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## RESEARCH ARTICLE

## Comparative Studies on the Efficiency of Different Chemicals to Control Citrus Scab (*Elsinoe Fawcettii*) in *Citrus Reticulata* Blanco

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### ABSTRACT

*Elsinoë fawcettii*, a fungus, is the cause of citrus scab. The disease is economically significant and common, and potential threat to citrus groves worldwide, particularly in areas present in warm and humid regions. The study was planned on the objective to evaluating the fungicides efficacy for the identification of most efficient fungicide to citrus scab. The study was conducted in three consecutive years (2017-18, 2018-19, and 2019-20) on 20 years old Kinnow plants, which were planted at a spacing of 6.1 m x 6.1 m. The foliar spray was applied before flowering in March, after fruit setting in April and in the months of August. Various chemicals including Azoxystrobin, Pyraclostrobin+Metiram, Difenoconazole+Azoxystrobin, Copper Oxychloride +Cymoxanil, Copper hydro oxide, Tebuconazole+Trifloxystrobin, Fluoxstrobin, Bordeaux mixture 1%, Azoxystrobin + Chlorothalnil, and Chlorothalonil+Thiophenate methyl were evaluated and results were compared with control. Diseased data for citrus scab disease was recorded after fifteen days of the last spray. The results revealed the superior efficacy of Difenoconazole+Azoxystrobin (1 ml/L of Water) and Bordeaux mixture (1%) in controlling citrus scab in Kinnow. These systemic fungicides offer significantly better disease suppression and yield improvement than other chemicals. A systemic chemical Fluoxstrobin (1 ml/L of Water) and control (non-treated with chemicals) were inferior to other treatments in controlling citrus scab incidence. Integrating these fungicides with timely applications, for Kinnow disease management programs, can substantially improve fruit quality and farmer profitability.

**Keywords:** disease incidence, foliar spray, kinnow, fungicides, disease management.

### INTRODUCTION

Kinnow mandarin (*Citrus reticulata* Blanco) is a commercially significant citrus cultivar extensively cultivated in Pakistan (Hayat *et al.*, 2022). Citrus is the world most widely grown fruit crop and having economic significance in the world. The fruit of citrus does have high content of minerals, dietary fiber and vitamins. Citrus fruits have positive effects on human health. Oranges account for more than 50% of total

production of citrus. Several factors both including abiotic and biotic are affecting citrus and affecting its fruit quality directly, causing prominent deformities and blemishes of its fruits (Van Zyl, 2019).

In Pakistan citrus holds a prominent position as a major cash crop, contributing significantly to the livelihoods of farmers and the national horticultural economy. In Pakistan total citrus was grown on area of 152,716

hectares with production of 2,321,802 MMT in the year 2020-21, including major share of Punjab province 2,246,473 MMT on 141,178 hectares (GOP, 2023).

However, its productivity is severely constrained by several major biotic stressors, including Asian citrus psyllid (*Diaphorina citri* Kuwayama), citrus whitefly (*Dialoeurodes citri* Ashmead), sooty mold (*Capnodium citri*), and dieback caused by *Colletotrichum gloeosporioides*, collectively leading to yield reductions of up to 70%. The findings validate the IPM approach as an ecologically and economically sustainable strategy for managing key pest complexes in Kinnow mandarin cultivation (Meena *et al.*, 2025). Citrus scab is one of the most dangerous diseases of citrus (Malik *et al.*, 2021).

To determine effective management strategies for citrus dieback and scab diseases, the individual and combined effects of mustard oil cake, urea, triple super phosphate (TSP), muriate of potash (MOP), cow dung, the fungicide Bavistin, irrigation, and pruning were evaluated (Alam *et al.*, 2020).

Citrus scab is caused by the fungus *Elsinoë fawcettii* (Dewdney, 2019). In one of previous study the citrus scab pathogen was effectively suppressed by bio-sulfur at 500-, 1000-, and 2000-fold dilutions. In field trials, the incidence of citrus scab on spring-flush leaves was 40.3% in untreated plots, compared to 5.3%, 10.3%, 12.3%, 15.3%, and 24.0% in plots treated with imibenconazole, 2-4 and 6-6 lime-Bordeaux mixtures (commonly adopted in eco-friendly cultivation), 500-fold diluted bio-sulfur, lime sulfur, and 1000-fold diluted bio-sulfur, respectively. For citrus fruit, disease incidence was 79.3% in the untreated control, and 4.0%, 33.8%, 42.0%, 43.3%, 44.8%, and 78.0% in treatments. Given that citrus scab infections can initiate as early as May with the emergence of the spring flush, prophylactic treatments should be applied by mid- to late April to optimize disease management, enhance control efficacy, and reduce labor and production costs (Oh *et al.*, 2020).

These are phytopathogenic fungi of significant concern in citrus cultivation due to their association with citrus scab diseases. Both species synthesize a distinctive group of red to orange pigments known as elsinochromes, a class of non-host-selective, photosensitizing phytotoxins essential for full virulence and necrotic lesion development on host tissues. *E. fawcettii* is the causal agent of citrus scab (previously categorized into sour orange scab and common scab) and infects a broad range of *Rutaceae* species and

hybrids globally. The biosynthesis of elsinochrome in *E. fawcettii* is tightly coordinated with conidiation, and emerging evidence suggests that shared environmental and physiological cues regulate both secondary metabolism and asexual reproduction. Further elucidation of these regulatory networks may provide novel targets for disease management. This review underscores the significance of *Elsinoë* spp. as critical pathogens in citrus pathosystems, pathogenicity, and the pivotal role of elsinochrome in disease development. (Chung, 2011). "Essential oils (EOs) are natural plant-derived products characterized by a complex composition of bioactive volatile compounds, many of which exhibit well-documented antibacterial properties. The combined application of citrus-derived essential oils demonstrated efficacy in suppressing the phytopathogenic bacterium "*Streptomyces scabiei*" (Prieto *et al.*, 2024).

