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Investigating the reaction of some commercial potato cultivars and fungicides on late blight disease control

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ABSTRACT

Late blight (LB) caused by Phytophthora infestans (Mont.) de Bary, is one of the most destructive diseases of potato and tomato worldwide. Globally, application of fungicide is an effective approach for potato LB management. Isolates of P. infestans were collected from different fields in Egypt and characterized using microsatellite markers and mitcondrial DNA. Twelve fungicides were applied in vitro to test their control P. infestans isolates. Nine potato cultivars were evaluated under artificial inoculation. Field trials were conducted to evaluate the efficacy of applied fungicides to LB management and potato productivity at El-Behera governorate in two seasons. Our results revealed that collected isolates of P. infestans were A1 mating type, 23_A1 clonal lineage and Ia/IR1 mitcondrial DNA. Furthermore, potato cultivar Burren was highly resistant and recorded the lowest area under disease progress curve (AUDPC). On the other hand, cultivars Alaska, Spunta, Lady Rosita and Mondial were highly susceptible and revealed the highest AUDPC values. In vitro, all fungicide applications inhibited linear growth of *P. infestans* isolates (EG-Pi1 and EG-Pi2). For field trial, Equation-Pro, and Consonto were the most effective fungicides in respect of LB management where lowest disease severity and AUDPC were recorded in both prophylactically and curatively during two seasons. Moreover, potato tuber production significantly increased with Equation - Pro application. The results of this study provided crucial information for potato breeding programs and crop improvement as well as successful management strategy for late blight challenge.

Keywords: Phytophthora infestans, Late blight, Potato, Cultivars, Fungicides

INTRODUCTION

Globally, the potato is the third-largest food crop after wheat and rice, representing high nutritional value in a balanced diet. The potato crop plays a crucial role in stabilizing global food security and serves as a cash crop for smallholder farmers (Olanya *et al.*, 2012). The global harvested area of potatoes is 17.8 million hectares with a production of 374.8 million metric tons. In Egypt, the harvested area and productivity of the potato crop are 213.3 thousand hectares and 5.2 million metric tons, respectively (FAOSTAT, 2022). In some seasons, national potato production has been low due to various factors such as poor quality of seed tubers, inadequate agricultural inputs, population dynamics of plant pathogens, and ineffective management strategies for potato pests.

Potato late blight, caused by the phytopathogenic oomycete *Phytophthora infestans* (Mont.) de Bary, is one of the most economically costly plant pathogens affecting potato and tomato crops, with yield losses ranging from 16 to 100% worldwide (Namanda *et al.*, 2004; Haverkort *et al.*, 2009). In the last two decades, new aggressive clonal lineages of *P. infestans* have emerged, exacerbating disease management (Cooke *et al.*, 2012; Hu *et al.*, 2012; Danies *et al.*, 2013; Arafa *et al.*, 2018 and 2020; El-Ganiany *et al.*, 2022). Epidemics of late blight initiate with internal and external primary inoculum (Mizubuti and Forbes, 2002). Internal inoculum sources include blighted potato tubers and infected volunteer plants within and around potato fields, while external inoculum includes infected debris from previous seasons, which plays a substantial role in the epidemiology of *P. infestans* in temperate, tropical, and subtropical regions (Mizubuti and Forbes, 2002).

Exploiting more resistant potato varieties is one of the most effective strategies to control late blight and increase potato productivity as well as prevent harming the environment (Tähtjärv *et al.*, 2013) Management of late blight requires aggressive measures that include the combined use of agricultural practices, scouting, sanitation, and most importantly the combination of host plant resistance with application of fungicides (Kirk *et al.*, 2005). The use of protectant and systemic fungicides for managing late blight has perhaps been the most studied aspect of this disease management in temperate countries (Olanya *et al.*, 2001). Protective fungicides principally inhibit spore germination and penetration. However, once the pathogen enters the leaves, these fungicides become ineffective. Therefore, the development of new oomycete-targeted fungicides to control outbreaks of *P. infestans* is indispensable. Several new chemicals with varying modes of action have been developed (Gisi and Sierotzki, 2008; Gisi *et al.*, 2012; Sauter, 2012; Toquin *et al.*, 2012). Moreover, active ingredients like mefenoxam, azoxystrobin, cymoxanil, cyazofamid, and mandipropamid are site-specific fungicides that pose a greater risk for resistance development and selecting resistant fungal isolates (Gisi *et al.*, 2011; FRAC, 2014).

The application of fungicide dramatically reduced the progress of late blight, with a corresponding increase in potato tuber yield (Khan *et al.*, 2003). Chemical fungicides are generally categorized as systemic/translaminar and protectant in nature. Indeed, mancozeb and metalaxyl are considered the oldest and most applied active ingredients by potato growers worldwide. Unfortunately, many farmers and agricultural investors use these fungicides at higher frequencies and rates in a season than recommended (Sharma and KC, 2004). For instance, growers in Nepal apply fungicides 10–15 times during the same season to manage late blight disease, based on agro-weather station data and the resistance/susceptibility of potato cultivars (Sharma *et al.*, 2011).

The overdose of chemical fungicides not only raises the cost of productivity but also has trade-offs for the environment and people's health. In Egypt, several potato cultivars exhibit different levels of resistance and/or tolerance to late blight under various environmental conditions. However, the resistance of these potato cultivars can be broken down in some epidemic seasons based on the introduction of new races of the pathogen. Additionally, the declined efficacy of commonly applied fungicides has been informed by farmers, research staff, and agricultural extension employees. These field observations and agricultural reports proposed the possibility of fungicide-resistant strains in different countries all over the world (Matson *et al.*, 2015). Therefore, the periodic evaluation of alternative active ingredients to make effective, consistent, and successful recommendations for late blight control is indispensable. Furthermore, fungicide resistance development of new races by late blight pathogen has been reported, it is therefore highly recommended fungicides at appropriate doses at field conditions. So, before the fungicide's treatment at the field trial, there is a great necessity to evaluate them first *in vitro* against aggressive isolates of *P. infestans* under different concentrations. The objectives of this study were to (i) identify genetic resources of potatoes associated with late blight resistance/tolerance; (ii) determine fungicide resistance in Egyptian isolates of *P. infestans in vitro*; (iii) evaluate the efficacy of fungicides for managing late blight in the Egyptian market.

MATERIALS AND METHODS

Plant material:

Nine potato cultivars *i.e.* Mondial, Alaska, Burren, Spunta, Diamont, Hermes, Cara, Draga and Lady Rossetta were obtained from Horticulture Research Institute, Agricultural Research Center (ARC), Dokki, Egypt. These cultivars were used in the current research under greenhouse and field conditions.

Samples collection of Phytophthora infestans:

Samples of late blight disease were obtained from naturally infected potato plants. These samples were collected from Kafrelshiekh, Garbyia, and El-Beheira governorates during growing season 2022. The infected fresh leaves were transferred from the open field to the laboratory in transparent polyethylene bags under cold conditions. The samples were washed with tap water to remove dust and incubated in a humid chamber at 18 °C for three days in darkness to the enhancement of sporulation.

