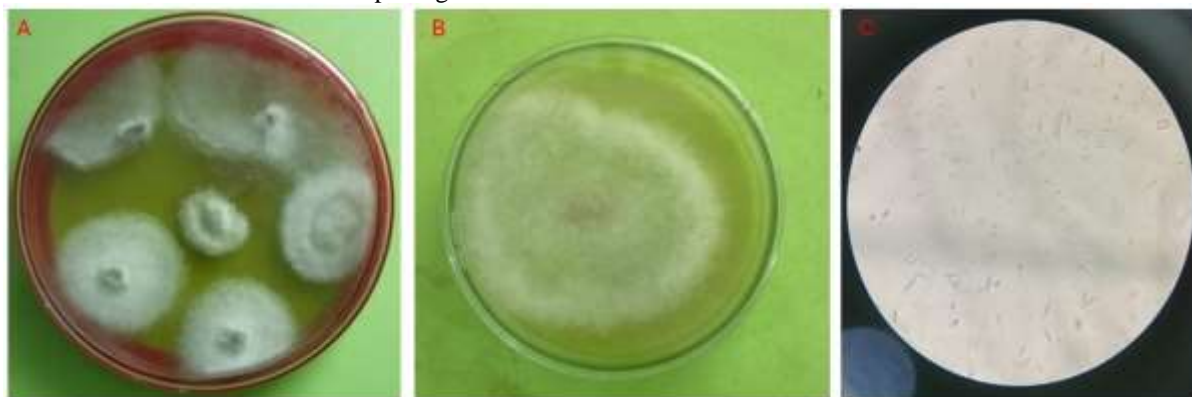


Fig. 5A, B, C. Morphological characteristics of *Fusarium sacchari* causing Fusarium wilt of sugarcane.



6. Pathogenicity test of *Fusarium sacchari* causing Fusarium wilt of sugarcane.

Effect of different fungicides on the mycelial colony growth of the *Fusarium sacchari*

The Fig. 7 shows result that all tested fungicides inhibited the linear colony growth of *Fusarium sacchari* under *in vitro* conditions. As the dose of the fungicides increases the growth of the test fungus decreases. Among all tested fungicides the Ready super (Pyraclostrobin) was found highly effective fungicides as compared to other fungicides. The lowest mycelial colony growth of the test fungus was recorded by Ready super (Pyraclostrobin) fungicide (8.11, 11.00 and 13.66 mm) at 15, 10 and 5 ppm doses followed by Parlor (Clothianidin) fungicide (22.33 mm), Dividend star (Difenoconazole) fungicide (28.33 mm) at 15 ppm doses. The highest mycelial colony growth of the test fungus was observed by Orandis opti (Oxathiapiprolin) fungicide (68.66 mm) at 5 ppm. Among the all-tested fungicides Dynasty CST (Azoxystrobin) and Orandis opti (Oxathiapiprolin) were found less effective as compared to other fungicides.

Effect of different fungicides on the growth inhibition % of the *Fusarium sacchari*

All fungicides inhibited the growth of test fungus at all used doses. The maximum inhibition % was recorded by Ready super (Pyraclostrobin), (90.74 and 87.77) at 15 and 10 ppm followed by Parlor (Clothianidin), (75.18) and Dividend star (Difenoconazole), (68.51) respectively. The lowest inhibition % was noted by Orandis opti (Oxathiapiprolin), (23.70) at 5 ppm. The highest dose 15 ppm of all tested fungicides inhibited the test fungus as compared to lowest dose 5 ppm. The Ready super (Pyraclostrobin) fungicide was recorded highly effective whereas, Parlor (Clothianidin) and Dividend star (Difenoconazole) were found moderately effective. The Orandis opti (Oxathiapiprolin) and Dynasty CST (Azoxystrobin) were found less effective as compare to other tested fungicides (Fig. 8).