Another safe fungicide, Fluoxastrobin, has been applied for control of fungicidal diseases of kinnow (Adaskaveg *et al.*, 2022). Pyraclostrobin and metiram are fungicidal agents employed for the management of fungal pathogens in crops. Pyraclostrobin is a systemic strobilurin-class fungicide that inhibits mitochondrial respiration by blocking electron transfer at the cytochrome  $bc_1$  complex (Complex III), thereby disrupting ATP synthesis and arresting fungal development. In contrast, metiram is a non-systemic dithiocarbamate fungicide that functions as a multi-site contact protectant. It acts by inhibiting essential enzymatic processes during spore germination and early fungal growth stages, thereby preventing infection establishment on plant surfaces (Horsfield *et al.*, 2010; Abd El-Rahman *et al.*, 2018). Tebuconazole and Trifloxystrobin were also successfully cured scab in stone fruits by reducing production of conidia (Lalancette *et al.*, 2015).

Among the fungal diseases, citrus scab has emerged as a prevalent and economically significant issue. The disease is characterized by the formation of superficial, corky lesions on the fruit surface, which greatly diminish the marketability of affected fruits, especially in the fresh produce sector. Citrus scab is prevalent in many humid, citrus-growing regions globally. The etiology of citrus scab is complex, involving multiple fungal species whose identification is complicated by the limitations of traditional morphological and cultural diagnostic methods. Therefore, accurate species-level identification

requires the use of molecular diagnostics and host specificity assays. Among the molecular tools available, enzyme-linked immunosorbent assay (ELISA) and polymerase chain reaction (PCR) are commonly employed, with PCR offering superior specificity and sensitivity for pathogen detection (Munir *et al.*, 2020). Disease management with chemical and biochemical fungicides applications, screening, natural controlling agents, disease free citrus nursery development is necessary for food security and safety (Jyothi and Shilpa, 2020). Keeping in view of the above-mentioned facts it is necessary to find out the most suitable fungicide against the citrus scab disease in Kinnow under subtropical and semi-arid regions of Punjab.

### MATERIALS AND METHODS

The experiment was conducted at Citrus Research Institute, Sargodha- 40100. The study was conducted in three consecutive years (2017-18, 2018-19, and 2019-20) on 20 years old kinnow plants having planting distance of 6.1 m x 6.1 m. Various treatments (chemical ingredients along with its concentrations) i.e., Azoxystrobin @ 1 ml/L of Water, Pyraclostrobin + Metiram @1.5 g/L of Water, Difenconazole + Azoxystrobin @1 ml/L of Water, Copper Oxychloride

+Cymoxanil@2 ml/L of Water, Copper Hydro Oxide 2.5 g/L of Water, Tebuconazole+Trifloxystrobin @0.5 g/L of Water, Fluoxstrobilin @1 ml/L of Water, Azoxystrobin + Chlorothalnil @1 ml/L, Chlorothalonil+Thiophenate methyl @1 ml/L, Bordeaux mixture @1% (1:1:100) were applied on kinnow plants and results were compared with control (non-treated plants) (Table 1). The foliar spray was applied before flowering in March, after fruit setting in April and in the months of August. Diseased data for citrus scab disease was collected /recorded after fifteen days of the last spray.

**Bordeaux Mixture Preparation:** Bordeaux mixture, a conventional fungicidal formulation, was synthesized by a few modifications in procedure developed by Nassar-Hardy (2011) by the admixture of aqueous solutions of copper (II) sulfate (CuSO<sub>4</sub>) and calcium hydroxide [Ca(OH)<sub>2</sub>], commonly referred to as hydrated lime. The regular preparation ratio of Bordeaux mixture is 10-10-100, which denotes copper sulfate (pounds), hydrated lime (pounds), and 100 gallons water, respectively. For proper mixing of copper sulfate solution in the lime solution to avoid precipitation, copper sulfate is gradually mixed in the lime solution with continuous stirring.

Table 1. Selected Chemicals used to control citrus scab (*Elsinoe Fawcettii*) in Kinnow

Treatments	Chemicals applied as foliar spray	Concentration of active ingredient
T1	Azoxystrobin	1 ml/L of Water
T2	Pyraclostrobin+Metiram	1.5 g/L of Water
T3	Difenoconazole+Azoxystrobin	1 ml/L of Water
T4	Copper Oxychloride +Cymoxanil	2 ml /L of Water
T5	Copper hydro oxide	2.5 g/L of Water
T6	Tebuconazole+Trifloxystrobin	0.5 g/L of Water
T7	Fluoxstrobilin	1 ml/L of Water
T8	Bordeaux Mixture 1%	(1:1:100)
T9	Azoxystrobin + Chlorothalnil	1 ml /L of Water
T10	Chlorothalonil+Thiophenate methyl	1 ml /L of Water
T11	Control	-

**Fruit Sampling:** Four to five branches were selected randomly from the selected trees from their different sides (south, north, west, east, center) to guarantee even representation.