Isolation of *P. infestans*:

This trial was conducted at the Vegetable Diseases Research Department, Plant Pathology Research Institute, ARC in Giza, Egypt. In regarding the potato leaves, and stems, once the samples showed signs of sporulation, the lesions were cut into small pieces, which including both infected and healthy parts. Afterward, these tissue pieces were placed under organic potato slices of the highly susceptible cultivar "Lady Rosetta" in sterilized Petri dishes that were converted and not turned over. The Petri dishes contained water agar (20g agar and distilled water up to 1 liter) to increase the moisture. The dishes were then incubated at 18°C for 7-10 days in the dark, depending on the isolate, until abundant sporulation appeared on the upper side of the slices. Eventually, sporangia were picked up from the top of the slices and transferred directly onto the recommended media. Rye agar media (Caten and Jinks, 1968) (60 g of organic rye grains, 20 g sucrose, and 20 g agar per liter) and pea agar (Jaime-Garcia *et al.*, 2000), (120 g of organic frozen peas and 20 g agar per liter) were used for isolation, growth and maintenance of *P. infestans* isolates. The mix of antibiotics *i.e.* Ampicillin 200 mg, Rifampicin 20 mg (Sigma-Aldrich), Nystatin 50 mg (Sigma-Aldrich), Pentachloronitrobenzene (PCNB) 50 mg and Benlate (50%WP) 50 mg per liter media were used. The developed colonies were purified and identified using the morphological and microscopic characteristics according to Martin *et al.* (2012). The purified cultures were reserved on rye slants at 18°C for further studies.

Mating type assay:

To determine the mating types of *P. infestans*, each isolate was paired with reference isolates A1 and A2 on pea agar media. The individual test isolates were placed 4 cm apart from each other on Petri dishes and incubated in darkness at 18 °C for around two weeks. After incubation, the presence or absence of oospores at the contact interface between the paired isolates was observed. Based on the presence of the oospores, the paired isolates were named as A1 or A2 accordingly. This method was explained by Cooke *et al.* (2003).

Molecular characterization of P. infestans isolates:

Total genomic DNAs of collected P. infestans isolates were extracted using the GeneJET Plant Genomic DNA Purification Mini Kit (Thermo Fisher Scientific). Quantity and quality of the extracted genomic DNAs were performed by electrophorese in 1% agarose gel and NanoDrop (Thermo Fisher Scientific Inc. Wilmington, DE, USA). Subsequently, the DNA samples were subjected to a 12-plex PCR using 12 simple sequence repeat (SSR) markers (D13, G11, Pi4B, Pi04, Pi63, Pi70, PinfSSR2, PinfSSR3 (also referred to as Pi02), PinfSSR4, PinfSSR6, PinfSSR8, and PinfSSR11) (Table 1) as previously described (Li et al. 2013). Each microsatellite marker was labeled with one of the four fluorescent dyes FAM, VIC, NED, or PET, depending on the expected size of each SSR product to avoid any overlapping allele size ranges. PCR reactions were prepared by multiplexing all the 24 primers in one PCR tube for each DNA sample using the QIAGEN Multiplex PCR Kit (Qiagen, Hilden, Germany). Each single 12.5 μ L PCR comprised 1 μ L of the DNA extract, 6.25 of 2× Qiagen Type-IT Multiplex PCR mix, 0.44 μ L of primer master mix and 4.81 µL of nuclease-free water. Amplification was conducted in a thermocycler (MWG-Biotech, Ebersberg, Germany), with initial denaturation at 95 °C for 5 min, then by 28 cycles of 95 °C for 30 s, 58 °C for 90 s, and 72 °C for 20 s, and a final extension at 60 °C for 30 min. Furthermore, the mitochondrial DNA of P. infestans isolates was amplified from the extracted genomic DNA using PCR with six primer pairs, P1, P2, P3, and P4 (Griffith and Shaw 1998) as well as HVRi and HVRii (Yang et al. 2013) (Table 1). The thermal cycling conditions were based on Griffith and Shaw (1998) and Yang et al. (2013). The amplicon DNA were electrophoresised on 2% agarose gels and visualized using UV gel documentation (Biocraft, Co. Japan) after staining with ethidium bromide.

Primer name	Forward primer sequence	Reverse primer sequence	Restriction enzyme	Reference
	·	SSR markers		
SSR3	ACTTGCAGAACTACCGCCC	GTTTGACCACTTTCCTCGGTTC	NA	Li <i>et al</i> . (2010)
Pi4B	AAAATAAAGCCTTTGGTTCA	GCAAGCGAGGTTTGTAGATT	NA	Knapova and Gisi (2002)
PiG11	TGCTATTTATCAAGCGTGGG	GTTTCAATCTGCAGCCGTAAGA	NA	Knapova and Gisi (2002)
Pi04	AGCGGCTTACCGATGG	GTTTCAGCGGCTGTTTCGAC	NA	Lees et al. (2006)
Pi63	ATGACGAAGATGAAAGTGAG G	CGTATTTTCCTGTTTATCTAACACC	NA	Lees et al. (2006)
Pi70	ATGAAAATACGTCAATGCTCG	CGTTGGATATTTCTATTTCTTCG	NA	Lees et al. (2006)
PinfSSR11	TTAAGCCACGACATGAGCTG	GTTTAGACAATTGTTTTGTGGTCGC	NA	Li et al. (2010)
PinfSSR2	CGACTTCTACATCAACCGGC	GTTTGCTTGGACTGCGTCTTTAGC	NA	Li et al. (2010)
PinfSSR4	TCTTGTTCGAGTATGCGACG	GTTTCACTTCGGGAGAAAGGCTTC	NA	Li et al. (2010)
PinfSSR6	GTTTTGGTGGGGCTGAAGTTT T	TCGCCACAAGATTTATTCCG	NA	Li et al. (2010)
PinfSSR8	AATCTGATCGCAACTGAGGG	GTTTACAAGATACACACGTCGCTCC	NA	Li et al. (2010)
D13	TGCCCCCTGCTCACTC	GCTCGAATTCATTTTACAGACTTG	NA	Li et al. (2010)
		Mitochondrial DNA haplotype		
P1	GCAATGGGTAAATCGGCTCA A	AAACCATAAGGACCACACAT	Cfol	Griffith and Shaw 1998
P2	TTCCCTTTGTCCTCTACCGAT	TTACGGCGGTTTAGCACATACA	Mspl	Griffith and Shaw 1998
Р3	ATGGTAGAGCGTGGGAATCA T	AATACCGCCTTTGGGTCCATT	EcoRI	Griffith and Shaw 1998
P4	TGGTCATCCAGAGGTTTATGT T	CCGATACCGATACCAGCACCAA	EcoRI	Griffith and Shaw 1998
HVRi	GTGGGTTCGAGTCCCACTAA	CTCACCCGTTCGCTATGTTT	NA	Yang et al. 2013
HVRii	ACGGAATTATCGGAAGATTT	AAATCCCTTTTATACTGTAATTTGA T	NA	Yang et al. 2013

Table 1. Oligonucleotide sequences used in this study.

Inoculum preparation:

Inoculum preparation of EG_Pi2 isolate was performed using susceptible potato cultivar leaves in six weeks-old plants. A sporangial suspension was obtained from 20 old-day rye agar Petri dishes at 18°C in the dark, where three ml sterilized distilled water were added to each Petri dish which contained pure culture of *P. infestans* and rubbed off the sporangia to make a suspension. Potato leaflets were placed onto moistened filter paper with sterilized distilled water in 140 mm sterilized Petri plates on the abaxial surface. The leaflets were injured at the center using a sterile 10 μ l micropipette tip and inoculated with a 30 μ l sporangial suspension. The Petri plates were incubated for 48 hrs at 18°C in darkness, then at 18°C with a 12-h photoperiod for 7 days where the mycelial covers all potato leaflets surface. Then, the sporulated leaves were gently shaken in sterilized distilled water to dislodge sporangia and produce sporangial suspension was adjusted via hemocytometer, which was adjusted to 15×10⁴ sporangia/ml. The suspension was chilled at 4 °C for 2-4 hours before artificial inoculation to cleave sporangia and release zoospores.