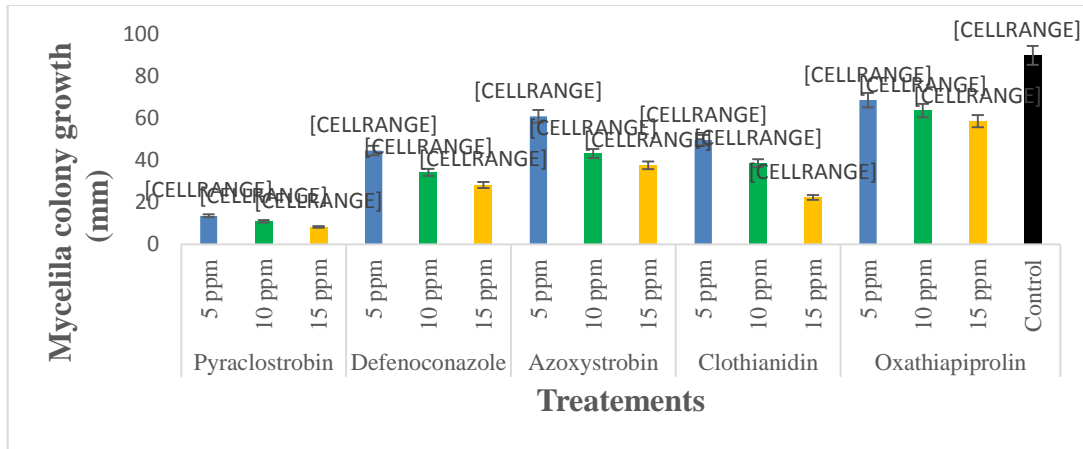


Fig. 7. Effect of different fungicides on the mycelial colony growth of *Fusarium sacchari*.

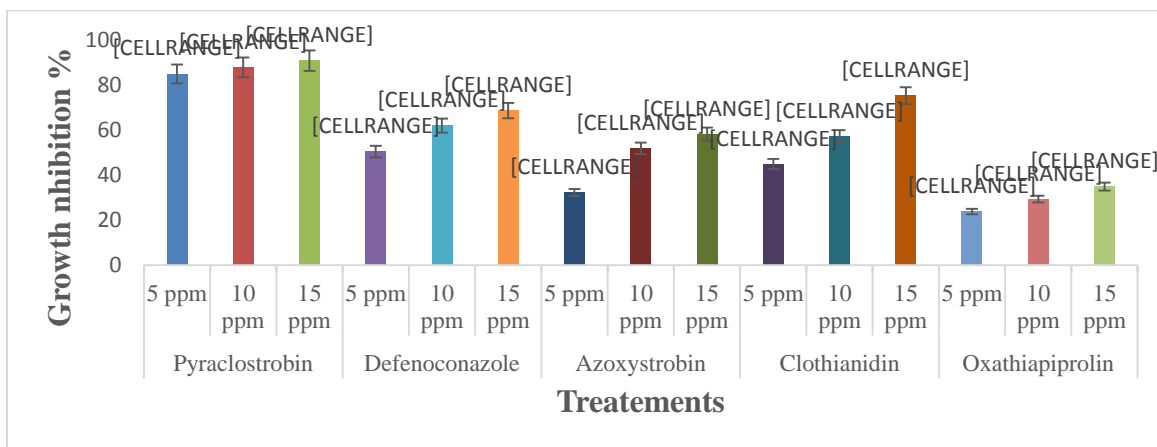


Fig. 8. Effect of different fungicides on the growth inhibition % *Fusarium sacchari*.

Effect of different Plant extracts on the mycelial colony growth of *Fusarium sacchari*

All tested plant extracts were found effective at higher dose against the test fungus. As the concentration of the plant extracts rises the efficacy of plant extracts also increases. The plant extracts of the Bitter apple leaves (*Citrullus colocynthis*) and Giant calotrope (*Calotrope gigantea*) were found highly effective at 15% concentration as compared to other plant extracts. The maximum mycelial colony growth was recorded by Cinnamon (*Cinnamomum verum*), (81.66 mm) followed by Moringa (*Moringa oleifera*), (71.66 mm) and Thron apple leaves (*Datura stramonium*), (76.33 mm) at 5% concentrations whereas, the 10% concentrations of all tested plant extracts were found moderately effective. The lowest colony growth of the test fungus was recorded by Bitter apple leaves (*Citrullus colocynthis*), (42.66 mm) and Giant calotrope (*Calotrope gigantea*), (44.66 mm) at 15% concentrations as compared to other plant extracts against the test fungus (Fig. 9).

