Egyptian Journal of Phytopathology,Vol. 52, No.1, pp 67-82 (2024)
DOI 10.21608/EJP.2024.358825

ORIGINAL PAPER

Impact of some agrochemical products on early fruit drop of certain Egyptian mango cultivars induced by fungal infection

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Received: 19 April 2024 / **Accepted**: 5 June 2024 / **Published online**: 7 June 2024

ABSTRACT

Early fruit drop is one of the most important diseases that usually decrease the production of mango trees in Egypt. The typical symptoms of early fruit drop were observed on four surveyed Egyptian mango cultivars (Hindy, Ewais, Keitt, and Montakhab El-Qanater) during 2022 and 2023 seasons. Among these cultivars, Montakhab El-Qanater recorded the highest percentage of fruit drop while Ewais showed the lowest percentage in both inspected seasons. *Fusarium moniliforme* Sh., *Lasiodiplodia theobromae* Pat. and *Alternaria alternata* (Fr.)Keissler, were isolated from early dropped fruits of inspected cultivars and recorded the highest frequencies, respectively. Pathogenicity test showed that isolated fungi were able to induce the early fruit drop disease with typical symptoms. Montakhab El-Qanater cv. was the most susceptible for fungal infection compared to Ewais cv. Infection with each of *F. moniliforme*, and *L. theobromae* was inherent to the high percentage of fruit drop compared with *A. alternata*. *In vitro*, the efficacy of five commercial agrochemical products (Bellis, Craft, Dovex, Score and Tekto) was evaluated on the isolated fungi. Generally, the linear growth of any of the isolated fungi was inhibited with 0.5ml/L of Score, 0.5g/L of Bellis and 1ml/L of Tekto, respectively. *In vivo*, the treatment with Score (50ml/100L) significantly decreased early fruit drop during the two experimental seasons on Montakhab El-Qanater and Ewais cultivars. The lowest efficacy was obtained with Craft (40ml/100L) and Dovex (25ml/100L). The physiological effects of the evaluated products were measured on total phenolic content and enzymatic activities of chitinase (CHT), peroxidase (PO) and polyphenol oxidase (PPO) in leave extract of Montakhab El-Qanater and Ewais cultivars. The total phenolic content and enzymatic activities were significantly promoted in both cultivars with the treatments of the evaluated chemicals. The highest values of total phenolic content in both cultivars were detected with Score, Bellis and Tekto, respectively. While, the greatest activity of measured enzymes was revealed with Bellis, Score and Tekto, respectively.

Keywords: Mango fruit drop, *F. moniliforme*, *L. theobromae*, fungal inhibition, enzymes activity and phenolic compounds.

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INTRODUCTION

Mango is one of the most important fruits in Egypt. Globally, Egypt ranks eighth in mango production and annually its production close to 1.5 million metric tons (FAO 2021). Among the mango cultivars, there is considerable variation in the quantity of production, fruit size, color, shape, flavor, texture, taste, and susceptibility to diseases, Also, Mango is liable to infection by several plant pathogenic fungi *i.e.*, blossom blight (anthracnose), dieback, fruit rot, root rot, malformation, powdery mildew, and early fruit drop of mango (Ragab *et al*., 1971; Abdalla *et al*., 2003; Ismail *et al*., 2012). In addition, early fruit drop of mango is the most disease affecting almost all commercial cultivars causing considerable reduction of quantity and quality of mango yields with different degrees, moreover the climatic changes can play important role in cultivars sensitive (reaction) to infection by the causal pathogens of mango fruit dropping. Egypt has a wide range of mango varieties, local traditional varieties, and newly introduced mangoes. Although the

best traditional varieties are Ewais and Hindi, the productivity of these varieties is recently decreased. Therefore, continuous efforts should be exerted to enhance the yield and quality for Egyptian varieties of mango (El-sheshetawy 2016). Under high humid conditions as prevailing in west Bengal excessive fruit drop and low fruit retention are important factors that determine the final yield in mango (Piyalid *et al*., 2017). Early fruit dropping is a disease of economic importance which occurs throughout India and other countries (Rattanpal, 2012). Regarding citrus trees, growers of Kinnow (*Citrus nobilis* × *Citrus deliciosa* L.) suffer a greater economic loss when early fruit dropping is accelerated by pathogenic infection, insects' injury, and physiological or hormonal imbalance due to poor orchard management. Fruit drops occur at various stages of fruiting and are categorized as pathological, insects and physiological fruit dropping (Patil, 2011). Citrus insects such as mealy bugs cause infestation on most of the part of citrus plants; a sooty mold develops which reduces photosynthetic activities and attracts pathogens to induce fruit dropping (Rattanpal, 2012). Ibrahim *et al*., (2007) found that species of fungi cause early fruit drop such as *A. altirnata*, C. *gloeosporioide*, *Botrytis* spp. and many other different fungi. As co-operative effect, fruit drop disease symptoms displayed rapidly under humid and warm conditions and ultimately infect the fruit, leading to fruit drop. Besides biotic factors, abiotic factors such as high temperature, water deficits, and wind velocity in the area also contribute to excessive early fruit dropping. The plant defense against pathogens often depends on attack through biochemical reactions that produce toxic substances to the pathogen or creating conditions that inhibit its development (Andersen *et al*., 2018). Plant induced resistance against plant pathogens occurs when any of these substances reach a concentration sufficient to inhibit the infection (Díaz-Puentes, 2009).The defensive response involves processes such as accumulation of phenolic compounds and induction of enzymatic activities (Cornide *et al*., 1994).The plant defense against pathogens depending on systemic acquired resistance in most cases is equally effective against the fungi, bacteria and viruses (Díaz-Puentes, 2012).This study focused on early fruit dropping after fruit-set especially those trees which showed outwardly natural early fruit drop, and internal symptoms of dropped fruits showing core browning in the field. In Egypt, to date much information is needed for the management of fruit dropping in mango. Furthermore, there are not enough studies that demonstrate the reaction of plant defense against fungal disease of tropical fruit diseases, such as mango. In the present study the reduction of early fruit dropping on both Montakhab El-Qanater and Ewais cultivars was measured by using, five available agrochemicals; Score 25% EC at 50ml/ 100 L, Bellis 38%WG 50g/ 100L, Tekto 50%SC 100ml /100L, Craft 30% EC40ml / 100L and Dovex 50% SC 25ml/ 100L as a spray on the trees after fruit-set. Also, the activity of defense-related enzymes (chitinase, polyphenol oxidase and peroxidase) as well as total phenolic content were determined. Therefore, the obtained results could provide more information for researchers and agronomists concerning manage the early fruit dropping of mango.

