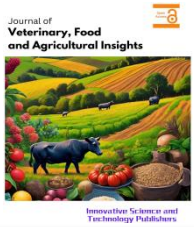
J. Vet. Food Agri. Insights. 01(1) 2024. 01-08
https://doi.org/10.xxxxx/jvfai.01.1.0051



Journal of Veterinary, Food and Agricultural Insights

ISSN: 1234-xxxx (Print), 1234-xxxx (Online)
https://isciencepress.com/journals/index.php/JVFAI/index

Research Article

Corresponding Author:
Name:
Atta ur Rehman Khan
Email:
aur.khan@iub.edu.pk

Article History:
Received: August 11, 2024
Revised: November 28, 2024
Accepted: December 13, 2024

Management of early blight of potato caused by *Alternaria solani* through fungicides under in vitro conditions

Ahmad Javed, Muhammad Adeel, Kashif Arshad, Tahir Mahmood, Muneeb Anwer, Talha Ikram, Muhammad Umer Shafique, Mudasir Hussain, Atta ur Rehman Khan*

Department of Plant Pathology, Faculty of Agriculture and Environment, The Islamia University of Bahawalpur, Pakistan.

ABSTRACT

Potato *(S olanum tuberosum L.)* is most frequently cultivated solanaceous crop worldwide. Early blight disease, caused by four types of Alternaria fungi, is a major threat to potato yield. The primary pathogen is *Alternaria solani*, which damages the leaf, stem, and potatoes, costing billions of dollars annually for management. The current study aimed to screening of potato lines, and their effective management. A screening methodology was used to assess the resistance of different potato cultivars against early blight caused by *A. solani*. The experiment followed a randomized complete block design (RCBD) with three replications, with a selection of 4-5 potato cultivars chosen for the screening process. Disease severity evaluations were conducted at predetermined intervals, starting from the appearance of early blight symptoms. The results indicated that all fungicides had significant effects on the number of spots/plant, plant height, plant weight, and no. of tubers. The application of different types of fungicides, such as Boscalid, Azoxystobin+Flutriafol, Tebuconazol, and Tebuconazole+Flutriafol, significantly reduced the effect of *A. solani* on potato. The most effective fungicide is the combinantion of Azoxystrobin+Flutriafol that exhibited the significant efficacy compared to all the other fungicides.

Keywords: *A. Solani*; Early blight; Fungicides; Potato

INTRODUCTION

Solanum tuberosum, commonly known as potato, is a highly prominent vegetable crop that holds immense global significance. Belonging to the Solanaceae family, this plant is a crucial provider of starchy nourishment in both sub-tropical and temperate areas (Kumari, 2012). The cultivation of *Solanum tuberosum*, commonly known as potatoes, is noteworthy for its expansion into tropical regions, particularly during the winter season. The potato, which traces its origins to South America, boasts a significant historical association with this geographical area (Hijmans and Spooner, 2001). Potato, considered the third most widely produced and consumed crop globally, surpassing only rice and wheat, is a staple food for nearly a billion people worldwide, consumed in various forms (Anwar et al., 2015). Recent data reveals that global potato production in 2021 reached an impressive 376 million metric tons, cultivated across an area of 18.13 million hectares (Mha). In the same timeframe, Pakistan contributed to this production with a total of 7.74 million metric tons of potatoes harvested from an area of 0.15 Mha (FAOSTAT, 2021).

In 2021, Pakistan successfully exported approximately 0.4 million tons of potatoes to various international markets, including Afghanistan, Sri Lanka, UAE, Russia, Qatar, Azerbaijan, Malaysia, and Iraq, among others. Several potato varieties are cultivated in Pakistan, such as Constance, Vogue, Kuroda, Esmee, Rudolph Santé, Arizona, and Meryem etc. The primary regions for potato cultivation

in Pakistan are Okara, Depalpur, and Sahiwal.

Potatoes, as a vital staple crop, play a significant role in promoting health and nutrition due to their nutrient-rich composition and potential health advantages. Carbohydrates are abundantly present in potatoes, offering a substantial energy source for the body (Burlingame et al., 2009). Moreover, potatoes are rich in dietary fiber, which aids in digestion and supports regular bowel movements, contributing to gastrointestinal health (Geliebter et al., 2013, Ahmad et al 2011). Additionally, potatoes are a valuable source of essential vitamins such as vitamin C and B vitamins, as well as minerals like potassium, which are crucial for maintaining proper nerve and muscle function (Wei et al., 2021).