The vulnerability of commercial potato cultivars to the potato late blight in vivo:

Nine commercial potato cultivars *i.e.* Mondial, Alaska, Burren, Spunta, Diamont, Hermes, Cara, Draga, Lady Rosetta were evaluated under greenhouse conditions during season 2022 using isolate EG_Pi2 of *P. infestans.* Plastic pots (50 cm in diameter) were filled with a mixture of sand and clay (1:1, v/v), and a single potato seed tuber was sown in each pot. Three plastic pots were employed as replicates for each cultivar. Furthermore, all plants were irrigated and fertilized based on the recommendation of The Ministry of Agriculture and Land Reclamation. After 50 days of the planting date, all tested plants were sprayed with *P. infestans* inoculum at a concentration of 15×10^4 of sporangia per ml. Late blight was assessed four times (three, seven, eleven, and fifteen) days post inoculation (DPI) where the final score of 15 DPI was analyzed. Disease severity of late blight was assessed according to Danesh *et al.* (1994) with a scale of 0 to 5 where 0.5 increments, as 0 = no disease symptoms, 0.5 = Less than 10% bilghted leaf area, 1 = 10-20% bilghted leaf area, 1.5 = 21-30% bilghted leaf area, 2 = 31-40% bilghted leaf area, 4 = 71-80% bilghted leaf area, 4.5 = 81-90% bilghted leaf area, and 5 = 91-100% bilghted leaf area.

Interestingly, tested potato cultivars were grouped into five categories as follows: highly resistant (HR): less than 10% of leaf area; resistance (R): 10 -15%; moderately resistant (MR): 16 - 25%; susceptible (S): 26 -50% and highly susceptible (HS): more than 50% as described by Duan *et al.* (2021) with some modifications:

Assessment of potato cultivars response to late blight under field conditions:

A total of nine commercial potato cultivars which used in the previous trial were evaluated under field conditions for late blight at Etai-Elbarod, El-Beheira governorate during the growing seasons of 2022 and 2023. Randomized complete block design (RCBD) was utilized in the current trial with three plots (replicates) per cultivar. Each plot (6 x 7 m2) contained twelve rows. All plots were regularly watered and fertilized weekly with NPK 20-20-20 and micronutrients. Disease severity was recorded four times (10 days intervals) 40, 50, 60, and 70 days after planting. Disease severity % was calculated according to the equation described by Descalzo et al. (1990) as follow:

$R = \left[\sum (a \times b)/N \times K\right] \times 100$

where: R = disease severity, a = number of infected leaves rated, b = numerical value of each grade, N = total number of examined plants, K = the highest degree of infection in the scale.

Area under disease progress curve (AUDPC) was estimated based on the obtained data to compare different responses of the tested cultivars according to Pandey *et al.* (1989) using the following equation:

 $AUDPC = D [1/2 (Y_1 + Y_K) + Y_2 + Y_3 + \dots + Y_{(K-1)}]$

Where:

D = days between each successive two readings, Y_1 = first disease record. Y_k = last disease record.

Efficacy of fungicides against *P. infestans*:

Chemical fungicides:

Twelve commercial fungicides with various active ingredients and mobility in the plant named Saver (Cymoxanil + Chlorothalonil); Consonto (Fenamidone+ Propamocarb–HCL); Infinito (Propamocarb-HCL+ Fluopicolide); Amesto (Azoxystrobin); Reval (Propamocarb – HCL); Dithane M 45 (Mancozeb);

Folio Gold (Chlorothalonil + Metalaxyl-M); Facomel MZ (Mancozeb+ Metalaxyl); Equation – Pro (Cymoxanil + Famoxadone); Cymonel (Cymoxanil); New copper (Copper oxychloride) and Fostyl (Fosetyl- aluminium) (Table 2) were applied *in vitro* to test the baseline sensitivity of Egyptian *P. infestans* isolates. Additionally, the obtained fungicides were applied on open field trials to determine their efficacy against late blight disease.

In vitro trial:

Stock solutions of the tested fungicides were prepared by dissolving them in sterile distilled water. Two *P. infestans* isolates (EG_Pi1 and EG_Pi2) were grown on rye agar medium as mentioned above for two weeks at 18°C. Fungicides were added into the recommended media (sterilized and molten pea media, 50 °C) via dilution of stock solutions to a recommended dose of each fungicide (Table 2) where the agar was mixed thoroughly before pouring into plates (15ml/plate) and allowed to solidify. A single agar plug (5mm) was removed from actively growing mycelia and placed on the center of each Petri plate. Three plates were used for each isolate and repeated twice. Three Perti dishes were used as a control (non-amended fungicides) for each isolate. All inoculated plates were placed in plastic boxes for incubation, and were kept at 18°C in the darkness for two weeks. Two perpendicular measurements were made of each colony and the average colony diameter was calculated. Moreover, linear growth and inhibition percentage over control were estimated for each isolate along with all treatments as follows

Growth inhibition = $\frac{\text{Growth in control plates} - \text{Growth in treatment plates}}{x \ 100}$

Growth in control plates

Efficacy of fungicides against potato late blight in open field:

The twelve fungicides assigned for late blight control were evaluated under natural conditions in hot spot regions along with two approaches prophylactical and curative (Table 2). This work was conducted in Etai-Elbarod, El-Beheira governorate during two successive growing summer seasons 2022 and 2023. A randomized complete block design (RCBD) with 3 replications was adopted in this respect. Certified potato tuber seeds of cv. Spunta were cut into single-eye seed pieces. Seed pieces were healed at room temperature for 48 h. The experimental unit was a plot measured (6×7 m² dimensions)) included 12 rows with 7 m long and 20 cm apart. A total of 39 (3x13) experimental plots were established. All cultural practices were applied according to the technical recommendation of the crop as normal. Interestingly, two independent application approaches of treatments (prophylactically and curatively) were run for each fungicide. The first symptom was observed after 37 days from the planting date on, 5th and 10th January 2022 and 2023, respectively. The treatments were applied at 10 days intervals with the recommended dose (Table 2). The disease severity was scored as previously described. Evaluation of the efficacy of each fungicide was computed according to the following formula adopted by Rewal and Jhooty (1985). % infection in the control - % infection in treatment

Efficacy (%) =

% Infection in the control

Table 2. Detailes of twelve commercial fungicides used in the current research for potato late blight management.