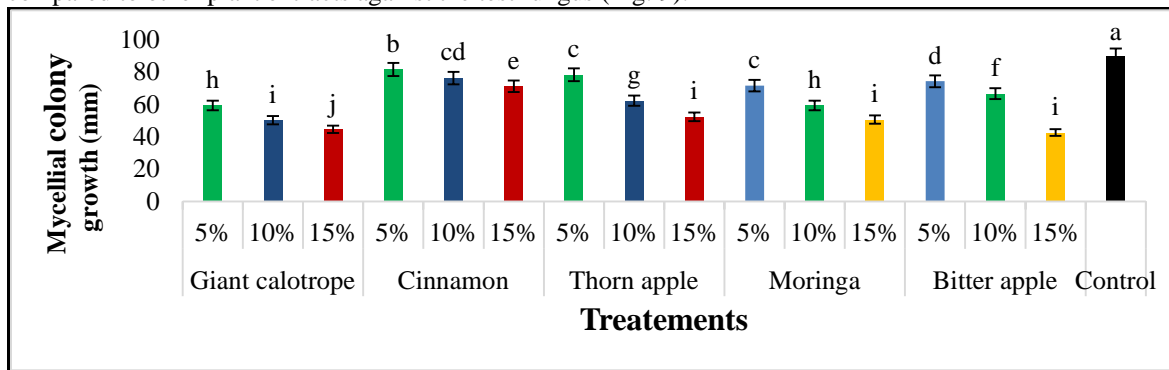


Fig. 9. Effect of different Plant extracts on the mycelial colony growth of *Fusarium sacchari*.

Effect of different Plant extracts on the growth inhibition % of the *Fusarium sacchari*

The results in figure 4.5 shows that the all-tested plant extracts inhibited the mycelial colony growth of the test fungus at all tested concentrations however the highest concentrations were found highly effective as compared to lower concentrations. The maximum inhibition % of the test fungus was recorded by Bitter apple leaves (*Citrullus colocynthis*), (52.59) followed by Giant calotrope (*Calotrope gigantea*), (50.33), Moringa (*Moringa oleifera*), (43.70) and Thron apple leaves (*Datura stramonium*), (41.85) at 15% concentration. The moderate inhibition % was recorded by Moringa (*Moringa oleifera*) (34.07), Thron apple leaves (*Datura stramonium*), (30.74) and Bitter apple leaves (*Citrullus colocynthis*), (25.92) at 10% concentrations. The lowest inhibition % of the test fungus was recorded by Cinnamon (*Cinnamomum verum*), (9.25) and (12.92) by Thron apple leaves (*Datura stramonium*) respectively (Fig. 10).

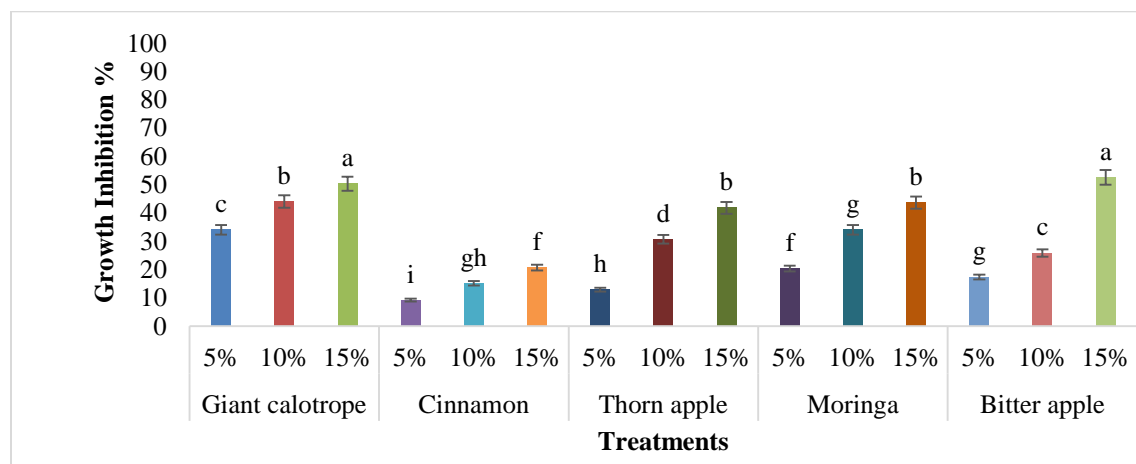


Fig. 10. Effect of different Plant extracts on the growth inhibition % of the *Fusarium sacchari*.