Potato early blight is a common fungal disease that affects potato plants. It is caused by the polycyclic Fungi Imperfecti Alternaria solani Sorauer. The disease is known to cause significant yield losses and can also affect the quality of the harvested potatoes. The fungus infects the leaves of the potato plant, causing the formation of dark brown lesions with concentric rings. These lesions can coalesce, leading to the death of the affected leaves (Saha and Das, 2017). Effective management strategies, such as crop rotation, fungicide application, and the use of resistant potato cultivars, are essential for controlling the disease and minimizing its impact on potato production (Pscheidt, 1986). The causal organism responsible for early blight was initially identified and documented by (Ellis and Martin in 1882) as Macrosporium solani. The initial documentation of the fungal organism as a parasitic entity and its correlation with the occurrence of potato leaf blight was reported in Australia by (Galloway, 1891). Alternaria solani, the pathogenic microorganism responsible for the occurrence of early blight disease, possesses the capability to endure within the soil environment in the form of mycelia, which are thread-like structures, as well as spores. The dispersal of these spores can occur through wind or water droplets, leading to the infection of foliage at any developmental stage (Mulder & Turkensteen, 2005). The manifestation of foliage infection symptoms is usually observed during the growth of tubers, while the progression of lesions intensifies as leaves approach their physiological maturity and senescence stage (Rotem, 1994). It has been observed that elevated temperatures have a positive correlation with the augmentation of both A. solani infection and sporulation rates. The germination of conidia and the subsequent infection of pathogens can be significantly influenced by environmental factors, including but not limited to high relative humidity, rainfall, and the accumulation of dew. It is noteworthy that early blight can have a significant impact on alternative host plants, including tomato, eggplant, pepper as well as solanaceous and non-solanaceous weeds (Jones et al., 1993; Mulder and Turkensteen, 2005). The present study aims to conduct an in-vitro assessment of various fungicides in order to determine their efficacy against a specific fungal pathogen.

MATERIALS AND METHODS

Isolation and Purification of Fungal Pathogen Pathogen

The potato plants were cultivated at the Vegetable Research area of Islamia University of Bahawalpur, adhering to the prescribed set of guidelines for crop cultivation in the specific region. Leaves exhibiting indications of early blight were collected from the field and carefully enclosed within polythene bags. The leaf samples underwent surface sterilization by treating them with a solution of 0.5% sodium hypochlorite. Subsequently, the samples were extensively rinsed with distilled sterilized water on multiple occasions. Leaf fragments of comparable size, comprising roughly equal proportions of infected and healthy tissue, were surgically removed. Subsequently, the leaf fragments were carefully positioned onto a culture medium consisting of potato dextrose agar (PDA), which was further enhanced with streptomycin to inhibit the growth of bacteria and minimize the risk of contamination. The Petri dishes were subjected to incubation at a controlled temperature of 25℃ ± 2℃ for a duration of one week, while maintaining a 12-hour cycle of darkness and light, as described by (Meena et al., 2017). The pathogen culture derived from the isolated spores was subjected to additional purification using the single spore isolation technique. The purified culture was then maintained on Potato Dextrose Agar (PDA) slants for subsequent investigations.

Identification of the Pathogen Culture Isolate

The fungal culture that was acquired was determined to be a potential Alternaria species using conventional taxonomic methods, which involved analyzing the physical characteristics of the colony

and spores. Conidia were extracted from the periphery of the isolated culture and mounted onto a microscopic slide coated with a lacto phenol solution to facilitate the examination of spore properties. The morphological traits of the fungal colony were methodically examined at regular intervals of 24 hours. The identification of the pathogen was accomplished through a meticulous procedure that entailed cross-referencing detailed accounts of the phytopathogen documented in numerous authoritative manuals and scholarly publications (Subramanium, 1971; Barnett and Hunter, 1998; Gilman and Joseph, 1998).