No.	Commercial name	Active ingredient	FRAC code	***Concentration/ Formulation type	Mobility in the plant	Recommended dose/100L				
1	Saver	Cymoxanil + Chlorothalonil	*27+ **M 05	42.5% SC	Systemic + Contact	300 cm ³				
2	Consonto	Fenamidone+ Propamocarb – HCL	11+28	45% SC	Systemic + Systemic	250 cm ³				
3	Infinito	Propamocarb-HCL+ Fluopicolide	28+43	68.75 % SC	Systemic+ Translaminar	100 cm ³				
4	Amesto	Azoxystrobin	11	25% SC	Systemic	50 cm ³				
5	Reval	Propamocarb - HCL	28	72.0% SL	systemic	250 cm ³				
6	Dithane M 45	Mancozeb	M 03	80.00 % WP	Contact	250 g				
7	Folio Gold	Chlorothalonil + Metalaxyl-M (Mefenoxam)	M 05 +4	53.75% SC	Systemic+ Systemic	300 cm ³				
8	Facomel MZ	Mancozeb+ Metalaxyl	M 03+4	72.00 % SP	Contact+ Systemic	250 g				
9	Equation – Pro	Cymoxanil + Famoxadone	27+11	52.5% WG	Systemic+ Systemic	40 g				
10	Cymonel	Cymoxanil	27	80 % WP	Systemic	50 g				
11	New copper	Copper oxychloride	M 01	50% WP	Contact	250 g				
12	Fostyl	Fosetyl- Al	P07	80 % WP	Systemic	250 g				

FRAC= Fungicide Resistance Action Committee.

* The numbers were assigned primarily according to the time of product introduction to the market.

** Different letters (A to P, with added numbers) are used to distinguish fungicide. groups according to their biochemical mode of action in the biosynthetic pathways of plant pathogens.

*** Percentage of active ingredient in commercial product.

M = chemical multi-site inhibitors.

P = host plant defense inducers.

SC= Suspension concentrate; SL= Soluble liquid; WP= Wettable powder; SP= Water soluble powder; WG= wettable granule. Estimation of yield:

Yield of potato weight of tubers (kg) per net plot for all treatments (twelve fungicides) and control was weighted after harvesting tubers at 140 days. The percentage of yield increase was calculated using the following equation:

Rate of increase (%) = (Yield of treatment- Yield of control/ Yield of control) x100

Statistical analysis:

The data collected was analyzed using Analysis of Variance (ANOVA) based on a randomized complete block design (RCBD). It is worth noting that the dataset analysis was performed with the help of the WASP statistical software. Mean differences were compared using the Least Significant Difference (LSD) method, as suggested by Hoshmand (2006).

RESULTS

Samples collection and characterization of *P. infestans*:

A total of twelve isolates of *P. infestans* were collected during the 2021/2022 growing season from three governorates: Kafrelsheikh, Garbyia, and EL-Behera (Table 3). The majority of the isolates (10) were obtained from Kafrelsheikh governorate. The isolates were collected from tomato crops (leaves, stems, and fruits) and potato crops (leaves). Eight of the *P. infestans* isolates were collected during the 2022 growing season, while the remaining four were taken during the second season (2022). Interestingly, all twelve *P. infestans* isolates belonged to the A1 mating type, as determined by an *in vitro* assay on organic rye grains medium, including two reference isolates (A1 and A2 mating type). On the other hand, A2 mating type and self-fertile isolation were not detected in this study. Twelve SSR markers were used to identify the collected Egyptian *P. infestans* isolates as a 23_A1 clonal lineage (as shown in Table 1). Furthermore, 12 isolates of *P. infestans* were analyzed using four restriction fragment length polymorphism (RFLP) markers to detect sequence variations in four regions of the mitochondrial genome (P1, P2, P3, and P4). The mtDNA polymorphisms grouped the tested isolates into group la. Moreover, hypervariable regions of two other isolates were analyzed, and only one type, IR1 type, was detected.

c	Isolato		Diant	Sampli	location		*Matin **mtDN		***Clonal
s. No.	name	Host	tissue	ng year	District	Governorate	g type	haplotyp e	lineage
1	EG_Pi1	Tomato	Stem	2022	Qallen	Kafrelsheikh	A1	la/IR1	23_A1
2	EG_Pi2	Potato	Leaves	2022	Kafrelzyat	Garbia	A1	la/IR1	23_A1
3	EG_Pi3	Tomato	Leaves	2022	Qallen	Kafrelsheikh	A1	la/IR1	23_A1
4	EG_Pi4	Tomato	Stem	2022	Qallen	Kafrelsheikh	A1	la/IR1	23_A1
5	EG_Pi5	Tomato	Leaves	2022	Qallen	Kafrelsheikh	A1	la/IR1	23_A1
6	EG_Pi6	Tomato	Fruits	2022	Qallen	Kafrelsheikh	A1	la/IR1	23_A1
7	EG_Pi7	Tomato	Stem	2022	Baltem	Kafrelsheikh	A1	la/IR1	23_A1
8	EG_Pi8	Tomato	Leaves	2022	Baltem	Kafrelsheikh	A1	la/IR1	23_A1
9	EG_Pi9	Tomato	Fruits	2022	Baltem	Kafrelsheikh	A1	la/IR1	23_A1
10	EG_Pi10	Potato	Leaves	2022	Rahmanyia	EL-Behera	A1	la/IR1	23_A1
11	EG_Pi11	Tomato	Fruits	2022	Sakha	Kafrelsheikh	A1	la/IR1	23_A1
12	EG_Pi12	Tomato	Stem	2022	Sakha	Kafrelsheikh	A1	la/IR1	23_A1

Table 3. Details of *Phytophthora infestans* isolates used in this study.

EG=Egypt

Pi= Phytophthora infestans

*Two reference isolates (A1 and A2 mating type) were used in this assay

**Six mitochondrial regions on the mitochondrial genome were amplified using six markers (Griffith and Shaw 1998; Yang *et al.*, 2013)

***twelve SSR markers were utilized for clonal lineage detection

Assessment of potato cultivars to late blight resistance under greenhouse conditions:

Nine different cultivars of potato were sprayed with inoculum of *P. infestans* isolates EG_1 (23-A1) during the night. The first symptoms of late blight were observed four days after inoculation under favorable conditions. After 15 days, the severity of the disease on each cultivar was scored and the data was presented in Table 4. The results showed that isolate EG_1 was virulent on all tested cultivars, but the severity of the disease varied, with a range from 8.7% to 59.6%. Among the tested cultivars, Burren showed the highest level of late blight resistance with a severity of only 8.7%,

followed by Diamont and Hermes with severity of 11.3% and 11.7%, respectively. Additionally, the area under the disease progress curve (AUDPC) was 41.0, 63.8 and 51 for Burren, Diamont, and Hermes, respectively (Fig. 1). On the other hand, the cultivar Mondial was found to be the most susceptible to *P. infestans* with a severity of 59.6%, followed by Lady Rosetta and Spunta with severity of 52.6% and 51.3%, respectively. The AUDPC values for these cultivars were 323.2, 276.4, and 273, respectively (Fig. 1). Potato cultivar that may have general partial resistance to virulent isolate. Furthermore, the tested cultivers were divided into five reactions (HR, R, MR, S and HS). Burren cultiver recorded the highest reaction (HR), whereas, Mondial and Leady Rossrta revealed the lowest reaction (HS).

Cultivar	3 DPI	7 DPI	11 DPI	15 DPI	Reaction*
Mondial	0	12.7	38.3	59.6 a	HS
Alaska	0	5.3	23.3	41.2 d	S
Burren	0	1.2	4.7	8.7 h	HR
Spunta	0	10.3	32.3	51.3 c	HS
Diamont	0	3.8	6.5	11.3 g	R
Hermes	0	2.5	4.4	11.7 g	R
Cara	0	6.6	12.6	25.0 e	MR
Draga	0	6.5	10.3	23.5 f	MR
Lady Rosetta	0	9.4	33.4	52.6 b	HS
L.S.D at 0.05				1.298	

Table 4. Evaluation of nine commercial potato cultivars for *P. infestans* resistance under greenhouse conditions.