DISCUSSION

Fusarium wilt of sugarcane caused by *Fusarium sacchari* is an important stalk disease affecting sugarcane productivity in Asian countries. Among the major diseases reported in the country, wilt is a serious disease affecting cane production and productivity different varieties. The disease is responsible for the elimination of many commercial varieties from cultivation in disease endemic regions in the country (Viswanathan *et al.*, 2013). The amount of dry or dead canes discovered in the field during harvest which can vary from 2 to 10 tons per hectare is normally used to compute losses resulting from wilt (Parthasarathy *et al.*, 1972). According to Sarma *et al.* (1976) disease incidence is greater in ratoon crops than in plant crops, and yield losses might be as high as 65%. Furthermore, fewer tillers were generated by sick plants than by healthy ones (Agnihotri and Singh, 1989).

During survey the most hit regions due to sugarcane *Fusarium* wilt were Missan Wadi 36% and Village Haji Muhammad Ibrahim Dars 28% in districts Tando Allahyar, the lowest disease incidence 18% was found at the village of Qazi Noor Ahmed. The moderate 22% disease incidence was recorded at Village Dad Khan Jarwar. This greater prevalence might be caused by the possible influence of sugarcane variety, as well as by unsuitable management approaches and the pathogen's ideal climatic circumstances (Agnihotri and Rao, 2002; Ishaq *et al.* 2023., Abro *et al.* 2023). A nationwide survey to ascertain the prevalence of wilt of sugarcane found that: 5–10% in Uttar Pradesh; the recent survey showed severe wilt incidence of up to 60% in East Godavari District, where the crop cultivation is in low land. However, 10% disease of sugar cane wilt disease incidence in areas of West Godavari district under upland conditions., it was also found that the disease present from trace levels to 5-6% in most of the upland regions of Tamil Nadu and Maharashtra where as in parts of Andhra Pradesh, Gujarat and Orissa disease incidence of up to 10-20% was recorded in deltaic regions (Viswanathan, 2013). Following the procedure of separation and identification of the pathogen causing sugar cane wilt disease, the separated colonies showed copious amounts of aerial, pale white mycelium that became purple 14 days after inoculation. There were many microconidia in false heads on ovoid, single celled monophialides or polyphialides, as well as elongated macroconidia with a bent apical cell and an underdeveloped basal cell with three septa. The isolated pathogen's morphological traits, conidia, and conidiophores aligned with *Fusarium sacchari*, the causal agent of sugarcane wilt (Leslie and Summerell, 2006; Ost *et al.*, 2022).

The findings of the pathogenicity test showed that when the infection progressed, the leaves of infected plants

first went yellow before becoming brown. The collar root area showed signs of white mycelium development. In addition, the stem's base became dark and sank. The plant gradually dried and toppled within 15 days of inoculation. The test pathogen was re-isolated aseptically on PDA plates from artificially wilt affected sugarcane plant, studied and compared its cultural and morphological characteristics with the original culture of *Fusarium* isolates obtained from naturally fusarium wilt diseased sugarcane plants. The similar types of results were recorded by (Bansal *et al.*, 2016). Sen and Kapoor, (1975) also reported that disease signs were observed and documented after the fungus was artificially inoculated.

For managing the *Fusarium sacchari* causing the fusarium wilt disease of sugarcane five different fungicides and botanical extracts were tested by food poisoning method. The result shows that fungicides are highly effective as compared to plant extracts. The Pyraclostrobin fungicide was recorded highly effective whereas, Clothianidin and Difenconazole were found moderately effective. The Oxathiapiprolin and Azoxystrobin were found less effective. The highest and lowest % of *Fusarium sacchari* inhibition were recorded at 15 and 10 ppm, respectively, among the fungicides that were evaluated. Our chemical controls result is in good agreement with those of Osti *et al.* (2022), who used the poison plate approach to assess the fungicides azoxystrobin, difenoconazole, hymexazole, cyprodinil, and thiabendazole against *Fusarium sacchari* at doses of 1, 10, 100, and 1000 $\mu\text{g mL}^{-1}$. The study found that the best % of mycelial colony growth suppression were reached by thiabendazole at 10, 100, and 1000 $\mu\text{g mL}^{-1}$ and difenoconazole at 1000 $\mu\text{g mL}^{-1}$. On the other hand, cyprodinil, thiabendazole, and difenoconazole showed the lowest % of inhibition. These fungicides have been shown in several *in vitro* control tests on *Fusarium* species to have not able growth suppression effects. (Albuquerque and Gusqui, 2018; Poussio *et al.*, 2023), found that treatment with azoxystrobin, carbendazim, copper phosphite, and thiabendazole completely inhibited *F. oxysporum* isolated from tomato (*Solanum lycopersicum*). Similarly, at 150 and 200 ppm, carbendazim completely inhibited *F. oxysporum*, according to Dahal and Shrestha's (2018) findings.