Pathogenicity Test

In order to validate the pathogenicity of the identified isolate of Alternaria spp., Koch's postulates were utilized. The experimental subjects consisted of robust potato plants belonging to a susceptible cultivar, chosen for their overall good health. The potato propagules underwent surface sterilization using a solution of 0.5% sodium hypochlorite, followed by multiple rinses with sterilized distilled water to ensure complete removal of any contaminants. Following the surface sterilization process, the plant propagules were subjected to air-drying before being carefully placed into pots containing sterilized soil. The specimens were permitted to undergo a growth period of one month under optimal environmental conditions. A suspension of spores from the Alternaria spp. isolate was prepared and uniformly applied to one-month-old potato plants. In order to preserve the required level of humidity, the plants that had been inoculated were enveloped with plastic bags. To establish a control group, sterilized distilled water was applied via spraying onto the plants designated as controls. The manifestation of symptoms was documented at 2, 4, and 8 days after inoculation (DAI). Subsequently, the phytopathogen was isolated once again from the leaves of the previously inoculated plants and subjected to a comparative analysis with the original culture in order to validate its identity.

Evaluation of the Fungicides against the Fungal Pathogen

The effectiveness of five fungicides, specifically Propiconazole 25EC, Carbendazim 50WP, Curzate 50WP, Mancozeb 75WP, and Copper oxychloride, in managing the pathogen was assessed utilizing the Poisoned Food Technique (Schmitz, 1930). The Potato Dextrose Agar (PDA) medium was enriched with fungicides at concentrations of 50, 100, 250, and 500 parts per million (ppm), respectively. A Petri plate devoid of any fungicides was used as the control. The media were carefully transferred into sterilized Petri plates and left undisturbed until they solidified. A pure culture of the Alternaria solani isolate, seven days mature, was inoculated by carefully positioning 5mm discs at the central region of each plate, including the control sample. The experimental treatments were replicated three times and securely sealed using paraffin wax strips. Subsequently, they were incubated at a controlled temperature of 25℃ ± 2℃. The progression of colony growth in each experimental group was observed at regular 24-hour intervals until the Petri dishes were fully colonized by the fungal organism. The percentage of inhibition in the fungicide treatments was determined by calculating the relative difference compared to the control, utilizing the formula provided by (Sundar et al., 1995).

$$\%\ \text{inhibition zone} = \frac{X - Y}{X} \times 100$$

Where, X= Growth of fungal pathogen on control plate, Y= Growth of fungal pathogen on fungicide treated plate

Statistical Analysis

Statistix 8.1 software will be used for the analysis in a Completely Randomized Design (CRD) to determine the significance and non-significance of fungicides in disease management associated with maize grains (Russel and Eisensmith, 1983).

Results

Effect of fungicides in reducing the mycelial growth of Alternaria solani

To effectively control Alternaria solani, an experimentation involving the application of three distinct fungicides, namely Mancozeb, Topsin M, and Difenoconazole, at varying concentrations of 100, 150, and 200 parts per million (ppm) was conducted using a statistical approach known as Complete Randomized Design (CRD). The assessment of these fungicides' impact on the mycelium development of Alternaria solani was performed at specific intervals of 3, 5, and 7 days.

Upon the lapse of 3 days, it was observed that all tested fungicides exerted a significant inhibitory effect on the mycelial growth of Alternaria solani. Notably, Mancozeb demonstrated a remarkable level of significance, yielding values of 0.54, 0.35, and 0.20 centimeters (cm) at concentrations of 100, 150, and 200 ppm, respectively. Topsin M also exhibited inhibition of Alternaria solani growth after 3 days, with values of 0.73, 0.63, and 0.47 cm at concentrations of 100, 150, and 200 ppm, respectively. In contrast, fungicide Difenoconazole displayed the least efficacy in suppressing mycelial growth, registering values of 0.78, 0.61, and 0.53 cm at concentrations of 100, 150, and 200 ppm, respectively.

After 5 days, a similar trend persisted, where all fungicides continued to significantly impede the mycelial growth of Alternaria solani. Mancozeb remained highly significant, yielding values of 0.62, 0.41, and 0.26 cm at concentrations of 100, 150, and 200 ppm, respectively. Topsin M also exhibited inhibition after 5 days, with values of 0.84, 0.73, and 0.54 cm at concentrations of 100, 150, and 150 ppm, respectively. However, Difenoconazole remained the least effective, recording values of 0.92, 0.74, and 0.57 cm at concentrations of 100, 150, and 200 ppm, respectively.