**Physicochemical Attributes:** Vernier caliper was used to measure the diameters of polar and equatorial of the fruit in centimeters (cm), while fruit peel thickness was also measured with vernier caliper. Analytical balance was used to measure the weight of fruit in grams. Mechanical juice extractor was used for juice collection

and subsequently juice was weighed. Organoleptic quality was assessed, on the basis of hedonic scale having the range 0-10, by the method described by Krum (1955). A group of three judges evaluated the taste and flavor attributes of the fruits by consuming fruits individually/independently. Refractometer (Model RL 1372) was used to determine total soluble solids (TSS) contents in fruit juice. Horwitz (1960) method was used to determine total titratable acidity (TA) in the juice, and was expressed in percentage with the following formula:

$$\text{acidity (\%)} = \frac{\text{Volume of juice (mL)} \times \text{NNaOH} \times 0.0064 \times 100}{\text{Volume of juice taken for titration (mL)}}$$

The total soluble solids to titratable acidity (TSS/TA) ratio was computed by dividing the TSS (%) by the TA (%), as outlined by Horwitz (1960).

**Disease Assessment:**

- Visually inspect each fruit for symptoms of citrus scab, which include raised, corky, scabby lesions. The assessment during peak disease expression

$$\text{Citrus Scab Incidence (\%)} = \left( \frac{\text{Number of infected fruits}}{\text{Total Number of Fruits Observed}} \right) \times 100$$

**Experimental design and statistical analysis:** The experiment was laid out under Randomized Complete Block Design (RCBD) with 9 treatments having 4 replications with 3 number of plants /replication and a total of 108 plants. Significance among treatment of means were compared by using Tuckey Highest Significant Difference Test (HSD) at 5 percent level and treatment means were compared according to Steel and Torie (1980).

**RESULTS**

There was significant difference among treatments applied as foliar spray on kinnow plants. During 2017-18, the citrus minimum scab disease incidence (2.25%) on kinnow fruits was found in (Fruit) treatment Difenonazole + Azoxystrobin at par with Bordeaux mixture (2.50%), followed by Pyraclostrobin + Metiram (3.25%), Copper Hydro Oxide (3.75%). Among chemicals least control/maximum scab incidence (6.25%) was observed Tebuconazole + Trifloxystrobin was applied. However, maximum scab incidence (22.25 %) was found

(usually mid to late fruit development stage) was conducted.

- Record the number of fruits showing typical scab symptoms per tree.

Calculation of Disease Incidence:

For each tree or treatment group, calculate the scab incidence (%) using the formula:

$$\text{Disease Incidence (\%)} = \left( \frac{\text{Number of infected fruits}}{\text{Total Number of Fruits Observed}} \right) \times 100$$

in control (Table 2). Significant results of chemical fungicides on kinnow plants were observed during 2018-19. The minimum citrus scab disease incidence (3.75%) on kinnow fruits was found in treatment Bordeaux mixture 1% followed by Copper Hydro Oxide (6.25%), Difenonazole + Azoxystrobin@1 ml/L of Water (8.25), and Tebuconazole + Trifloxystrobin (9.25). Among chemicals least effective influence for scab control was observed at Copper Oxychloride. Non treated plants (control) showed maximum disease incidence (32.25%) (Table 2).

During 2019-20, the minimum citrus scab incidence on kinnow fruits was found in treatment T8 (Bordeaux mixture 1%) (3%), followed by Difenonazole + Azoxystrobin@1 ml/L of Water (4%), and Copper hydro oxide (6%). Among chemical foliar applications maximum incidence (9%) was observed in Azoxystrobin, Pyraclostrobin + Metiram, Fluoxystrobin. However, overall maximum citrus scab infected in control (26%) (Table 3).

Table 2. Effect of various chemicals used to control citrus scab (*Elsinoe Fawcettii*) in Kinnow during 2017-18 and 2018-19

Treatments	Disease Incidence (%) 2017-18	Disease Incidence (%) 2018-19	Disease Incidence (%) (Fruit) 2019-20
T1 = Azoxystrobin	4.00±0.21 D	10.75±0.52 C	9±0.46 B
T2 = Pyraclostrobin+Metiram	3.25±0.15 E	11.25±0.51 C	9±0.43 B
T3 = Difenonazole+Azoxystrobin	2.25±0.11 F	8.25±0.42 E	4±0.23 E
T4 = Copper Oxychloride	3.75±0.19 DE	12.75±0.52 B	7±0.34 C
T5 = Copper Hydro Oxide	2.75±0.12 FG	6.25±0.31 F	6±0.31 D
T6= Tebuconazole+Trifloxystrobin	6.25±0.31 B	9.25±0.42 D	8±0.39 B
T7 = Fluoxastrobin	4.75±0.22 C	8.75±0.39 DE	9±0.43 B
T8 = (Bordeaux mixture 1%)	2.50 ±0.17 G	3.75±0.19 G	3±0.24 F
T9 = Chlorothalonil+Thiophenate methyl	4.70±0.22 C	8.70±0.39 DE	9±0.40 B
T10 = Azoxystrobin+Chlorothalnil	4.78±0.22 C	8.80±0.39 DE	9±0.40 B
T11 = Control	22.25±1.2 A	32.25±1.51 A	26±1.31 A

Means sharing different letters in a row or in a column are statistically significant ( $P \leq 0.05$ ). The values after ± indicate standard deviations

A significant difference ( $P \leq 0.5$ ) in yield was observed in plants treated with various chemicals. Average of three years data showed that maximum yield (82 Kg/plant)

was recorded in plants treated with Difenonazole + Azoxystrobin followed by Bordeaux mixture 1% (81 kg/plant). However, plants treated with Azoxystrobin

recorded 68 kg/plant, Pyraclostrobin + Metiram 73 Kg/plant, Copper Oxychloride 73 Kg/plant, Copper Hydro Oxide 78 Kg/plant, Tebuconazole + Trifloxystrobin 74 Kg/plant, Fluoxastrobin 77 Kg/plant, while control recorded minimum fruit yield (65 Kg/plant).