MATERIALS AND METHODS

1. Survey of mango fruit drop disease:

The present study was carried out during the two inspected seasons 2022 and 2023 in different orchards located at Qalioubia and Giza governorates, Egypt. Survey of mango fruit dropping was performed on trees (15-year-old) of four Egyptian traditional mango varieties; Hindy, Ewais, Keitt, and Montakhab El-Qanater which showed early fruit dropping, with internal documented symptoms. The survey included 15 trees; 3 replicates were randomly chosen for each cultivar. The percentages of fruit dropping were recorded during April and May each year (Fig. 3).

Percentages of drop (%) was calculated using the following formula:

Percentages of drop (%) for trees= Number of mango trees showing fruit drop/Total number of tested mango trees × 100.

Percentages of drop (%) for fruits=Number of dropped fruits (one month after full fruit setting) /Total number of tested fruits in mango tress × 100.

2. Fungi associated with fallen fruits:

Dropped mango fruits (one month after full fruit setting showing core browning in the field) were collected from the fields of Qalioubia and Giza governorates during 2022 and 2023 seasons. Isolation was carried out from two parts, stem end (pedicel) and internal parts of mango fruit. For isolation trials, samples were collected from trees showing dropped fruits with internal sections showing core browning. Samples were brought to the laboratory and washed thoroughly with sterilized distilled water (*SDW*). The infected tissues were cut into small portions 0.5 cm, surface sterilized by dipping in 1% sodium hypochlorite for 0.5 min., followed by three subsequent washings with *SDW*, then they were dried between two folds of sterilized filter paper. The sterilized portions were placed aseptically in 9 cm petri dishes containing PDA medium. The plates were then incubated at 25 ±2 °C for 7 days for development of fungal growth. The growing fungal colonies were purified using the hyphal tip technique. Purified fungi were identified based on their morphological characteristics, according to Sutton (1980). Pure culture stocks of the isolated fungi were kept on PDA slants at 4°C for further studies. The frequency (%) of each fungal species was calculated by following equation:

Frequency (%) = Number of colonies of each fungal species/total number of all fungi×100.

3. Pathogenicity test:

Pathogenicity tests were performed on healthy mango fruit (one month after full fruit setting) cv. Ewais and cv. Montakhab El-Qanater in a private mango orchard located on Qalioubia governorate, Egypt. Conidial suspensions of the isolated fungi, *Fusarium moniliforme*, *Lasiodiplodia theobromae* and *Alternaria alternata*, were prepared from 7-8 days-old cultures grown on PDA in petri plates (∅9cm) according to (Hussien *et al*. 2018) and adjusted to 10^6 spores/ml. The fruits were injected, each fruit with 0.1ml of the spore's suspension using a micropipette according to (Khamis *et al*. 2015). The selected trees were uniform in vigor as possible. The fertilization program and other agricultural practices were the same for all trees. Three replicated (two trees per replicate and 5 fruits per each tree). The same number of fruits was injected with water as a control. The wounds were covered with melted wax 50°C (Deverall, 1967). Percentages of dropped fruits were calculated 30 days after artificial inoculation by the following formula:

$$\text{Percentage of dropped fruits}(\%) = \frac{\text{Number of droped fruits}}{\text{Total number of inoculated fruits}} \times 100$$

4. Bioassays of the evaluated agrochemicals:

Five agrochemicals (Table 1) were evaluated In vitro on the linear growth of isolated fungi, and *In vivo* on experimented mango cultivars.

4.1. *In vitro* experiment:

The effects of each agrochemical concentration on the linear growth of *F. moniliforme*, *L. theobromae*, and *A. alternata* were assessed *in vitro*. A 5 mm Ø fungal mycelia plug, taken from the developing edge of one week old culture was placed in the middle of a petri dish contained 10 ml PDA media adjusted with the concentrations of the therapies that were being evaluated. As a control, untreated PDA plates were inoculated with desired fungus. The entire experiment was done twice, with three plate replicates for each treatment. The diameter of colony growth (mm) for each fungus was measured after 5-7 days of incubation at 25±2°C. The reduction in colony growth was calculated

according to the formula suggested by Abd-Alla *et al.* (2014) as follows: Reduction in colony growth (%) = Control - Treatment / Control × 100.

Table (1): Tested Compounds.

Product	Active ingredients	Status	Conc./100 L water
Craft 30%	Difenoconazole 15%+ Propiconazole 15%	EC	40 mm
Dovex 50%	Azoxystrobin 20% +Tebuconazole 30%	SC	25mm
Score 25%	Difenoconazole 25%	EC	50 mm
Bellis 38%	Boscalid 25.2% + Pyraclostrobin 12.8%	WG	50g
Tekto 50%	Thiabendazole	SC	100 mm

4.2. *In vivo* experiment under field conditions:

This study was carried out in the 2022 and 2023 seasons in a 12-year-old cv. Montakhab El-Qanater and cv. Ewais mango orchard at El-Qanater El-Khayria Horticultural Research Station, Agricultural Research Centre, to assess the effectiveness of the agrochemical products listed before that have been approved by the Ministry of Agriculture, Egypt, for preventing early mango fruit dropping in natural field settings. As usual, the suggested horticultural techniques were followed. Each treatment was applied to 6 trees as three replicates. Every therapy was sprayed four times, separated by fourteen days. The above-mentioned suggested dose was used to spray the investigated chemicals for the first time during the start of fruit set. The same number of trees was sprayed with water to serve as a control. The percentage of dropped fruits was calculated as mentioned previously. Efficiency of the tested treatments was calculated after 60 days of the first application of the tested agrochemical products according to the following formula:

$$\text{Efficiency}(\%) = \frac{\text{Mango fruit drop (\%) in the control} - \text{mango fruit drop(\%)in the treatment}}{\text{Mango fruit drop (\%) in the control}} \times 100$$

5. Impact of the evaluated products on defense related enzymes and total phenolic content:

To determine the effect of the tested agrochemicals, Craft 30%EC 40mm /100L, Dovex 50% SC 25mm/100L, Score 25% EC 50 mm /100L, Bellis 38% WG 50g/100L and Tekto 50% SC 100 mm/100L water on the defense enzyme activities and total phenolic content in two mango cultivars, Ewais and Montakhab El-Qanater. Mango leaf samples were taken from the plants 48 hours following the most recent field application of the tested agrochemical products and the control. Each sample was comprised from three replicates of at least six distinct mango trees and immediately put in liquid nitrogen to measure the total phenolic content, chitinase (CHT), peroxidase (PO), and polyphenol oxidase (PPO) activities. (Monir *et al*., 2021; Shehata and Hassan 2023).

Leaves extraction:

To determine the activity of chitinase enzyme (CHT), peroxidase (PO) and polyphenol oxidase (PPO) enzymes as well as total phenolic contents, using glass homogenizer, 10g fresh mango leaves and 0.1sodium phosphate buffer PH=7 at 100 mg/1 ml containing 14 mM β-mercaptoethanol at the rate 1/3 w/v. The homogenates centrifuged for 40 min at 1500 rpm at 5^{o}C and stored at 4±1°C for further studies.

Chitinase activity (CHT):

Chitinase (CHT) activity was measured according to Boller, *et al*. (1983). To prepare the reaction mixture, 1ml mango crude extracts, 1 ml of phosphate buffer pH=7, 1 mL of dinitrosalycilic acid and 2

ml of 0.3% (w/v) colloidal chitin were incubated at 37ºC for 30 min.

Polyphenol oxidase activity (PPO):

Polyphenol oxidase (PPO) activity was measured according to Matta and Dimond (1963). To prepare the reaction mixture, 1ml mango crude extract, 1ml solution of phosphate buffer pH=7, 1ml catechol and the tube was completed with distilled water to 5ml.

Peroxidase activity (PO):

Peroxidase (PO) was measured according to Allam and Hollis (1972). The reaction mixture was made up of 1 ml of crude mango extract, 0.5 ml of phosphate buffer solution (pH = 7), 0.3 ml of pyrogallol, 1 ml of H_2O_2, and 5 ml of distilled water in the tube.

The activity of the enzymes was measured with a spectrophotometer, and the results were represented as changes in the mixtures absorbance at 425 nm for peroxidase, 495 nm for polyphenol oxidase, and 540 nm for chitinase every 0.5 minutes for five minutes.

Total phenolic content (TPC):

Total phenolic content (TPC) in mango leaves was evaluated according to Zieslin and Ben-Zaken (1993). To prepare the reaction mixture, 1ml mango crude extracts, 0.2 ml folin-Ciocalteau reagent, Sodium carbonate solution Na_2Co_3 7.5% (Dilute 7.5gm anhydrous sodium carbonate into 100m/L distilled water), 40 µl gallic acid and the tube was filled with distilled water to 5ml. TPC was expressed as the change in the absorbance of the mixtures every 0.5 min. for 5 minutes at 750 nm using Spectrophotometer.

Statistical analysis

All obtained data during both seasons were subjected to analysis of variance method according to Snedecor and Cochran (1990). Duncan's Multiple Range tested (Duncan, 1955) was used to compare differences among means.

RESULTS

1. Survey for mango fruit drop disease:

Survey during the two seasons 2022 and 2023 was carried out on some Egyptian traditional mango varieties in Qalioubia and Giza governorate. Typical symptoms of mango fruit drop disease were observed in all examined mango fruits where internal sections of dropped fruits showed core browning. Mango dropped fruits were observed in all examined locations. Data in Figs. (1, 2 and 3) show that Montakhab El-Qanater cv. showed the highest percentage of drop (%) for trees and percentage of drop (%) for fruits in the first and the second season. Meanwhile, Ewais cv. showed the lowest percentage of drop (%) in both seasons.

2. Fungi associated with mango fruit drop disease:

Pure cultures of the isolated fungi were identified as *Fusarium moniliforme* Sh., *Lasiodiplodia theobromae* Pat. and *Alternaria alternata* (Fr.)Keissler, based on culture characteristics and microscopic observation. Data in Table (2) show the three associated isolated fungi with mango early fruit drop symptoms. *F. moniliforme* showed the highest frequency from the two governorates during the two years of the investigation (49.07% - 46.29%). While *L. theobromae* recorded from 25.91% to 35.18% and the lowest frequency was *A. alternata* that recorded from 24.99% to 18.50%.