The results of tested plant extracts against *Fusarium sacchari* showed the inhibition of the mycelial colony growth of the test fungus at all tested concentrations. However, the highest concentrations were found highly effective as compared to lowest concentrations. The maximum inhibition % of the test fungus was recorded by Bitter apple leaves (*Citrullus colocynthis*), (52.59) followed by Giant calotrope (*Calotrope gigantea*), (50.33), Moringa (*Moringa oleifera*), (43.70) and Thorn apple leaves (*Datura stramonium*), (41.85) at 15% concentration. The moderate inhibition % was recorded by Moringa (*Moringa oleifera*), (34.07), Thorn apple leaves (*Datura stramonium*), (30.74) and Bitter apple leaves (*Citrullus colocynthis*), (25.92) at 10% concentrations. The lowest inhibition % of the test fungus was recorded by Cinnamon (*Cinnamomum verum*), (9.25) and (12.92) by Thorn apple leaves (*Datura stramonium*) respectively. Sagar *et al.* (2007) who assessed the effectiveness of fourteen plant extracts at 5 and 10% concentrations against *Pythium aphanidermatum* and *F. solani* caused ginger rhizome rot *in vitro*, corroborate our findings. The mycelial colony growth inhibition of the extracts tested against *F. solani* was highest for the powder extract of *Ferula feotida* (68.51%), followed by the leaf extract of *Ocimum* (60.16%). Furthermore, under *in vitro* conditions, aqueous leaf extracts (2.0%) from 15 plants showed the greatest inhibition against *Alternaria lini*, the causative agent of leaf and bud blight in linseed, with *Azadirachta indica* showing the highest % (67.7%), followed by *Lawsonia inermis* (63.0%), *Datura metel* (39.2%), *Calotropis procera* (37.8%), *Lantana camara* (36.6%), and *Citrus medica* (Singh and Singh, 2007). In an experiment carried out *in vitro*, Vijaya *et al.*, (2007) evaluated the fungicidal toxicity of twelve plant extracts at 5 to 10% concentrations against sett rot disease in sugarcane. They discovered that extracts with a 10% concentration worked noticeably better than ones with a 5% concentration. Garlic extract, at 10%, showed the greatest mycelial growth inhibition of all the extracts at 53.13%, indicating its greater effectiveness over the other plant extracts.

Conclusions

The highest disease incidence 36% was found at Missan Wadi followed by Haji Muhammad Ibrahim Dars, village Dad Khan Jarwar and village Masoo Bozdar respectively. The minimum disease incidence 18% was found at Qazi Noor Muhammad Laghari. The *Fusarium sacchari* was isolated from all collected diseased samples and identified on the basis of morphological characteristics. The disease is associated by *Fusarium sacchari* which was confirmed through soil inoculation method. Five different chemical fungicides were tested, among them Ready super (Pyraclostrobin), Parlor (Clothianidin) and Dividend star (Difenconazole) were found highly effective at 15 ppm. Whereas, the Dynasty CST (Azoxystrobin) and Orandis opti (Oxathiapiprolin) fungicides were found less effective. Among the five tested plant extracts the Bitter apple leaves (*Citrullus colocynthis*) and Giant calotrope (*Calotrope gigantea*) were found highly effective 15% concentration whereas, the Moringa (*Moringa oleifera*) and Thorn apple leaves (*Datura stramonium*) were found moderately effective at 10% concentration, while Cinnamon (*Cinnamomum verum*) was found lowest effective at 5% concentration. The overall results shows that the chemicals are highly effective as compare to plant extracts.

CONFLICT OF INTEREST

The authors declare no conflict of interest

AUTHORS' CONTRIBUTION

GHJ, MAA and UA designed the experiment. GHJ, MU, JUH and RA conducted the experiment. GHJ, MAA, JUH and GBP provided critical guidance and oversight throughout the research process. GHJ, JUH and GBP carried out data analysis. MAA and GHJ edited the manuscript and improve English language. All authors have read and approved the final manuscript.