Table 3. Effect of various chemicals on fruit diameter, fruit length, fruit peel thickness, and yield of Kinnow (average of three years)

Treatments	Fruit size (mm)	Fruit Diameter (mm)	Peel Thickness (mm)	Yield (Kg/plant)
T1 = Azoxystrobin	53±2.6 C	67±3.2 CD	3.54±0.17 B	68±3.75 D
T2 = Pyraclostrobin+Metiram	52±2.1 D	68±3.4 C	3.49±0.14 C	73±3.79 C
T3 = Difenconazole+Azoxystrobin	57±2.4 AB	65±3.5 D	3.57±0.15 B	82±4.11 A
T4 = Copper Oxychloride	61±2.5 A	69±3.3 BC	4.05±0.16 A	73±3.87 C
T5 = Copper Hydro Oxide	56±2.4 B	67±3.4 CD	3.66±0.14 AB	78±3.45 AB
T6= Tebuconazole+Trifloxystrobin	60±2.5 A	72±3.7 A	3.51±0.15 BC	74±3.49 B
T7 = Fluoxastrobin	56±2.4 B	70±3.8 B	3.59±0.17 A	77±3.71 B
T8 = Bordeaux mixture 1%	53±2.2 C	66±3.8 D	3.53±0.18 AB	81±3.29 A
T9=Chlorothalonil+Thiophenate methyl	55±2.4 BC	66±3.4 D	3.50±0.14 BC	77±3.45 B
T10=Azoxystrobin+Chlorothalnil	54±2.4 CD	64±3.4 E	3.50±0.14 BC	76±3.45 AB
T11 = Control	53±2.3 C	61±3.4 F	3.54±0.13 B	65±3.36 E

Difenconazole+Azoxystrobin (17%) and Bordeaux mixture 1% (16.6%) showed significantly ( $P \leq 0.5$ ) higher titratable acidity (%) during 2017-18. Similarly, during 2018-19, the maximum titratable acidity (%) was calculated on plants treated with Difenconazole+Azoxystrobin (16.3%) and Bordeaux

mixture 1% (17.5%). However, during 2019-20, the maximum titratable acidity (%) was calculated on plants treated with Difenconazole+Azoxystrobin (16.7%) and Bordeaux mixture 1% (17.6%) during 2019-20. While minimum value of TA was found in control in three consecutive years (Figure 1).

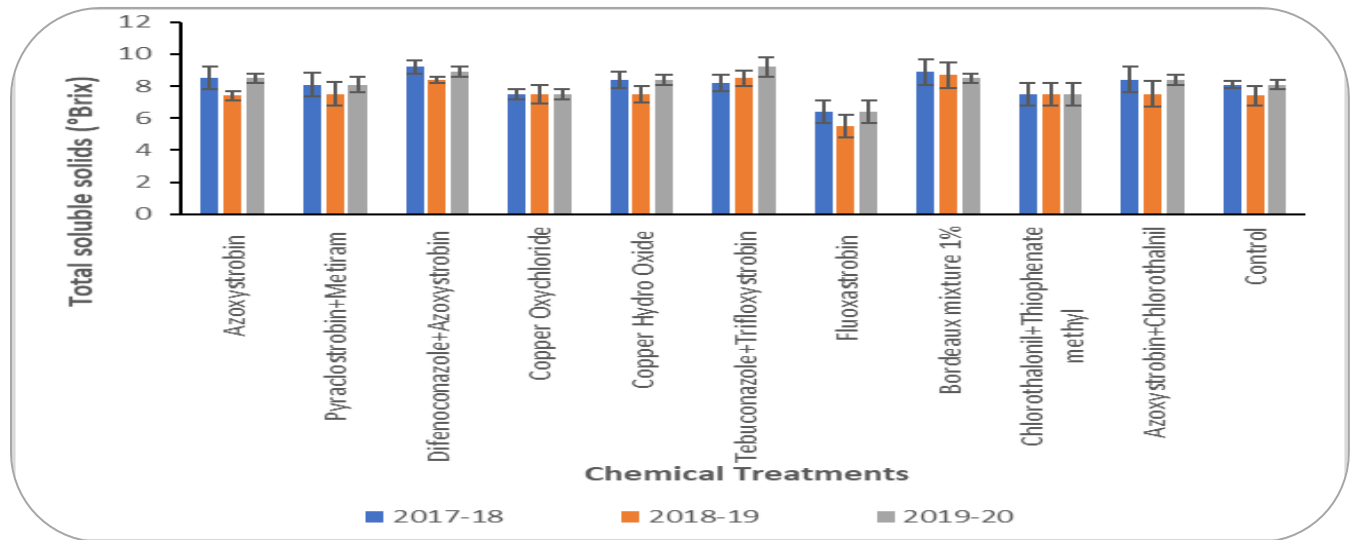


Figure 1. Effect of various chemicals on titratable acidity (%) of Kinnow during 2017-18, 2018-19 and 2019-20.