Values in the same column followed by the same letter are not significantly differed at P < 0.05 level.

*Cultivar reaction taken after 70 days from planting date, where HS = Highly susceptible, S = Susceptible, MR = Moderately resistant, R = Resistant and HR = Highly resistant.

DPI= Days post inoculation.



Fig. 1. Area under disease progress curve (AUDPC) of nine potato cultivars under greenhouse conditions.

Assessment of potato cultivars to late blight resistance under field conditions:

During the growing season of 2022, nine commercial potato varieties were evaluated in an open field at EL-Behera governorate under natural infection.

The data presented in Table (5) and Fig. (2) showed that the potato cultivars had the same reaction trend under greenhouse conditions. The Burren cultivar was highly resistant and recorded the lowest disease severity (Fig. 3) and area under disease progress curve at 10.0% and 107, respectively, compared to other cultivars in the study.

Diamont and Hermes potato cultivars displayed moderate resistance reaction at 70 days post-planting, with disease severity at 19.3% and 21.4%, respectively, as well as 363 and 426.5 AUDPC, respectively. Conversely, Mondial, Lady Rosetta, Spunta, and Alaska potato cultivars showed a highly susceptible response with a disease severity of 68.7%, 62.3%, 60.7%, and 55.9%, respectively, and the highest area under disease progress curve of 1002.0, 936, 931.5, and 717.0, respectively. These findings suggest that the potato cultivars have a general partially resistant response to the virulent isolate.

Table 5. Evaluation of nine potato cultivars for late blight disease under natural infection at EL-Beheragovernorate during two growing seasons 2022/2023 and 2023/2024*

Cultivars		Decetion***			
	40 days**	50 days	60 days	70 days	Reaction
Mondial	10.9	20.7	39.7	68.7 a	HS
Alaska	5.3	10.8	30.3	55.9 d	HS
Burren	0	0.0	5.7	10.0 i	HR
Spunta	10.4	20.3	37.3	60.7 c	HS
Diamont	4.7	9.4	14.9	19.3 h	MR
Hermes	5.3	11.6	17.7	21.4 g	MR
Cara	8.5	21.4	28.9	47.8 f	S
Draga	8.3	23.6	29.3	49.3 e	S
Lady Rosetta	9.3	20.1	37.7	62.3 b	HS
L.S.D at 0.05				1.368	

Values in the same column followed by the same letter are not significantly differed at P < 0.05 level.

* Data presented in the Table referee to mean of two seasons 2022/2023 and 2023/2024.

** Number refer to days' post planting date

***Cultivar reaction taken after 70 days, where HS = Highly susceptible, S = Susceptible, MR = Moderately resistant, R = Resistant and HR = Highly resistant.



Fig. 2. Area under disease progress curve (AUDPC) of nine potato cultivars under open field conditions during two growing seasons 2022/2023 and 2023/2024.



Fig. 3. Disease severity percentage at six cultivars A= Burren B= Diamont, C= Hermes, D= Lady Roseta, E= Mondial and F= Spunta.

Efficacy of fungicides against *P. infestans*:

Laboratory test:

Twelve fungicides were evaluated for their effectiveness against *P. infestans* isolates (EG-Pi1 and EG-Pi2) *in vitro*, as shown in Table 6 and Fig. 4 & 5. Most of the fungicides tested achieved complete inhibition (100%) of *P. infestans* with both isolates (tomato and potato). Consonto, Infinito, Amesto, Dithane M 45, Folio Gold, Facomel MZ, Equation-Pro, Cymonil, New Copper, and Fostyl completely stopped the growth of *P. infestans* isolates. However, Saver and Reval recorded linear growth of 2.4 and 2.2 cm, respectively, with the tomato isolate, and 0.5 and 1.78 cm, respectively, with the potato isolate, compared to the control treatment. The data also showed that Saver and Reval achieved inhibition of 73.3% and 75.6%, respectively, with EG-1, and 94.4% and 80.0%, respectively, with EG-2.

	Pure statute	EG-1	***	EG-2****		
5. NO.	Fungicide	L.G.*(cm)	inhi.**%	L.G(cm)	inhi.%	
1	Saver	2.4 b	73.3	0.5 c	94.4	
2	Consonto	0.5 c	94.4	0.5 c	94.4	
3	Infinito	0.5 c	94.4	0.5 c	94.4	
4	Amesto	0.5 c	94.4	0.5 c	94.4	
5	Reval	2.2 b	75.6	1.8 b	80.0	
6	Dithane M 45	0.5 c	94.4	0.5 c	94.4	
7	Folio Gold	0.5 c	94.4	0.5 c	94.4	
8	Facomel MZ	0.5 c	94.4	0.5 c	94.4	
9	Equation – Pro	0.5 c	94.4	0.5 c	94.4	
10	Cymonel	0.5 c	94.4	0.5 c	94.4	
11	New copper	0.5 c	94.4	0.5 c	94.4	
12	Fostyl	0.5 c	94.4	0.5 c	94.4	
13	Control	9.0 a	0.0	9.0 a	0.0	

Table 6. Efficacy of twelve fungicides on the linear growth and inhibition percentage of mycelial growth of two *P* infestance isolates

L.G*= Linear growth; % inhi** = % Inhibition.

****Collected from potato crop.

Values in the same column followed by the same letter are not significantly differed at P < 0.05 level.

^{***}Collected from tomato crop.



Fig. 4. Efficacy of twelve fungicides on the linear growth of *P. infestans* isolate (EG-Pi1).1= Propan, 2= Consonto, 3= Infinito, 4= Amesto, 5= Reval, 6= Dithane M 45, 7= Folio Gold, 8= Facomel MZ, 9= Equation – Pro, 10= Cymonel, 11= New copper, 12=Fostyl and C= Control



Fig. 5. Efficacy of twelve fungicides on the linear growth of *P. infestans* isolate (EG-Pi2). 1= Propan, 2= Consonto, 3= Infinito, 4= Amesto, 5= Reval, 6= Dithane M 45, 7= Folio Gold, 8= Facomel MZ, 9= Equation – Pro, 10= Cymonel, 11= New copper, 12=Fostyl and C= Control

Field trial and fungicides application:

During the growing season of 2022/2023 twelve fungicides were used as a preventive treatment for late blight management in the Etai Elbarood district of El-Behera governorate. The results showed that all of the fungicides significantly reduced the symptoms of late blight disease in the Spunta potato cultivar. The severity of the disease ranged from 7.8% to 20.1% (Table 7). The best fungicides based on the results were Equation-Pro, Consonto, Propan, Infinito, Amesto, and Folio Gold, which had disease severity percentages of 7.8%, 8.0%, 9.4%, 9.7%, 10.0%, and 11.0%, respectively, compared to the control treatment (72.2%). These fungicides also recorded the highest efficacy percentages, ranging from 84.7% to 89.2%. On the other hand, Dithane M 45 and New copper had the highest disease severity percentages of 16.9% and 20.1%, respectively, and the lowest efficacy percentages of 72.2% and 76.6%, respectively (Table 7).