REFERENCES

- Abdul, M., A. Javaid, A. Shoaib and R. Bajwa (2015). Antifungal potential of methanolic extracts of *Syzygium cumini* and *Azadirachta indica* against *Fusarium solani*. *International Journal of Agriculture and Biology*, 17(5): 1035–1040.
- Abro, M. A., G. H. Jatoi, G. B. Poussio, N. Koondhar, I. M. Figari and W. R. Arshad (2022). The Efficient Use of Different Fungal Bioagents for Eco-Friendly Management of Fusarium Wilt Disease of Tomato in Sindh, Pakistan. *Pakistan Journal of Phytopathology*, 34(2): 103-115.
- Agnihotri, V. P. and G. P. Rao (2002). A century status of sugarcane wilt in India. In S. B. Singh, G. P. Rao, and S. Eswaramoorthy (Eds.), *Sugarcane Crop Management* (pp. 145–160). Studium Press.
- Agnihotri, V. P. and K. Singh (1989). *Fusarium* wilt of sugarcane: Impact on tillering and plant growth. *Sugarcane Wilt: New Insights Into Pathogen Identity, Variability and Pathogenicity*, FPSB, 6(SI2): 30–39.
- Akhtar, K. P. and A. Javai (2018). Antifungal activity of medicinal plant extracts against red rot pathogen of sugarcane. *Pakistan Journal of Phytopathology*, 30(2): 175–181.
- Booth, C. (1971). *Fusarium: Laboratory Guide to the Identification of the Major Species*. Commonwealth Mycological Institute, Kew, Surrey, England.
- Corcoran, E., C. De Lucia and M. Giampietro (2020). Biofuel production: Considerations for sustainability. *Renewable and Sustainable Energy Reviews*, 131: 110022.
- FAOSTAT. (2023). *Food and Agriculture Organization of the United Nations*. <https://www.fao.org/faostat/>
- Farber, A., Y. Elad and M. Harel (2019). Integrated disease management in sugarcane. *Plant Disease Management Reports*, 13(2): 104–112.
- Farooq, S., A. Khan and M. Nadeem (2019). Sugarcane production and its role in Pakistan's economy. *Pakistan Journal of Agricultural Sciences*, 56(1): 145–152.
- Gupta, R., V. Sharma and P. Pathak (2011). *Fusarium* wilt of sugarcane and its integrated management. *Sugar Tech*, 13(4): 305–311.
- Ishaq, H., M. A. Khan, I. Ahmed, S. M. A. Shah, U. Ali, G. H. Jatoi and S. Iftikhar (2023). Antifungal Exploitation of Fungicides and Plant Extracts Against *Fusarium Oxysporum* F. Sp. *Melongenae* Causing *Fusarium* Wilt of Eggplant. *Pakistan Journal of Biotechnology*, 20(01): 59-67.
- Jaiman, R. K. and A. Jain (2010). Fungicidal management of *Fusarium* wilt in sugarcane. *Journal of Mycology and Plant Pathology*, 40(2): 295–297.
- Khalikar, P. V., A. J. Deshmukh and P. P. Patil (2011). Management of sugarcane wilt using fungicides and bioagents. *Sugar Tech*, 13(1): 34–38.
- Leslie, J. F. and B. A. Summerell (2006). *The Fusarium Laboratory Manual*. Blackwell Publishing.
- Bansal, Y., J. Chander, N. Kaistha, N. Singla, S. Sood and A. D. van Diepeningen (2016). *Fusarium sacchari*, a cause of mycotic keratitis among sugarcane farmers—a series of four cases from North India. *Mycoses*, 59(11): 705-709.
- Manzoor, M., A. Rehman and Q. Abbas (2023). Socioeconomic role and prospects of sugarcane in agricultural economies. *Agricultural Economics Review*, 16(3): 251–264.
- Moosa, A., M. Bashir and M. Shafique (2017). Fungicidal control of wilt disease in sugarcane. *Pakistan Journal of Phytopathology*, 29(2): 201–208.
- Nelson, P. E., T. A. Toussoun and W. F. O. Marasas (1983). *Fusarium: Diseases, Biology, and Taxonomy*. Pennsylvania State University Press.
- Obongoya, B. O., S. O. Wagai and G. D. Odhiambo (2010). Bioactivity of plant extracts against *Fusarium oxysporum* causing wilt in beans. *African Crop Science Journal*, 18(4): 213–222.
- Medina-Osti, F., A. Gutiérrez-Díez, S. Ochoa-Ascencio and S. R. Sinagawa-García (2022). *In vitro* sensitivity of *Fusarium sacchari* isolated from sugar cane to five fungicides. *Revista mexicana de fitopatología*, 40(3): 447-457.