There was significant difference ( $P \leq 0.5$ ) among treatments applied regarding TSS. The maximum total soluble solids were calculated on plants treated with Difenconazole+Azoxystrobin (9.2°Bx) and Bordeaux mixture 1% (8.9°Bx) during 2017-18. However, during 2018-19, the maximum total soluble solids were calculated on plants treated with Difenconazole +

Azoxystrobin (9.2°Bx) and Bordeaux mixture 1% (8.7°Bx). However, during 2019-20, the maximum total soluble solids (Brix) were calculated on plants treated with Difenconazole+Azoxystrobin (8.9°Bx) and Tebuconazole+Trifloxystrobin (9.2°). While on overall bases minimum TSS was recorded in control during consecutive years (Figure 2).

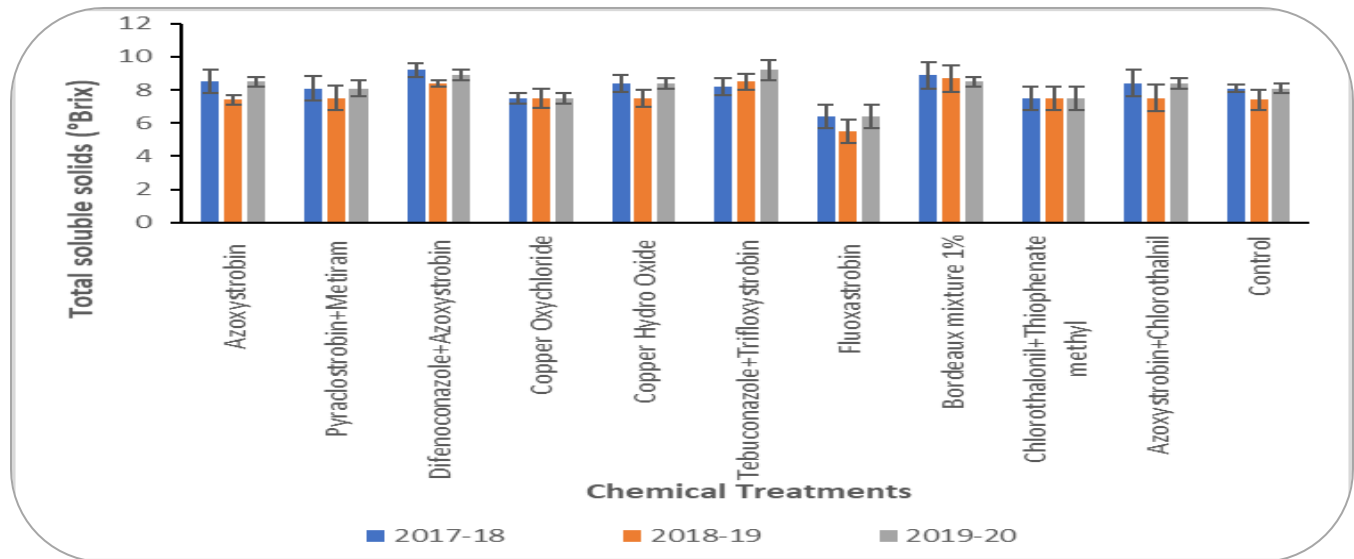


Figure 2. Effect of various chemicals on total soluble solids (°Brix) of Kinnow during 2017-18, 2018-19 and 2019-20.

A significantly ( $P \leq 0.5$ ) higher average fruit juice weight (g) was calculated on plants treated with Difenoconazole+Azoxystrobin (8.9 g) and Bordeaux mixture 1% (9.3 g) during 2017-18. However, during 2018-19, it was observed that maximum average fruit juice weight (g) was calculated on plants treated with Difenoconazole+Azoxystrobin (8.3 g) and Bordeaux mixture 1% (8.7 g). However, during 2019-20, the

maximum average fruit juice weight (g) was calculated on plants treated with Azoxystrobin at par with Difenoconazole+Azoxystrobin (9.3 g) and Bordeaux mixture 1% was at par with Tebuconazole+Trifloxystrobin (9.2 g). While during three consecutive years on overall bases, the minimum fruit juice weight was found where no chemical was sprayed (control) (Figure 3).