3. Pathogenicity test:

This trial was implemented to prove the responsibility of the most frequent isolated fungi regarding the fruit dropping of mango. Pathogenicity test revealed that all the recovered fungal species were able to induce early fruit drop of mango with typical outside and inside symptoms. Results in Table (3) and Figs (4, 5 and 6) reveal that the tested fungi were significantly differed in their pathogenic potentiality. The susceptibility of examined cultivars showed that the highest percentage of dropped fruits was inherited to Montakhab El-Qanater cv., and Ewais cv., respectively. During two successive years (2022 and 2023) *F. moniliforme* infection

was correlated with fruit drops and recorded the highest percentage on cv. Ewais and cv. Montakhab El-Qanater, being 81.49 and 86.66%, respectively. While infection of *L. theobromae* recorded 69.99 and 73.33 %. The lowest percentage of fruit drop was associated with *A. alternata* infection, being

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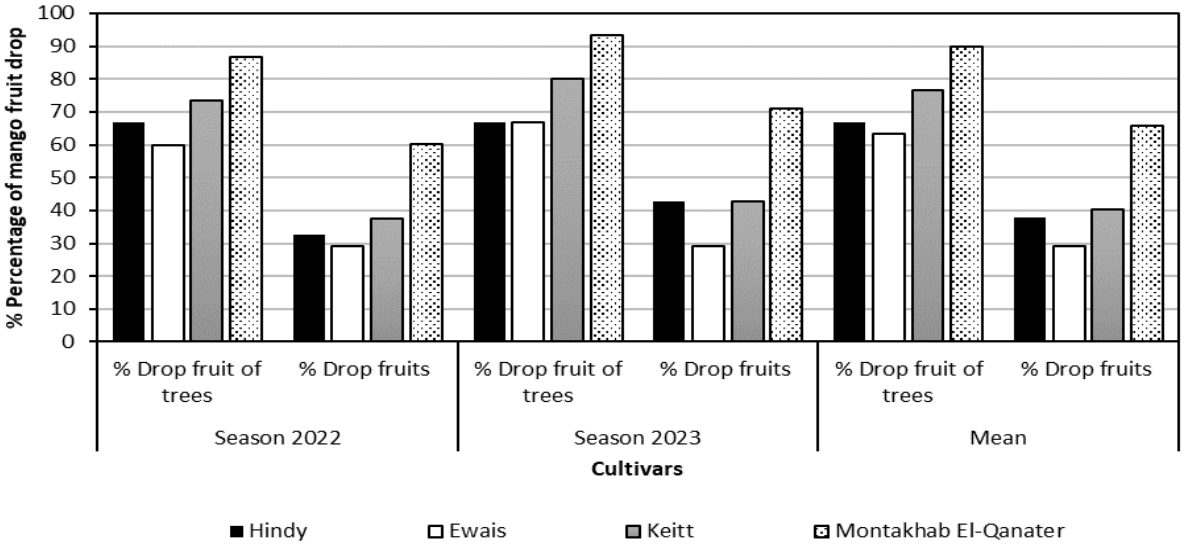

Fig. (1): Percentage of mango fruit drop on the surveyed cultivars in Qalioubiya governorate

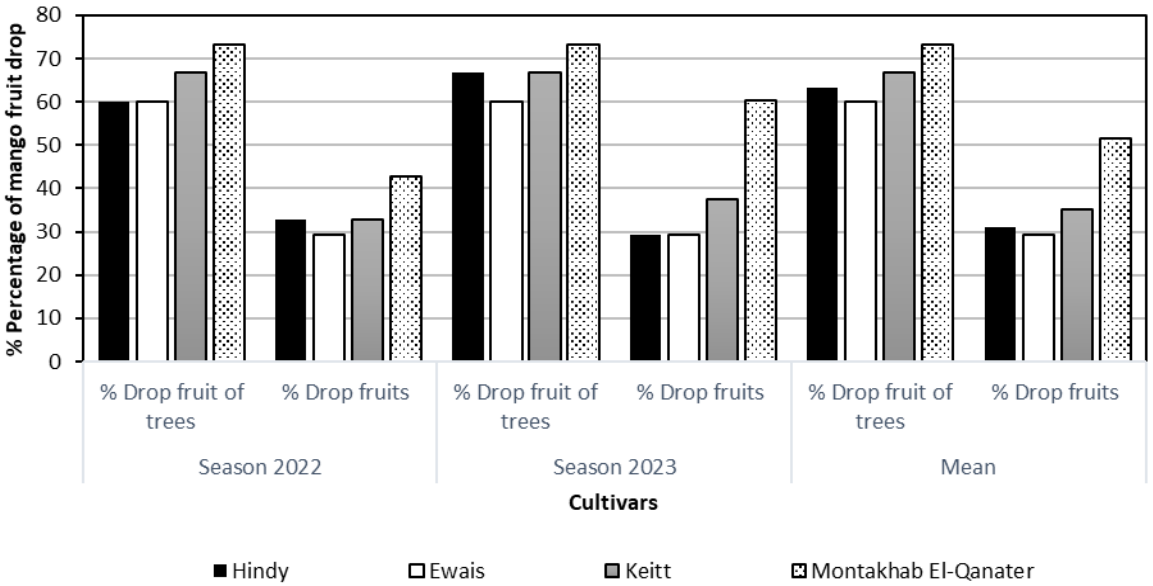

Fig. (2): Percentage of mango fruit drop on the surveyed cultivars in Giza governorate.

Table (2): Frequency (%) of fungi isolated from dropped mango fruits collected from the fields in Qalioubiya and Giza governorates during 2022 and 2023 seasons.

Isolated fungus	Isolation frequency %									
	2022					2023				
	Hindy	Eweais	Keitt	Montakhab El-Qanater	Mean	Hindy	Ewais	Keitt	Montakhab El-Qanater	Mean
L. theobromae	22.22b	25.92b	18.53c	37.03a	25.93b	37.03b	33.33b	25.92b	44.44a	35.18b
F. moniliforme	59.26a	55.55a	44.44a	37.03a	49.07a	44.44a	40.75a	55.55a	44.44a	46.29a
A. alternate	18.52c	18.53c	37.03b	25.94b	25c	18.53c	25.92c	18.53c	11.12b	18.53c
Total	100	100	100	100	100	100	100	100	100	100

Within each column, the same letter/s indicates no significant difference among treatments at ($p < 0.05$).

61.66 and 61.66 %, respectively. Data of the pathogenicity test also indicated that Montakhab El-Qanater was more susceptible than cv. Ewais during the two seasons.

Table (3): Percentage of fruit drop caused by the isolated fungi during 2022 and 2023 on cv. Ewais and cv. Montakhab El-Qanater.