- Panigrahi, S., S. Behera and B. Mishra (2021). Nutritional and medicinal properties of sugarcane juice. *International Journal of Current Microbiology and Applied Sciences*, 10(3): 178–187.
- Parthasarathy, S. V. (1972). *Sugarcane in India*. K.C.P. Ltd.
- Parthasarathy, S. V. (1972). Studies on sugarcane wilt. *Indian Sugar*, 22(4): 23–27.
- Paul, A. K., S. Sultana and M. M. Hossain (2022). Incidence of sugarcane wilt disease in Bangladesh. *Bangladesh Journal of Plant Pathology*, 38(1): 45–50.
- Poussio, G. B., G. H. Jatoi, M. I. Khaskheli, S. T. Qaz, S. A. Siddiqui and M. Mengwar (2023). Investigating the Efficacy of Diverse Biopesticides for Managing Fusarium Wilt Disease of Tomato. *Journal of Agriculture and Veterinary Science*, 2(3): 167-178.
- Poongothai, M., R. Viswanathan and P. Malathi (2014a). Identification of *Fusarium sacchari* as the causal agent of sugarcane wilt. *Australasian Plant Disease Notes*, 9(1): 121–125.
- Poongothai, M., R. Viswanathan, P. Malathi and A. Ramesh Sundar (2014b). *Fusarium sacchari* causing sugarcane wilt: Variation in morphological characteristics. *International Sugar Journal*, 116: 54–63.
- Qureshi, M. H., G. Abbas and A. M. Qazi (2020). Sugar industry and sugarcane productivity in Pakistan. *Pakistan Journal of Agricultural Research*, 33(4): 635–641.
- Rafiq, M., A. A. Khan and G. Shabbir (2021). Potential of plant extracts in the control of soil-borne pathogens. *International Journal of Agricultural and Biological Research*, 7(1): 44–50.
- Rajput, L. B., G. A. Shah and G. H. Jatoi (2021). Status of major sugarcane diseases in Pakistan and their management. *Journal of Plant Pathology Research*, 5(2): 112–119.
- Raza, M. A., I. Haider and M. A. Khan (2023). Sugarcane juice derivatives and their commercial significance. *Food Science and Nutrition*, 11(2): 934–941.
- Sana, M., A. Younis and A. Raza (2017). Antifungal activity of various plant extracts against *Fusarium oxysporum*. *Pakistan Journal of Botany*, 49(2): 873–878.
- Sarma, M. N. (1976). Wilt disease: Incidence higher in ratoon than plant crop, up to 65% yield loss. *Variability in Sugarcane Wilt*, North Coastal Zone Report.
- Sen, B. and I. J. Kapoor (1975). Systemic fungicides for the control of wilt of peas. *Journal of Vegetable Science*, 2(1): 76–78.
- Sharma, R., R. Singh and A. Dwivedi (2010). Cultural control of sugarcane wilt pathogen. *Indian Phytopathology*, 63(3): 342–347.
- Shoaib, A., M. E. Akhtar and I. U. Haq (2018). Impact of sustainable sugarcane production on rural livelihoods in Pakistan. *Journal of Sustainable Agriculture*, 12(1): 85–92.
- Viswanathan, R. (2013). Sugarcane diseases and their management. *Sugar Tech*, 15(3): 239–247.
- Viswanathan, R., P. Malathi, M. Poongothai and A. Ramesh Sundar (2013). Status of sugarcane wilt pathogen identity and prevalence. *Sugar Tech*, FPSB, Indian Sugarcane Research.
- Wani, A. B., M. Mushtaq and K. R. Hakeem (2023). Biomass valorization of sugarcane bagasse for sustainable bioenergy. *Renewable Energy Reviews*, 157: 112018.

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