C No	Fungicides		Disease s			
3. NO.		Zero time	10 DPA	20 DPA	30 DPA	EITICACY %
1	Saver	0.0	3.3	7.1	9.40 g	86.9
2	Consonto	0.0	3.2	5.1	8.00 h	88.9
3	Infinito	0.0	3.7	6.8	9.70 g	86.6
4	Amesto	0.0	3.9	6.8	10.0 g	86.1
5	Reval	0.0	4.2	7.1	11.3 f	84.3
6	Dithane M 45	0.0	5.9	9.0	16.9 c	76.6
7	Folio Gold	0.0	4.8	7.3	11.0 f	84.7
8	Facomel MZ	0.0	4.1	8.7	12.9 e	82.1
9	Equation – Pro	0.0	3.0	5.3	7.80 h	89.2
10	Cymonel	0.0	4.1	8.0	14.2 d	80.3
11	Kabakeet (New copper)	0.0	7.3	13.4	20.1 b	72.2
12	Fostyl	0.0	4.3	7.3	11.4 f	84.2
13	Control	0.0	23.7	51.9	72.2 a	-
	L.S.D at 0.05				0.769	

Table 7. Efficacy of twelve fungicides as a prophylactically application for late blight control at Etai Elbarood, El-
Behera governorate during growing season 2022/2023.

DPA= Days post application

Values in the same column followed by the same letter are not significantly differed at P < 0.05 level.

The Table (8) presents data that demonstrates the effectiveness of twelve fungicides during the second growing season of 2023/2024. The severity of the disease ranged from 8.0% to 21.7% for the tested fungicides, while the control treatment had a severity of 73.8%. A similar trend was also observed during the first season of 2022/2023 for the same fungicides.

Table 8.	Efficacy of twelve fungicides as a prophylactically application for late blight control at Etai Elbarood, E	-
	Behera governorate during growing season 2023/2024.	

C N	Funciaida		Disease	Effica 0 /		
5. NO	Fungicide	Zero time	10 DPA	20 DPA	30 DPA	Efficacy %
1	Saver	0.0	4.4	7.8	9.9 fg	86.6
2	Consonto	0.0	4.2	6.4	8.2 h	88.9
3	Infinito	0.0	4.7	6.9	9.2 fg	87.5
4	Amesto	0.0	4.9	7.0	9.4 fg	87.2
5	Reval	0.0	4.7	7.2	10.3 g	86.0
6	Dithane M 45	0.0	6.9	10.3	17.8 c	75.9
7	Folio Gold	0.0	4.3	7.1	11.2 f	84.8
8	Facomel MZ	0.0	5.2	8.2	13.4 e	81.8
9	Equation – Pro	0.0	3.2	5.9	8.0 h	89.2
10	Cymonel	0.0	5.0	8.2	15.2 d	79.4
11	Kabakeet (New copper)	0.0	7.1	10.4	21.7 b	70.6
12	Fostyl	0.0	4.8	6.8	11.0 f	85.1
13	Control	0.0	24.6	52.7	73.8 a	0.0
	L.S.D at 0.05				0.892	

DPA= Days post application

Values in the same column followed by the same letter are not significantly differed at P < 0.05 level.

The results of AUDPC of twelve fungicides with prophylactic application at Etai Elbarood, El-Behera governorate during the 2022 and 2023 growing seasons in an open field were presented in Fig. 6. The results show that the tested fungicides can be grouped into two categories based on their AUDPC values. The first category includes fungicides with the lowest AUDPC values (less than 200), which are Equation – Pro (122.0 and 131.0), Consonto (123.0 and 147.0), and Infinito (153.5 and 162.0) during the two seasons. The second category includes fungicides that have high AUDPC values (more than 200), such as Dithane M 45 (233.5 and 261.0) and New copper (307.5 and 283.5) during the two seasons.



Fig. 6. Area under disease progress curve under of twelve fungicides with prophylactically application under natural infection during two growing seasons 2022 and 2023.

The Tables (9 and 10) show that using fungicides significantly reduced the severity of late blight disease when applied as a curative treatment, compared to leaving the plants untreated. Four fungicides - Equation-Pro, Amesto, Infenito, and Consonto - resulted in the lowest percentage of disease severity: 10.0%, 10.4%, 10.8%, and 11.0%, respectively, in the first season, and 10.4%, 11.0%, 11.4%, and 11.6%, respectively, in the second season. This is compared to untreated plots which recorded 73.8% and 76.7% disease severity, respectively. These four fungicides also had the highest efficacy rates: 86.4%, 85.9%, 85.3%, and 85.1%, respectively, in the first season, and 86.4%, 85.7%, 85.1%, and 84.8%, respectively, in the second season. On the other hand, the highest recorded disease severity of late blight was observed when using Dithane M 45 and New copper, with 29.3% and 25.7%, respectively, in 2022, and 29.8% and 26.8%, respectively, in 2023. These fungicides also had the lowest efficacy rates of 60.3% and 65.2%, respectively, in 2022, and 61.1% and 65.1%, respectively, in 2023.

			Disease severity %					
S. No.	Fungicide	Zero	10	20	30	Efficacy %		
		time	DPA*	DPA	DPA			
1	Saver	4.7	7.4	9.2	11.9 g	83.9		
2	Consonto	5.5	7.3	9.9	11.0 gh	85.1		
3	Infinito	5.8	7.4	8.9	10.8 h	85.3		
4	Amesto	6.4	8.2	9.1	10.4 h	85.9		
5	Reval	7.9	9.3	11.4	13.4 f	81.8		
6	Dithane M 45	6.3	9.4	14.7	29.3 b	60.3		
7	Folio Gold	4.9	7.1	9.4	11.3 g	84.7		
8	Facomel MZ	4.3	7.9	13.4	17.2 e	76.7		
9	Equation – Pro	5.1	7.0	8.8	10.0 h	86.4		
10	Cymonel	6.7	8.4	13.2	18.9 d	74.4		
11	Kabakeet (New copper)	6.3	8.5	14.8	25.7 c	65.2		
12	Fostyl	5.2	7.3	10.0	12.4 g	83.2		
13	Control	4.9	27.9	57.3	73.8 a	-		
	L.S.D at 0.05				0.876			

 Table 9. Efficacy of twelve fungicides as a curatively application for late blight control in open field condition at Etai Elbarood, El-Behera governorate during season 2022/2023.

DPA= Days post application

Values in the same column followed by the same letter are not significantly differed at P < 0.05 level

S. No.	Fungicide	Zero	10	20	30	Efficacy %
		Time	DPA*	DPA	DPA	
1	Saver	4.8	7.7	9.9	12.7 g	83.4
2	Consonto	5.8	7.6	9.6	11.6 h	84.8
3	Infinito	5.9	7.8	8.4	11.4 h	85.1
4	Amesto	6.8	8.4	9.8	11.0 h	85.7
5	Reval	7.7	9.7	11.7	13.9 f	81.9
6	Dithane M 45	6.7	9.7	14.9	29.8 b	61.1
7	Folio Gold	4.4	7.6	9.8	11.5 h	85.0
8	Facomel MZ	4.6	8.2	13.9	17.9 e	76.7
9	Equation – Pro	5.4	7.4	9.2	10.4 i	86.4
10	Cymonel	6.9	8.7	14.2	19.3 d	74.8
11	Kabakeet (New copper)	6.8	8.6	15.8	26.8 c	65.1
12	Fostyl	5.9	7.7	10.8	13.2 f	82.7
13	Control	4.8	28.4	59.3	76.7 a	0
	L.S.D _{at 0.05}				0.794	

Table 10. Efficacy of twelve fungicides as a curatively application for late blight control in open field condition	
at Etai Elbarood, El-Behera governorate during season 2023/2024.	

DPA= Days post application

Values in the same column followed by the same letter are not significantly differed at P < 0.05 level.