Tested fungi	Percentage of drop (%)					
	2022			2023		
	Ewais	Montakhab El-Qanater	Mean	Ewais	Montakhab El-Qanater	Mean
L. theobromae	66.66b	73.33b	69.99b	70.00b	76.66b	73.33b
F. moniliforme	76.66a	86.33a	81.49a	83.33a	90.00a	86.66a
A. alternata	60.00c	63.33c	61.66c	56..66c	66.66c	61.66c
Uninfected	16.66d	26.66d	21.66d	16.66d	23.33d	19.99d

Within each column, the same letter/s indicates no significant difference among treatments at ($p < 0.05$).

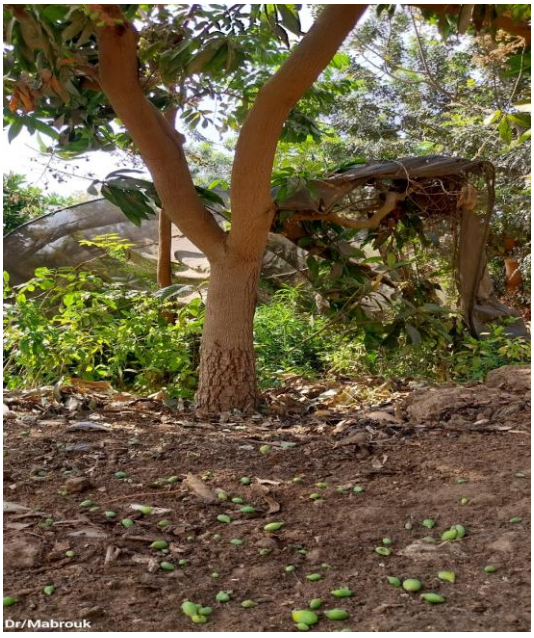

Fig. (3): Mango tree cv. Montakhab El-Qanater showing early fruit drop.

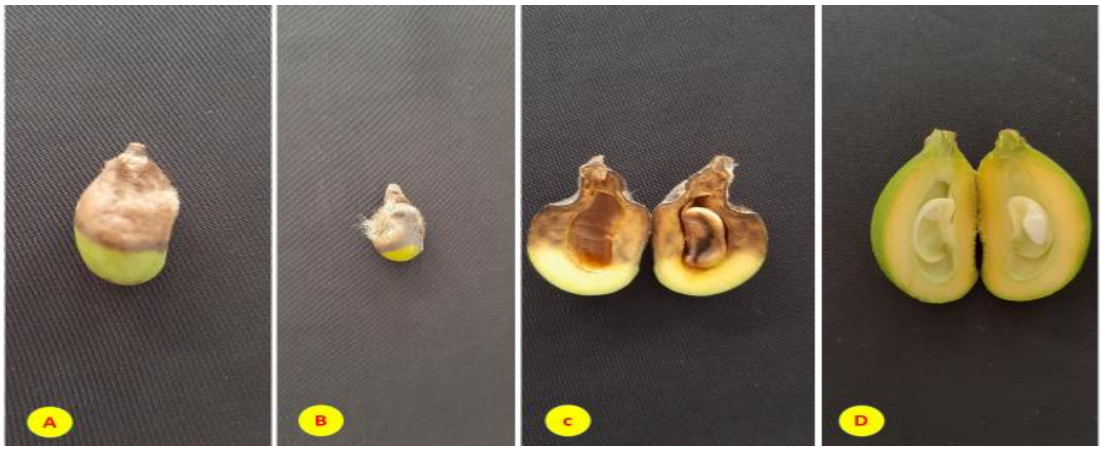

Fig. (4) : Symptoms of *Fusarium moniliforme* infection on inoculated immature fruits. (A-B); external symptoms, (C); internal symptoms showing browning of fruit core, (D); Uninfected fruit.

Fig. (5) : Symptoms of *Lasiodiplodia theobromae* infection on inoculated immature fruits. (A-B); external symptoms, (C); internal symptoms showing browning of fruit core, (D); Uninfected fruit.

Fig. (6): Symptoms of *Alternaria alternata* infection on inoculated immature fruits. (A-B); external symptoms, (C); internal symptoms showing browning of fruit core, (D); Uninfected fruit.

4. Evaluation of agrochemical products:

The efficiency of five tested commercial products namely, Score, Bellis, Tekto, Craft and Dovex were tested *in vitro* and *in vivo*.

4.1. *In vitro* experiment:

The obtained data in Table (4) reveal that the second concentration for all evaluated products had more effect on fungal linear growth compared to the first concentration for all the isolated fungi. The highest reduction was recorded by Score 25% EC, Bellis 38%WG and Tekto 50%SC, respectively compared to Craft 30% EC and Dovex 50% SC. Additionally, all isolated fungi mycelial growth was fully suppressed by Score 25% EC, Bellis 38%WG, and Tekto 50%SC (Table,4).

4.2. *In vivo* experiment:

Under field conditions, the experiments conducted during 2022 and 2023 seasons showed that all treatments using the tested agrochemicals reduced the early fruit drop compared with water treatment on both Ewais and Montakhab El-Qanater cultivars (Table 5). The highest efficacy was recorded by Score 25% EC. While 50g/ 100L of Bellis 38%WG and 100ml/100L of Tekto 50% SC showed a moderately effect. The lowest efficacy was determined by each of 40ml /100L of Craft 30% EC and 25ml/100L of Dovex 50% SC concentrations, respectively.

Table (4): Effect of the tested agrochemical products on mycelial growth of the isolated fungi *in vitro*.