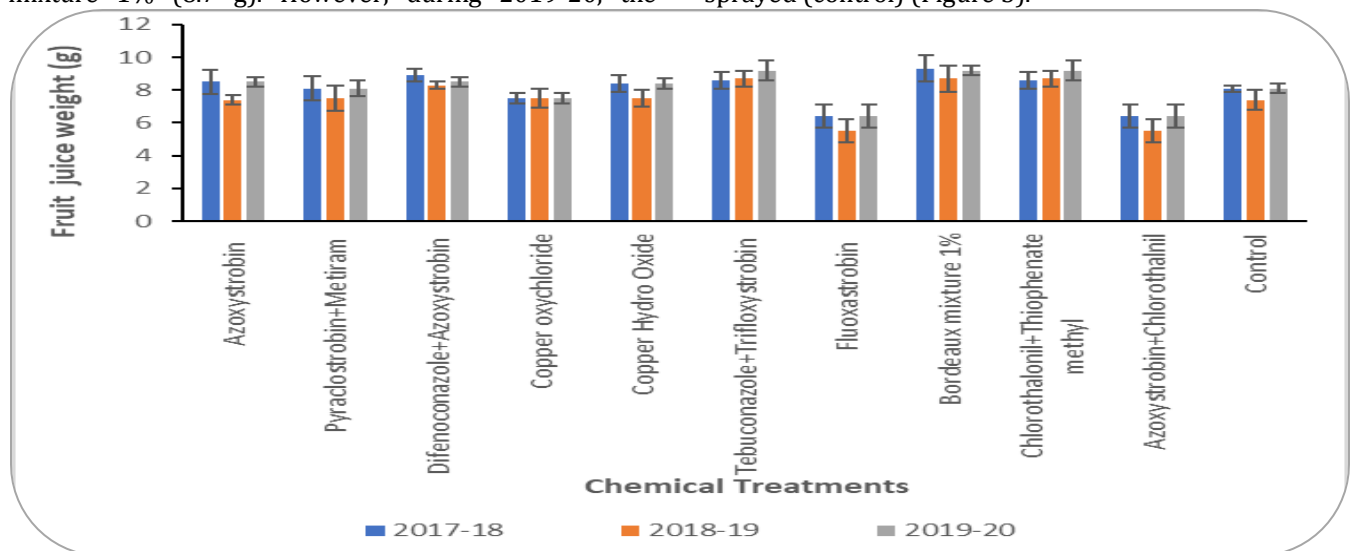


Figure 3. Effect of various chemicals on fruit juice weight (g) of Kinnow during 2017-18, 2018-19 and 2019-20.

A significantly ( $P \leq 0.5$ ) maximum average peel of fruit weight (g) was recorded on plants treated with Difenoconazole + Azoxystrobin (34.2 g) and Bordeaux mixture 1% (34.6 g) during 2017-18. However, during 2018-19, the maximum average fruit juice weight (g) was calculated on plants treated with Difenoconazole + Azoxystrobin (34.6 g) and Bordeaux mixture 1% (33.9 g) during 2019-20. However, during 2019-20, the

maximum average fruit juice weight (g) was calculated on plants treated with Azoxystrobin at par with Difenoconazole + Azoxystrobin (34.7 g) and Bordeaux mixture 1% was at par with Tebuconazole + Trifloxystrobin (33.5 g). While during three consecutive years on overall bases, the minimum average peel of fruit weight (g) was found where no chemical was sprayed (control) (Figure 4).

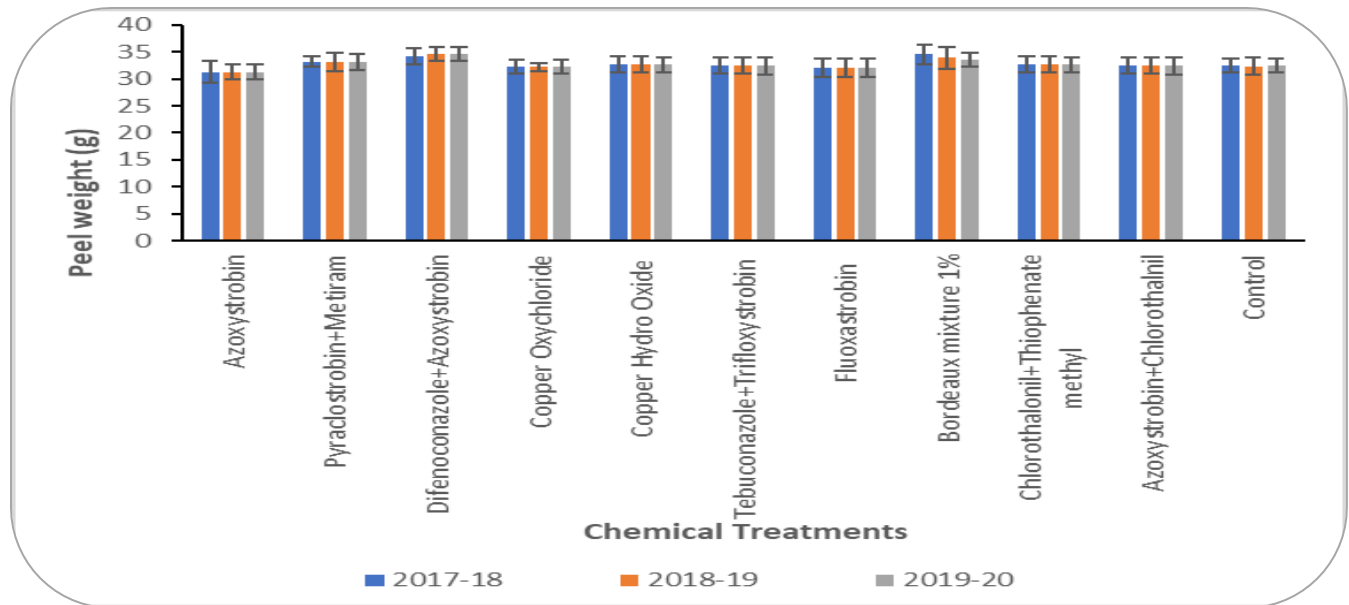


Figure 4. Effect of various chemicals on peel weight (g) of Kinnow during 2017-18, 2018-19 and 2019-20.