Data presented in Fig (7) demonstrated the area under disease progress curve (AUDPC) of twelve fungicides as a curative application in open fields during two seasons 2022 and 2023. AUDPC values of fungicides Equation – Pro, Infinito, Folio Gold, Consonto, Propan, Amesto. and Fostyl ranged from 234.5 to 261.0 in the first season 2019, whereas, in the second season ranged from 245.0 to 280.5 compared to control treatment 1245.5, and 1284.5, respectively. On the other hand, AUDPC values of Reval, Facomel MZ, Cymonel, New copper and Dithane M 45 ranged from 313.5 to 419.0 (2022) and ranged from 333.5 to 428.5 (2023).



Fig. 7. The area under disease progress curve of twelve fungicides as a curatively application in open field during two seasons 2022 and 2023.

The following results demonstrate the average production of the Spunta cultivar when affected by natural infection of *P. infestans*, with the application of twelve different fungicides - both prophylactically and curatively - during the 2022 and 2023 growing seasons in the Etai Elbarood district of the El-Behera governorate (Table 9). All of the applied fungicides led to a significant increase in potato tuber yield when compared to untreated plants. Among the fungicides, Equation - Pro demonstrated the highest productivity per plot, followed by Amesto,

Consonto, Infinito and Folio Gold, with yields of 152.5, 151.1, 150.4, 150.2 and 149.8 Kg, respectively, when applied prophylactically. Similarly, when applied curatively, these fungicides produced yields of 148.3, 147.0, 147.3, 145.8 and 144.3 Kg/plot, respectively, compared to the control's yield of 79.7 Kg/plot. According to Table 9, this product has shown a rate of increase of 91.34% and 86.07% when used as a prophylactic or curative method, respectively. However, the lowest yield was observed with New copper and Dithane M 45 which produced 129.8 and 132.8 Kg/plot, respectively, when used prophylactically; and 124.3 and 128.9 Kg/plot, respectively, when used curatively.

Table 11. Mean value of Spunta cultivar production and rate of increase under natural infection of *P. infestans*with application of twelve fungicides as prophylactically and curatively at Etai Elbarood, El-Beheragovernorate during two seasons 2022 and 2023.

		Prophylactically			Curatively		
S. No.	Fungicide	*Yield/plot	Yield/fed.	**Rate of	Yield/plot	Yield/fed.	Rate of
		(42 m²) (kg)	(ton)	increase	(42 m²) (kg)	(ton)	increase
1	Saver	147.3 e	14.73 b	84.81	143.2 e	14.32 b	79.67
2	Consonto	150.4 c	15.04 a	88.71	147.3 b	14.73 ab	84.82
3	Infinito	150.2 c	15.02 a	88.45	145.8 c	14.58 ab	82.93
4	Amesto	151.1 b	15.11 a	89.58	147.0 b	14.70 ab	84.44
5	Reval	148.4 d	14.84 b	86.19	143.3 e	14.33 b	79.80
6	Dithane M 45	132.8 h	13.28 d	66.62	128.9 h	12.89 d	61.73
7	Folio Gold	149.8 c	14.98 b	87.95	144.3 d	14.43 b	81.05
8	Facomel MZ	142.1 f	14.21 c	78.29	137.1 f	13.71 c	72.02
9	Equation – Pro	152.5 a	15.25 a	91.34	148.3 a	14.83 a	86.07
10	Cymonel	135.1 g	13.51 d	69.51	131.2 g	13.12 d	64.62
11	Kabakeet (New	129.8 i	12.98 e	62.86	124.3 h	12.43 e	55.95
	copper)						
12	Fostyl	147.5 e	14.75 b	85.07	143.8 e	14.38 b	80.42
13	Control	79.7 j	7.970 f	0	79.7 i	7.970 f	0
L.S.D at 0.05		0.784	0.260		0.943	0.325	

*Mean of two growing seasons

**Rate of increase = (T-C/C) x 100.

Values in the same column followed by the same letter are not significantly differed at P < 0.05 level

DISCUSSION

The Egyptian population of P. infestans over two consecutive growing potato and tomato seasons has been identified into one predominant clonal lineage o 23_A1 based on simple sequence repeat markers and A1 mating type according to in vitro assay with standard isolates (A1 and A2). The current study didn't reveal a dynamic population amongst P. infestans isolates collected from Egypt where no A2 mating type and self-fertile isolates were detected. Our previous work revealed that A1 mating type in the Egyptian isolates of *P. infestans* has not only established itself in the Egyptian environment, but it also exhibits high genetic divergence and polymorphism (Arafa et al., 2018 and 2020). On the other hand, other studies are in opposite line with our results which reported that A1 and A2 mating types, as well as self-fertile isolates. were detected in the Egyptian population of P. infestans (El-Ganainy et al., 2016 and 2022; El-Korany, 2002) This indicated that there might be other issues affecting A2 mating type and oospores formation in the current study where only A1 mating type was observed under natural infection of potato and tomato crops in Egypt. Investigating such factors may open new insights for combating late blight and could be an important subject of upcoming trials. The oomycete P. infestans is a heterothallic pathogen and needs two mating types (A1 and A2) for sexual reproduction. However, the term heterothallic is not absolute in case of the late blight pathogen where a lot of reports mentioned that oospores have been produced from a single isolate (Fyfe and Shaw, 1992; Fry et al., 2009; Arafa et al., 2020; El-Ganainy et al., 2022).

Different potato varieties exhibit varying levels of tolerance or resistance to late blight, a fungal disease caused by *P. infestans*. Chemical fungicide treatment at long intervals is not sufficient to control foliar late blight in potato varieties with low levels of resistance, especially in hot spot regions. Conversely, varieties with high resistance levels exhibit minimal symptoms of foliar blight. This suggests that the need for fungicide treatment to manage *P. infestans* varies greatly among cultivars and is likely to be greater in commercial open fields than in small-plot trials. As such, potato cultivars Burren, Diamont, and Hermes are highly recommended for both potato breeding programs and commercial production due to their superior levels of resistance to late blight. It is possible that certain types of potatoes contain genes that provide resistance to late blight, but further research is necessary to identify these genes and locate the regions of the genome that control this resistance. To

effectively make use of resistant potato varieties, it is important that the resistance be strong, multi-genic, and quantitatively measurable (known as durable resistance). Field experiments have shown that the fungicides Equation-Pro, Consonto, Infinito, Amesto, Folio Gold, and Saver are the most effective options for managing potato late blight during both crop seasons (2022 and 2023), using either the protectant or curative application methods.

Various fungicides used in the management of potato and tomato crops contain different active ingredients such as Cymoxanil, Chlorothalonil, Famoxadone, Fenamidone, Propamocarb-HCL, Fluopicolide, Azoxystrobin, and Metalaxyl-M (Mefenoxam) with different formulas. The application of these active ingredients has been found to reduce the severity of potato late blight and area under disease progress curve and increase potato productivity compared to other treatments and non-treated control under the Egyptian conditions. Therefore, these fungicides can be considered a useful approach in integrated late blight management strategies in potato and tomato crops.

It is interesting to note that the combination of Propamocarb-HCL with fenamidone (Consonto fungicide) or with fluopicolide (Infinito fungicide) has shown promising results in reducing disease severity and AUDPC values in all tested plots for potato production. These positive results can be attributed to the fact that these active ingredients belong to different chemical groups, thus having different modes of action and target site. Propamocarb-HCL belongs to the carbamates group with cell membrane permeability as its target site, fenamidone belongs to QoI-fungicides that affect the cytochrome bc1 (cyt b) gene, whereas fluopicolide belongs to the benzamides chemical group that affects the cytoskeleton and motor protein through delocalization of spectrin-like proteins (FRAC, 2022).