Upon reaching the 7-day mark, the trend persisted, with all fungicides significantly restraining the mycelial growth of Alternaria solani. Mancozeb maintained its high level of significance, with values of 0.71, 0.50, and 0.34 cm at concentrations of 100, 150, and 200 ppm, respectively. Topsin M also exhibited continued inhibition after 7 days, registering values of 0.96, 0.84, and 0.66 cm at concentrations of 100, 150, and 200 ppm, respectively. Nonetheless, fungicide Difenoconazole remained the least effective in curbing mycelial growth, recording values of 1.14, 0.77, and 0.66 cm at concentrations of 100, 150, and 200 ppm, respectively.

Table 1: Analysis of variance table for the evaluation of fungicides against *Alternaria solani* after 3 days.

Source	DF	SS	MS	F	P
Treatment	2	0.2882	0.14410	161.11	0.0000*
Concentration	3	11.8911	3.96371	4431.47	0.0000*
Trt x Conc	6	0.0850	0.01417	15.85	0.0000*
Error	24	0.0215	0.00089		
Total	35	12.2858			

Grand Mean= 0.8664 CV= 3.45 * = Significant at P < 0.05

Table 2: Assessment of mean values for fungicides treatments on *Alternaria solani*after 3 days of incubation.

Treatments	Concentrations		
	100 ppm	150ppm	200ppm
	Mean (cm)	Mean (cm)	Mean (cm)
Mancozeb	0.54DE	0.35F	0.2G
Topsin M	0.73B	0.63C	0.47E
Difenoconazole	0.78B	0.61CD	0.53DE
Control	1.86A	1.85A	1.81A
HSD		5.098	

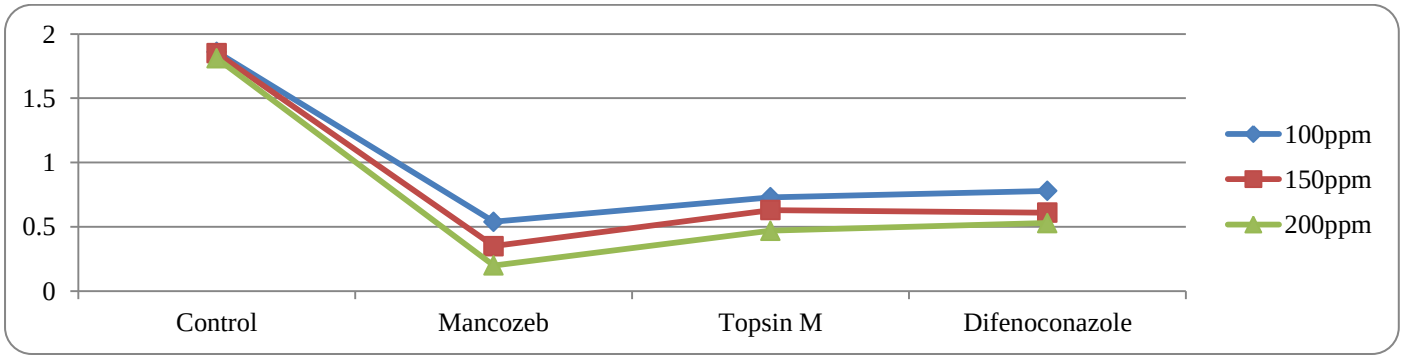


Figure.1: Graph showing effect of fungicides on development of *Alternaria solani* after 3rd day.

The impact of different fungicides including Mancozeb, Topsin M, and Difenoconazole on the growth of *Alternaria solani* is depicted in figure. The graph illustrates that after a 3-day period Mancozeb exhibited the highest efficacy among all the fungicides particularly at a concentration of 200 ppm.

Table 3: Analysis of variance table for the evaluation of fungicides against *Alternaria solani* after 5 days.