There was significant difference ( $P \leq 0.5$ ) in mean values of rag weight when foliar spray of selected fungicides was applied. The maximum average rag of fruit weight (g) was calculated on plants treated with Difenoconazole + Azoxystrobin (43.7 g) and Bordeaux mixture 1% (43.7 g) during 2017-18. However, during 2018-19, the maximum average rag weight (g) was calculated on plants treated with Difenoconazole + Azoxystrobin (43.4 g) and Bordeaux

mixture 1% (43.7 g). However, during 2018-19, the maximum average rag weight (g) of fruit was calculated on plants treated with Azoxystrobin at par with Difenoconazole + Azoxystrobin (34.7 g) and Bordeaux mixture 1% was at par with Tebuconazole + Trifloxystrobin (43.9 g). While during three consecutive years on overall bases, the minimum average rag of fruit weight (g) was found where no chemical was sprayed (control) (Figure 5).

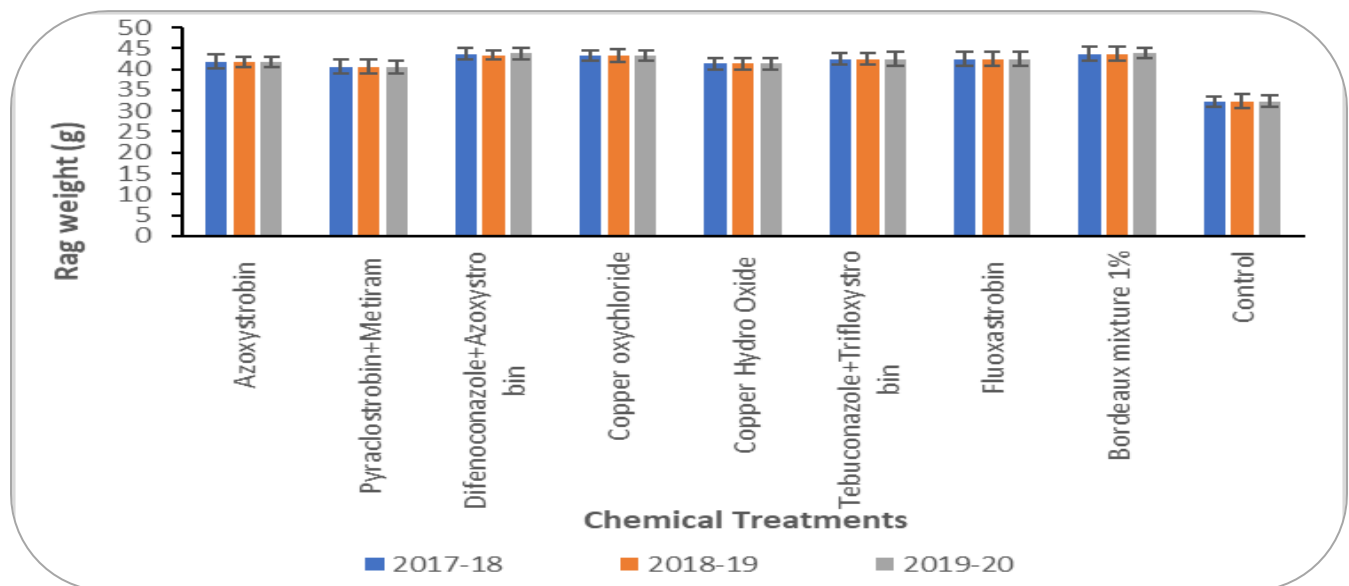


Figure 5. Effect of various chemicals on rag weight (g) of Kinnow during 2017-18, 2018-19 and 2019-20.

**DISCUSSION**

In present study a solution of Difenoconazole + Azoxystrobin was applied as foliar spray which was found

most effective in controlling citrus scab. Meena *et al.* (2025) demonstrated that the IPM treatment consistently exhibited the lowest mean densities of psyllid and

whitefly populations, as well as minimal severity of sooty mold and dieback, in comparison to conventional farmer practices and controls. Disease progression, quantified via the Area Under Disease Progress Curve, and pest population fluctuations, monitored from the 11<sup>th</sup> to 52<sup>nd</sup> Standard Meteorological Week (SMW), revealed a treatment-dependent trend, with maximum infestation, followed by farmer practice and IPM. Interestingly, the highest populations of natural enemies (coccinellids, *Chrysoperla* spp., and spiders) were observed in the untreated plants (control), followed by the IPM and then the conventional practice. Economically, IPM treatment yielded the highest average fruit production and the most favorable benefit-to-cost (B:C) ratio compared to farmer practices and controls.

Our results of Difenoconazole+Azoxytrobina are similar to a previous experiment on citrus conducted by Mondal *et al.* (2005), the influence of salicylhydroxamic acid, an alternative oxidase inhibitor, on the sensitivity of five fungal species to azoxytrobina and pyraclostrobina was evaluated. Given that *Alternaria alternata* exhibited mycelial insensitivity to azoxytrobina, the effect of azoxytrobina alone and in combination with SHAM was assessed specifically at the spore germination stage for this species. Arutselvan (2023) revealed that in citrus disease management Bordeaux mixture has special attention due to its unique properties of developing immunity in citrus plants.