Fenamidone is a type of fungicide that belongs to the imidazole chemical group. It has a single-site mode of action and can be used for both protective and curative applications. This active ingredient is effective in controlling phytopathogenic oomycete diseases and fungal spots on potato crops and other horticultural crops. In many countries, Fenamidone is often used in combination with other fungicides like mancozeb to reduce the potential risk of fungal races developing resistance to the fungicides (Johnson *et al.*, 2000; Muchiri *et al.*, 2009; Lal *et al.*, 2015). Azoxystrobin is a systemic fungicide that has a wide range of applications and was first introduced to the agricultural market in 1998. It belongs to the group of Qol-fungicides (Quinone outside Inhibitors) that prevent hyphal growth and spore germination by disrupting mitochondrial respiration through blocking electron transport at the cytochrome complex (III). Azoxystrobin can be used on various plant species such as potato, tomato, grapevines, apples, bananas, and other crops to combat different diseases. The fungicide Amesto (which contains Azoxystrobin as its active ingredient) has been found to be highly effective with low AUDPC values in controlling late blight. It has also shown a high productivity rate and rate of increase in both cropping seasons, regardless of the application approach (protectant or curative).

There have been reports of *P. infestans* isolates having resistance to metalaxyl in different countries including India, USA, Canada, Egypt, Bangladesh, China, Japan, Morocco, and Nepal. The resistance to fungicides within populations may be due to pathogenic variability during the sexual reproduction process of *P. infestans*, as well as the selection of the fittest clonal lineage isolates. However, our study found that when metalaxyl-M (Mefenoxam) was combined with other fungicides, such as chlorothalonil (fungicide Folio Gold), there was a high efficacy percentage (84.7 and 84.9%) in controlling late blight, and low values of AUDPC (211 and 211.8) as an average in both seasons, when applied as a protectant and curative. Chlorothalonil is an active ingredient with multi-site contact activity and chemical group phthalonitriles which can be used as a protectant fungicide (FRAC, 2022). Additionally, mixing of chlorothalonil and cymoxanil in one formula (Saver, commercial name) demonstrated effective application to manage late blight through reducing disease severity and AUDPC values as well as increasing potato tuber yield and rate of increase (145.3 Kg/Plot and 82.3) compared to nontreated control (79.7 Kg/plot) under natural infection.

Although, Mancozeb is a multisite (a broad-spectrum) protectant fungicide with more than six decades of application for fungal and oomycetes diseases, its application is less likely to be effective for late blight control in Egypt. Previous research conducted in various regions (Kathmandu Valley and Chitwan) of Nepal reported that *P. infestans* might have also developed tolerance strains to mancozeb (Sharma *et al.*, 2011; Gaire *et al.* 2014). On the other hand, Khadka *et al.* (2020) mentioned that Mancozeb decreased total late blight disease severity and increased potato production in the western plains of Nepal. Therefore, tolerance or fungicide resistance development of *P. infestans* population to mancozeb fluctuates in different regions within and/or between countries worldwide. Copper oxychloride is a contact fungicide with a group name inorganic (electrophiles) that has a long history of controlling oomycete pathogens and other plant diseases (Schutte et al., 1997; Kumar *et al.*, 2017; FRAC, 2022). Our findings demonstrated that fungicide Kabakeet (New copper) recorded the highest disease severity and lowest efficacy with 26.8 and 65.1%, respectively, as well as the highest value of AUDPC (412). Additionally, copper oxychloride gave the lowest potato yield and rate of increase with 127 Kg/Plot and

59.4, respectively, (average) compared to Equation – Pro which recorded the highest potato production and rate of increase with 150.4 Kg/Plot and 88.7(average), respectively.

CONCLUSION

Around the world, tomatoes and potatoes are severely damaged by late blight (LB). Fungicides work well to control LB. Egypt is where *P. infestans* isolates were gathered and subjected to in vitro testing. Field testing revealed that whereas Alaska, Spunta, Lady Rosita, and Mondial were very sensitive, Burren was highly resistant. With the lowest disease severity and maximum yield of potato tubers, Equation-Pro and Consonto were determined to be the most effective fungicides for managing LB. Strategies for crop improvement and potato breeding initiatives must take these findings into consideration.

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دراسة رد فعل بعض أصناف البطاطس التجارية والمبيدات الفطرية على مكافحة مرض الندوة المتأخرة

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يعتبر مرض الندوه المتأخرة المتسبب عن الكائن Phytophthora infestans (Mont.) de Bary، هو واحد من أكثر الأمراض تدميرًا للبطاطس والطماطم في جميع أنحاء العالم. ويعد استخدام مبيدات الفطريات عالميا طريقة فعالة لمكافحة الندوة المتأخرة في البطاطس. وفي هذة الدراسة تم جمع عزلات P. infestans من عدة حقول مختلفة في مصر وتم تشخيصها باستخدام المعلمات الدقيقة والحمض النووي للميتوكوندريا. كذلك تم أستخدام اثنى عشر مبيداً لآختبار قدرتها على مكافحة عزلات P. infestan في المعمل. كما تم تقييم تسعة أصناف من البطاطس تحت ظروف العدوي ا الصناعية. وأجريت تجارب حقلية لتقييم كفاءة استخدام المبيدات الفطرية في مكافحة الندوة المتأخرة وتقيمم إنتاجية أصناف البطاطس محل التجربة بمحافظة البحيرة خلال موسمين. وأوضحت النتائج أن عزلات P. infestans التي تم جمعها من الحقول كانت تتبع مجموعة التزاوج A1، والنسب النسلي يتبع A1_23 والحمض النووي للميتوكوندريا la/IR1. علاوة على ذلك، أوضحت الدراسة أن صنف البطاطس برن حقق مقاومة عالية وسجل أقل مساحة واقعه تحت منحني تطور المرض (AUDPC). ومن ناحية أخرى، كانت أصناف ألاسكا، سبونتا، ليدي روزيتا ومونديال شديدة الحساسية للمرض وسجلت أعلى مساحة تحت منحني تطور المرض AUDPC. وفي المعمل، أدى أستخدام جميع المبيدات الفطريه إلى تثبيط النمو القطري لعزلات EG-Pi1) P. infestans و EG-Pi2). وفي تجربه الحقل، أظهر استخدام مبيدي Equation-Pro وConsonto فعالية عالية في مكافحة المرض حيث تم تسجيل أقل نسبة مئوية للشدة المرضية وأقل مساحة واقعه تحت منحني تطور المرض AUDPC وذلك عند أستخدام المبيدات في الصوره الوقائية. والعلاجية خلال الموسمين محل الدراسه. علاوة على ذلك، زاد إنتاج درنات البطاطس بشكل ملحوظ عند أستخدام مبيد أكوجن - برو. وقد قدمت نتائج هذه الدراسة معلومات مهمة لبرامج تربية البطاطس وتحسين المحاصيل بالإضافة إلى استراتيجية الإدارة الناجحة لمكافحة اللفحة المتأخرة.

الكلمات المفتاحية: Phytophthora infestans، اللفحة المتأخرة، البطاطس، الاصناف، المبيدات الفطرية