Sources	DF	SS	MS	F	P
Treatment	3	20.4919	6.83063	11076.7	0.0000*
Concentration	2	0.3726	0.18631	302.12	0.0000*
Trt x Conc	6	0.1341	0.02235	36.24	0.0000*
Error	24	0.0148	0.00062		
Total	35	21.0134			

Grand Mean= 1.0600 CV= 2.3* = Significant at P < 0.05

Table 4: Assessment of mean values for fungicides treatments on *Alternaria solani*a fter 5 days of incubation.

Treatments	Concentrations		
	100 ppm	150ppm	200ppm
	Mean (cm)	Mean (cm)	Mean (cm)
Mancozeb	0.62E	0.41G	0.26H
Topsin M	0.84C	0.73D	0.54F
Difenoconazole	0.92B	0.74D	0.57EF
Control	2.34A	2.35A	2.35A
HSD	3.533		

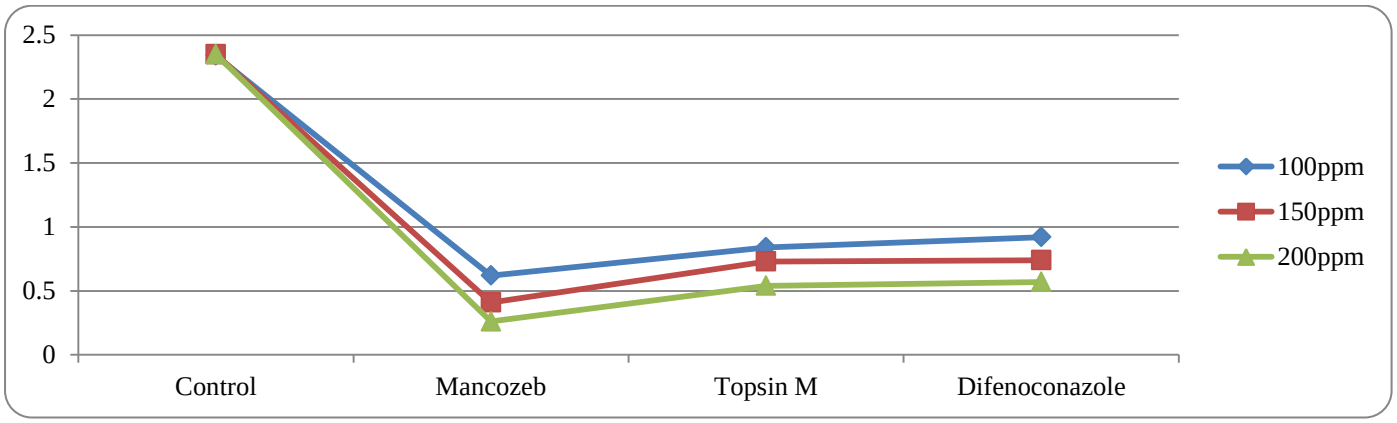


Figure 2: Graph showing effect of fungicides on development of *Alternaria solani* after 5thday.

The impact of different fungicides, including Mancozeb, Topsin M, and Difenoconazole, on the growth of *Alternaria solani*is depicted in the figure. The graph illustrates that after a 5-day period, Mancozeb exhibited the highest efficacy among all the fungicides, particularly at a concentration of 200 ppm.

Table 5: Analysis of variance table for the evaluation of fungicides against *Alternaria solani* after 7 days.

Sources	DF	SS	MS	F	P
Treatment	3	32.9701	10.9900	9487.80	0.0000*
Concentration	2	0.5164	0.2582	222.91	0.0000*
Trt x Conc	6	0.2139	0.0357	30.78	0.0000*
Error	24	0.0278	0.0012		
Total	35	33.7282			

Grand Mean 1.2814 CV 2.66 * = Significant at P < 0.05

Table 6: Assessment of mean values for fungicides treatments on *Alternaria solani* after 7 days of incubation.