Treatment	Conc. /L	*L. theobromae*		*F. moniliforme*		*A. alternata*		Mean	
		L(mm)	R%	L(mm)	R%	L(mm)	R%	L(mm)	R%
Craft 30% EC	0.2ml	45.0b	50	20.0b	77.77	15.0b	83.33	26.66b	70.37
	0.4ml	15.0e	83.33	10.0c	88.88	10.0c	88.88	11.66e	87.04
Dovex 50% SC	0.12ml	40.0c	55.55	20.0b	77.77	15.0b	83.33	25.0c	72.22
	0.25ml	25.0d	72.22	10.0c	88.88	10.0c	88.88	15.0d	83.33
Score 25% EC	0.25ml	15.0e	83.33	10.0c	88.88	10.0c	88.88	11.66e	87.04
	0.5ml	0.0f	100	0.0d	100	0.0d	100	0.0f	100
Bellis 38%WG	0.25g	15.0e	83.33	10.0c	88.88	10.0c	88.88	11.66e	87.04
	0.5g	0.0f	100	0.0d	100	0.0d	100	0.0f	100
Tekto 50%SC	0.5ml	15.0e	83.33	10.0c	88.88	10.0c	88.88	11.66e	87.04
	1ml	0.0f	100	0.0d	100	0.0d	100	0.0f	100
Water	--	90a	-	90a	-	90a	-	90a	-

Within each column, the same letter/s indicates no significant difference among treatments at ($p < 0.05$).
L= linear growth of mycelium & R%= the percentage of mycelial growth reduction

Table (5): Effect of the tested agrochemicals on the percentage of fruit drop of cv. Ewais and cv. Montakhab El-Qanater under field conditions.

Treatment	Conc./ 100L water	2022				2023			
		Ewais		Montakhab El-Qanater		Ewais		Montakhab El-Qanater	
		FD%	Eff.%	FD%	Eff.%	FD%	Eff.%	FD%	Eff.%
Craft 30% EC	40ml	20b	41.17	20.71c	50.00	26.25c	52.27	26.40b	53.52
Dovex 50% SC	25ml	20b	41.17	21.42b	48.28	27.5b	50.00	26.40b	53.52
Score 25% EC	50ml	15d	55.88	17.85e	56.90	18.75e	65.90	19.20e	66.19
Bellis 38% WG	50g	17c	50.00	19.28d	53.45	18.75e	65.90	20.80d	63.38
Tekto 50%SC	100ml	17c	50.00	19.28d	53.45	21.25d	61.36	23.2c	59.15
Water	--	34a	-	41.42a	-	55a	-	56.8a	-

Within each column, the same letter/s indicates no significant difference among treatments at ($p < 0.05$).
FD= fruit drop % & Eff= efficacy of the tested commercial product

5. Impact of the tested agrochemicals on the activity of defense related enzymes and total phenolic content:

Data in Figs (7, 8) show that all tested treatments increased the enzymatic activities of chitinase (CHT), peroxidase (PO) and polyphenol oxidase (PPO) as well as the accumulation of total phenolic content compared with untreated trees of Ewais and Montakhab El-Qanater cultivars, respectively. Concerning the enzymatic activities of all evaluated enzymes, they reached the highest values in both tested cultivars with the treatment of Bellis 38% WG, Score 25% EC and Tekto 50% SC, respectively. While the lowest values were recorded after treatment with Craft 30% EC and Dovex 50% SC, respectively. Meanwhile, the highest values of total phenolic content in the two tested mango cultivars were remarked with Score 25% EC, Bellis 38%WG and Tekto 50%SC, respectively. While the lowest amounts were recorded with Craft 30% EC and Dovex 50% SC, respectively.

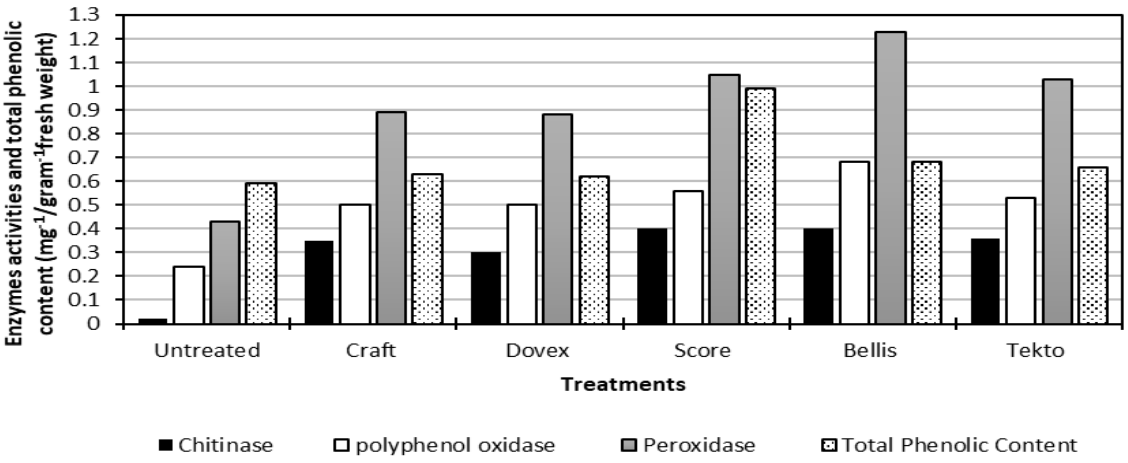

Fig. (7): Effect of the tested agrochemicals on the activity of defense enzymes and total phenolic content in mango Ewais cultivar

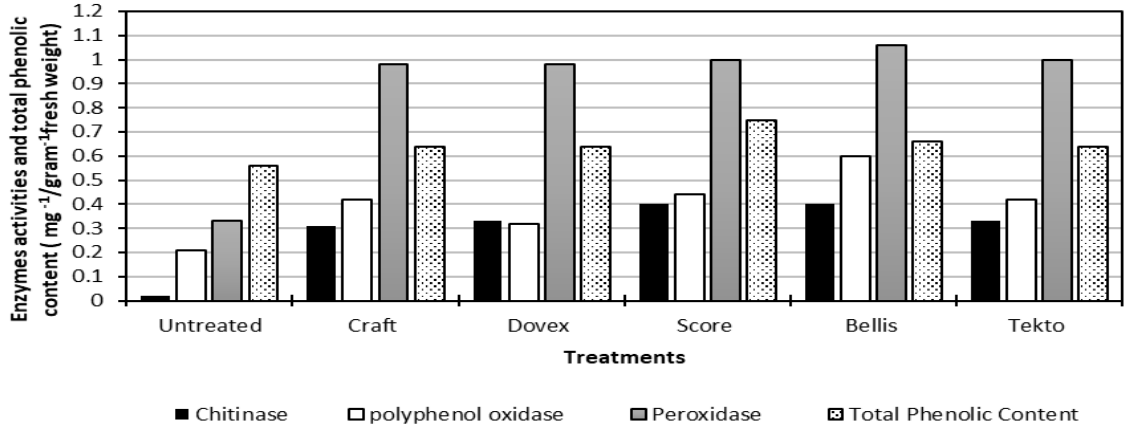

Fig. (8): Effect of the tested agrochemicals on the activity of defense enzymes and total phenolic content in mango Montakhab El-Qanater cultivar