A field trials of Wang *et al.* (2016), revealed that azoxytrobina alone, as well as in combination with difenoconazole, provided high levels of disease suppression against tobacco brown spot. Three applications of azoxytrobina at 0.094, 0.19, and 0.28 kg a.i./ha, and the Azoxytrobina+Difenoconazole mixture at 0.15, 0.22, and 0.29 kg a.i./ha achieved control efficacy ranging from 86.00% to 89.67% and 86.14% to 89.23%, respectively. In contrast, difenoconazole alone (0.12 kg a.i./ha) resulted in moderate disease control, with efficacy ranging from 55.14% to 58.41%. These results indicate that azoxytrobina, either individually or in combination with difenoconazole, represents a promising chemical control strategy for managing tobacco brown spot under field conditions.

The scab is a fungal disease which infected the fruits in current research however, in one of previous research described that antifungal efficacy of ten fungicides—thiophanate-methyl, difenoconazole, hexaconazole, Metalaxyl + Mancozeb, metiram, mancozeb, azoxytrobina,

dodine, chlorothalonil, and fosetyl-aluminum—was evaluated using the poisoned food technique at concentrations of 25, 50, 100, 200, and 400 ppm. The results indicated that the majority of fungicides demonstrated maximum mycelial growth inhibition at the highest concentration tested (400 ppm). Specifically, Metalaxyl + Mancozeb, mancozeb, thiophanate-methyl, metiram, and fosetyl-aluminum exhibited inhibitory effects of 100%, 84.86%, 65.57%, 52.96%, and 49.55%, respectively, at 400 ppm. A premix formulation of azoxytrobina and difenoconazole exhibited substantial antifungal activity, achieving 94.95% inhibition at 300 ppm. Notably, difenoconazole, hexaconazole, dodine, and chlorothalonil completely suppressed *A. citri* mycelial growth across all tested concentrations, indicating their superior efficacy. Jamar *et al.* (2017) also proved that optimum level of lime (a kind of Bordeaux mixture) is best for protection against citrus scab.

In present research the incidence of disease was due to the fact that infected citrus tissues typically develop erumpent, wart-like scab lesions, which are indicative of fungal colonization and associated cellular damage. Elsinochrome belongs to the perylene quinone class of fungal phytotoxins. These compounds act as photosensitizers, capable of absorbing visible light and facilitating the generation of reactive oxygen species (ROS), such as superoxide radicals and singlet oxygen, through interaction with molecular oxygen. These ROS are responsible for oxidative membrane damage, lipid peroxidation, and electrolyte leakage, culminating in localized host cell death and symptom expression (Prieto *et al.*, 2024).

The present study aimed to detect and identify the scab-causing fungal species affecting citrus cultivars in Punjab, Pakistan. A total of 46 symptomatic citrus samples were collected and subjected to PCR-based analysis. Of these, 13 samples tested positive for citrus scab. Based on the amplicon size observed in gel electrophoresis, the causal agent was putatively identified as *Elsinoë australis* (Jyothi and Shilpa, 2020).

During current research, applications time was carefully selected i.e., applied before flowering in March, after fruit setting in April and in the months of August. Diseased data for citrus scab disease was collected /recorded after fifteen days of the last spray. In one of previous research, the efficacy of disease control declined progressively with increasing intervals between fungicide application and pathogen inoculation, findings indicate that applications

of pyraclostrobin during the spring flush of citrus trees are significantly more effective in managing melanose, scab, and *Alternaria* brown spot compared to treatments with famoxadone or copper hydroxide (Mondal *et al.*, 2007). Khan *et al.* (2008) conducted research and proved that juice content exhibited dynamic variation influenced by multiple factors, generally increasing with fruit maturation and subsequently declining post-physiological maturity. Throughout the observation period, juice content remained inconsistent. 'Feutrell's Early' recorded the highest juice content (52.50%) on December 3<sup>rd</sup>, which declined to the lowest value on December 30<sup>th</sup>. Fruits picked on December 18<sup>th</sup> and January 30<sup>th</sup> had significantly ( $P < 0.05$ ) low juice contents compared to other treatments. Highest juice content, i.e., 55.50%, was noted in fruits picked/harvested on April 23<sup>rd</sup>, while the lowest (49.70%) was recorded on 7<sup>th</sup> January. Fruit juice content was recorded high during ripening compared to early stages; however, delayed harvesting reduced juice content, showing deterioration in juice content after maturity. This shows that quality of fruits is significantly affected by over-ripening (Gill *et al.* (1986). The difference in yield may be attributed to climatic variations during the period of research. Our research reveal that combination of both fungicides Difenconazole and Azoxystrobin provides very effect control of citrus scab owing to their synergistic effect. Both fungicides have been reported to be lethal to scab fungus by disrupting its fungal growth and cell membrane integrity, and ultimately hindering its energy production. Our results showed very promising control of citrus scab disease with these two fungicides.

#### CONCLUSION

Three consecutive year's study concluded the superior efficacy of difenoconazole and azoxystrobin foliar spray in controlling citrus scab in Kinnow Mandrin. Among systemic fungicides difenoconazole+azoxystrobin and Bordeaux mixture 1% offer significantly better disease suppression and yield improvement than Fluoxstrobin and control (no foliar spray of any chemical). Further 1% Bordeaux mixture effectively controlled the incidence of scab.

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Malik A. Rehman	:	Conceived the data and supervised the research
Shafqat Ali	:	Proof-read the paper
Mujahid Ali	:	Executed the research
Salman Ahmad	:	Analyzed the data
Bushra Zulfiqar	:	Helped in research
Erum Rashid	:	Formatted the paper