Treatments	Concentrations		
	100 ppm	150ppm	200ppm
	Mean (cm)	Mean (cm)	Mean (cm)
Mancozeb	0.71EF	0.5G	0.34H
Topsin M	0.96C	0.84D	0.66F
Difenoconazole	1.14B	0.77DE	0.66F
Control	2.91A	2.95A	2.89A
HSD	5.098		

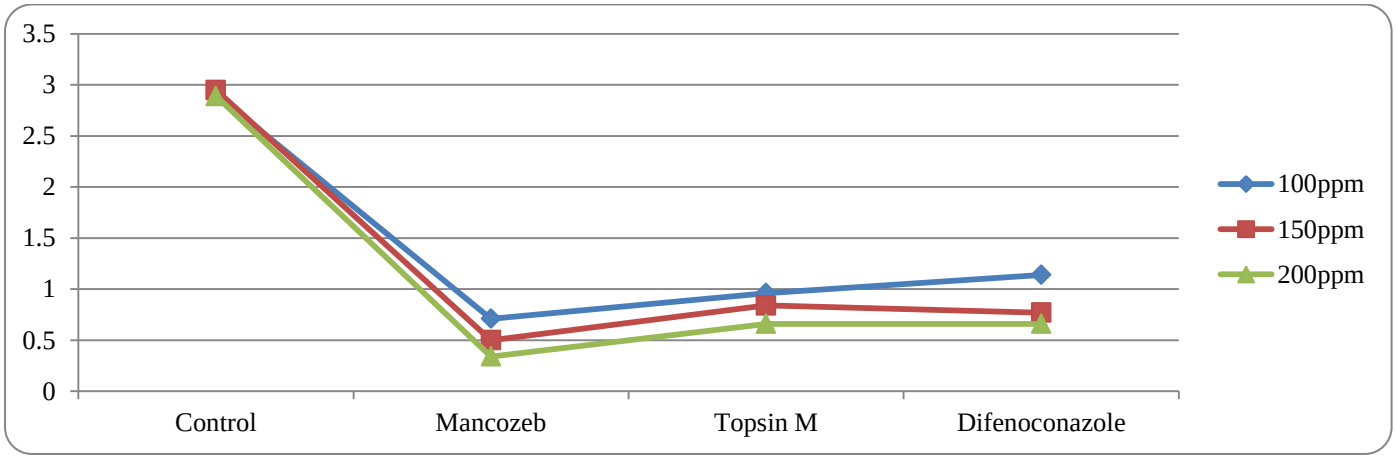


Figure 3: Graph showing effect of fungicides on development of *Alternaria solani*after 7thday.

The impact of different fungicides including Mancozeb, Topsin M, and Difenoconazole on the growth of *Alternaria solani* is depicted in the figure. The graph illustrates that after a 7-day period, Mancozeb exhibited the highest efficacy among all the fungicides particularly at a concentration of 200 ppm.

DISCUSSION

Solanum tuberosum, is widely grown crop and consumed in many ways. Potatoes, originally from the Andes in South America, are now a global food crop due to their nutritional value, adaptability to many climates, and culinary diversity (Bradeen et al., 2011). Potatoes are known for their underground tuberous structures and include carbs, vitamins (especially C and B6), and minerals. Potato growing improves food security and agricultural economics by using both traditional and modern methods (DeFauw et al., 2012).

A field study examined the efficiency of several fungicides in mitigating Alternaria solani early blight. Boscalid, Azoxystrobin+Flutriafol, Tebuconazole, and Tebuconazole+Flutriafol were tested for disease control. Azoxystrobin+Flutriafol was the most effective fungicide in reducing disease severity. Other therapies were more effective than Tebuconazole in disease control. Even though their efficacy varied, all fungicides suppressed the illness better than the control group. These data show that Azoxystrobin+Flutriafol may be the best early blight management strategy and that the tested fungicides reduced disease severity in field settings.

Experimental fungicides were tested to combat early blight (Alternaria solani) in Australian potatoes. Boscalid, azoxystrobin, and difenoconazole worked well three days before or after inoculation. Applying Boscalid a day earlier fully reduced illness. Field trials in different regions showed that 4-6 spray applications, combining boscalid + metiram early on, improved early blight control and tuber yields by 20%. Similar to the current study, growers can prevent illness and boost output by using protective fungicides like boscalid early and curative ones like difenoconazole for existing infections (Horsfield et al., 2010).