DISCUSSION

Mango trees are affected by many factors that can cause early fruit drop disease, *i.e*. insufficient pollination, poor tree nutrition, diseases, insects, and several other factors. In the present study, mango fruits dropping was observed in surveyed governorates; Qalioubiya and Giza during 2022 and 2023. Montakhab El-Qanater cv. showed the highest percentage of fruit drop for trees and percentage of drop for fruits in the first and second seasons. While, Ewais cv. showed the lowest percentage of fruit drop in both seasons. Normally, mango trees produce large numbers of flowers of which only a small proportion set fruit (Kostermans and Bompard, 1993). Only perfect (hermaphrodite) flowers can set fruit. However, the number of perfect flowers per inflorescence varies between cultivars and also, from year to year, depending on the location of the inflorescence in the tree (Singh, 2005). Successful fruit set is dependent on the use of selective pollinizers (Ram *et al.*, 1976) and pollen viability has been considered to be a major factor limiting yields in mango (Davenport and Nunez-Elisea, 1997). Fruit drop might be caused by several factors, such as nutrient deficiency, disturbances in embryogenesis and/or embryo abortion, sink competition between fruits, and abiotic and biotic stress (Chadha, 1993). In a previous study, the same effect was recorded on guava trees, annually, guava fruit drop occurs during October and November, when the fruit accomplished the lemon size (Khamis *et al*., 2015). Among biotic agents, *F. moniliforme, Lasiodiplodia theobromae* and *Alternaria alternata* were isolated from dropped fruits. *F. moniliforme* recorded the highest frequency in surveyed governorates during the two seasons. While *A. alternata* recorded the lowest frequency. The pathogenicity test revealed that, all isolated fungi were able to induce early fruit drop of mango with typical external and internal symptoms, particularly *Fusarium moniliforme,* and *Lasiodiplodia theobromae*, respectively. *In vitro* investigation, the highest percentage of fruit drop was observed on Montakhab El-Qanater cv., and Ewais cv., respectively. The susceptibility of investigated cultivars showed that *F. moniliforme* infection was correlated with fruit drop and recorded the highest percentage on cv. Ewais and cv. Montakhab El-Qanater. While the lowest percentage of fruit dropped was associated with *A. alternata* infection. The previous study in Egypt documented that *L. theobromae* was considered as the main causal agent of fruit drop, fruit rot and dieback of mango (Ragab *et al.,* 1971) and Goudarzi, *et al*., (2021) in Iran. Also, under warm and humid conditions *A. altirnata*, *Colletotrichum gloeosporioide*, *Botrytis* spp and a many other different fungi can cause early fruit dropping (Ibrahim et *al*., 2007). Moreover, on apple fruit, *A. alternata*, *Penicillum expansum* and *P. ramulosum* were associated with core rot of apple fruit in South Africa (Combrink *et al*., 1985). Fruit drop of English walnut also associated with brown apical necrosis and caused by *A. alternata* and *F. semitectum* (Belisario *et al*., 2010). Also, *Altrnaria* spp. and *Colletotrichum cladosporioides* were the main pathogens causing core browning and moldy core of apple fruit in China (Gao *et al*., 2013). Similarly, *Colletotrichum acutatum* and *C. gloeosporioides* caused fruit drop in citrus after bloom (Marques *et al*., 2013). In Egypt, *A. alternata*, *Lasiodiplodia theobromae*, *Fusarium semitectum*, and *Pestalotia psidii*, were isolated from dropped guava fruit samples collected from different orchards (Khamis *et al*., 2015).

All evaluated products had significantly suppressed the linear growth of the isolated fungi. Particularly the double dose compared to the single dose of evaluated products. The highest reduction was recorded by Score 25% EC, Bellis 38%WG and Tekto 50%SC, respectively compared to Craft 30% EC and Dovex 50% SC. Also, Score 25% EC, Bellis

38%WG and Tekto 50%SC completely inhibited the mycelial growth of all isolated fungi. Under field conditions, all treatments of assessed agrochemical products reduced the early fruit drop compared with water treatment on both Ewais and Montakhab El-Qanater cultivars. The highest efficacy was recorded by Score 25% EC. While 50g/ 100L of Bellis 38%WG and 100ml/100L of Tekto 50% SC showed a moderate effect. The lowest efficacy was determined by 40ml /100L of Craft 30% EC and 25ml/100L of Dovex 50% SC, respectively. Generally, the management of the diseases and pests, particularly during flowering and fruit set, has a significant reduction in fruits drop and improvement yields. The emerging strobilurin class of systemic fungicides promises to make control even more effective (Goodwin and Clough 1997). Significantly, Score 25% SC (Difenoconazole) and Amistar 25% SC (Azoxystrobin) reduced incidence and severity of early blight and increased tomato fruit yields (El-Kholy *et al*., 2021). Additionally, Score 25% EC (Difenoconazole 25%) and Bellis 38% WG (25.2% boscalid+12.8% pyraclostrobin) had more effect on apple scab infections and yield productivity (Radwan and Hassan, 2019). Under field conditions, both Homai 80%WP (Thiophanate methyl 50%+Thiram 30%) and Kemazed 50% WP (Carbendazim) showed 78.8 and 69.0% reduction of guava fruit drop, respectively at 70 g/100 l water (Khamis *et al*., 2015).

In the present study, all estimated products induced the enzymatic activities of chitinase (CHT), peroxidase (PO) and polyphenol oxidase (PPO) as well as the accumulation of total phenolic content compared with untreated trees of Ewais and Montakhab El-Qanater cultivars respectively. Concerning the enzymatic activities of all assessed enzymes, they were measured at the highest values in both tested cultivars with the treatment of Bellis 38% WG, Score 25% EC and Tekto 50% SC, respectively. While the lowest values were recorded after the treatment with Craft 30% EC and Dovex 50% SC, respectively. However, the highest values of total phenolic content in the two tested mango cultivars were remarked with Score 25% EC, Bellis 38%WG and Tekto 50%SC, respectively. While the lowest amounts were recorded with Craft 30% EC and Dovex 50% SC, respectively. These results are in the same trend with those recorded on mango, grape, avocado and date palm. Graham and Sticklen (1993) reported that chitinase enzyme is produced by several plant varieties such as grape as a defense against fungi attack. The upregulation of chitinase activity could increase the degradation process of chitin content which is the main component in the cell wall of fungal cells (Horn, 1977). The preceding study documented that several plants contain comparatively low levels of the enzyme chitinase and once produce higher levels of the enzyme when they are subjected to attack. Fungal diseases, mechanical damage and chewing insects elicits release of a variety of plant hormones which stimulate production of the enzyme chitinase (Giannikis *et al* 1998). Additionally, triggering defense-related enzymes, benomyl and carbendazim treatment of rice plants prevented bacterial leaf blight (Gomathinayagam *et al*., 2008). Similarly, treating tomato plants with the endophytic bacterium *Pseudomonas fluorescens* strain (6328) enhanced their resistance to *Fusarium oxysporum* by triggering the genes that encode defense-related enzymes, such as chitinase, peroxidase, β-1,3-glucanase, and phenylalanine ammonialyase (M'Piga *et al*., 1997). According to Hassan (2015), the accumulation of total phenolic content upregulated in Zebda cultivar more than Ewais and Hindy cultivars either in healthy or naturally infected one and twigs inoculated with *L. theobromae* and *F. moniliforme*. While the reduction of total phenols was associated with increased susceptibility of mango twigs to the dieback disease. The highest total phenols and free phenols were detected in the least

susceptible cultivar of Zebda. Regarding the enzymatic activities, polyphenol oxidase and peroxidase enzymes upregulated by using lemongrass (*Cymbopogon citratus*), rosemary (*Rosmarinus officinalis* L), kombucha filtrate, biocides Bio Arc (Local), (*Bacillus megaterium*) and Bio Zeid (Local) (*Trichoderma album*) and the two synthetic fungicides of Occidor and Antracol in Zebda, Ewais and Hindy cultivars compared to untreated cultivars (Hassan 2015).

In addition, peroxidase and polyphenol oxidase activities upregulated in two cultivars of avocado; Hass and Fuerte after dipping the scions in Amistar Top and or Bellis. However, after dipping the scions in Star copper or Bio Zeid, the lowest values were noted (Monir *et al*., 2021). Other studies were performed on some cultivars of date palm trees treated with fungicides; Uniform and/or Tshgarin documented that the highest values of total phenol in Barhey cv. while the lowest value was recorded in Zaghloul cv. Polyphenol oxidase activity has also upregulated in Barhey cv. with the treatment of Tshgarin while the lowest levels were obtained by using bio fungicide Serenade ASO. (Shehata and Hassan 2023). Hassan *et al*., (2023) found that, the efficacy of some agrochemicals *i.e*., salicylic acid, lemongrass oil, thyme oil, Imazalil (fungicide) and the biocontrol agent such as Biocontrol T34 (*Trichoderma asperellum* strain T34) against fruit rot of pear and their effect on fruit quality. All treatments kept fruit quality parameters and revealed significant effect considering total soluble solids, total phenolic content, titratable acidity, fruit firmness, polyphenol oxidase activity and peroxidase activity. Moreover, all investigated treatments were more effective against *L. theobromae* causing pear fruit rot disease.

CONCLUSIONS

In this study, the obtained data of the survey conducted in the orchards of mango during 2022 and 2023 revealed that the early fruit drop was common in the surveyed governorates Qalioubiya and Giza. Also, fungal isolates (*F. moniliforme*, *L. theobromae* and *A. alternata*) belonging to different genera were associated with early fruit drop disease of mango. *F. moniliforme* and *L. theobromae* caused the highest percentage of frequency in the two governorates. While the lowest frequency was caused by *A. alternata*. Typical symptoms of fruit drops were observed in all examined mango fruits. Montakhab El-Qanater cultivar showed the highest percentage of fruit drop. Meanwhile, Ewais showed the lowest percentage of drop. Score 25% EC 0.5ml/L, Bellis 38%WG 0.5g/L and Tekto 50%SC 1ml/L completely inhibited the growth of all the isolated fungi. Under filed conditions Score gave the highest efficacy for controlling fruit drop of mango during the two seasons on both tested cultivars, followed by Bellis and Tekto. While Craft and Dovex gave the lowest efficacy. There are positive relationships between enzymatic activities of chitinase (CHT), peroxidase (PO) and polyphenol oxidase (PPO) as well as total phenolic content and resistance developed in cv. Ewais and cv. Montakhab El-Qanater after applications of the tested agrochemical products in comparison to control.

Acknowledgement

The authors wish to express their sincere thanks, deepest gratitude and appreciation to all colleagues' and staff members in Plant Pathology Research Institute, El-Qanater El-Khayria Horticultural Research Station, (ARC) and Faculty of Agriculture-Ain Shams University, for their cooperation, efforts and facilities which made this investigation possible.

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Egyptian Journal of Phytopathology, Print ISSN:1110-0230&Online ISSN:2090-2522