Foliar fungicides, both premium single-site mode-of-action "specialty" and conventional protectant multisite, are essential for potato early blight management in the US. Individual participant data meta-analysis was used to evaluate fungicide efficacy, tuber yields, and early blight management timing. Individually applied specialty and standard fungicides like chlorothalonil and mancozeb were compared. There were significant variations ($P < 0.0001$) in overall efficacy and yield among treatments. The study found significant differences ($P < 0.0001$) in early blight development among treatments during the growth season. Compared to conventional fungicides, specialty fungicides reduced disease severity from growth initiation ($P = 0.0139$) to tuber maturation ($P = 0.0009$). Even though they cost more, specialized fungicides were best for early blight treatment in North Dakota and Minnesota (S. Yellareddygari et al., 2016).

Azoxystrobin (a strobilurin) and epoxiconazole (a sterol biosynthesis inhibitor) fungicides were tested on phyllosphere fungi, potato senescence, and yield in field and controlled circumstances. Fungicides boosted yield and green leaf area retention in field experiments without evident disease. One trial found that azostrobin maintained green leaf area and yielded better than epoxiconazole. Plant senescence was accelerated by fungicides that inhibited saprophytic fungi on leaves and stimulated *Altrnaria* spp. defence responses. Azoxystrobin reduced defence reactions more than epoxiconazole. Fungicides reduced accelerated senescence caused by saprophytic fungus inoculation without yield benefits. Azoxystrobin reduced infection-induced respiration and saprophyte spore germination and growth in growth chamber tests. The research implies azoxystrobin's improved defence inhibition may boost field yield (Christ, 1990).

Potato early blight control was studied again. Potato early blight (Alternaria solani) reduces yields and is controlled by regular fungicide applications. Azoxystrobin, released in 1999 in the US, originally showed great control. Azoxystrobin and other Quinone outside inhibitor (QoI) fungicides covered 80% of potato acreage in three years. A. solani isolates from Wisconsin showed reduced azoxystrobin sensitivity by 2001. Field studies in 2001–2003 evaluated fungicide programmes and resistance management. The then-recommended azoxystrobin and chlorothalonil programme controlled early blight under favourable conditions. In vitro sensitivity of A. solani isolates from these trials was highest in untreated, chlorothalonil-alone, and azoxystrobin-chlorothalonil alternating plots. Three single-nucleotide polymorphisms (SNPs) lowered azoxystrobin sensitivity, with the TTA mutant often recovered in field trials. Wild-type isolates were 22%, 4%, and 22% in 2001, 2002, and 2003 (Ding et al., 2019; S. K. Yellareddygari et al., 2016).

CONCLUSION

The study demonstrates the effectiveness of various chemical-based fungicides, including Azoxystrobin, Tebuconazole, Flutriafol, and Boscalid, in managing potato diseases. All tested fungicides effectively suppressed the disease, with Azoxystrobin and Flutriafol being the most effective combination for early light management. This highlights the positive impact of these fungicides on disease severity in field conditions.

REFERENCES

Ahmad, N., M.A. Khan, S. Ali, N.A. Khan, R. Binyamin, A.F. Sandhu and A. Rehman, 2011. Epidemiological studies and management of potato germplasm against PVX and PVY. Pakistan Journal of Phytopathology. 23(2): 159-165.

Anwar, D., D. Shabbir, M.H. Shahid and W. Samreen. 2015. Determinants of potato prices and its forecasting: A case study of Punjab, Pakistan. University Library of Munich, Germany https://mpra.ub.uni-muenchen.de/66678.

Barnett HL, Hunter BB. Illustrated genera of imperfect fungi. 1998. (No. Ed. 4). American Phytopathological Society (APS Press).

Bradeen, J. M., Haynes, K. G., & Kole, C. (2011). Introduction to potato. *Genetics, genomics and breeding of potato*, 1-19.

Burlingame, B., Mouillé, B., & Charrondiere, R. 2009. Nutrients, bioactive non-nutrients and anti-nutrients in potatoes. Journal of food composition and analysis, 22(6), 494-502.

Christ, B. J. (1990). Influence of potato cultivars on the effectiveness of fungicide control of early blight. *American Potato Journal*, *67*, 419-425.

DeFauw, S. L., He, Z., Larkin, R. P., & Mansour, S. A. (2012). Sustainable potato production and global food security. In *Sustainable potato production: global case studies* (pp. 3-19). Springer.

Ding, S., Halterman, D. A., Meinholz, K., & Gevens, A. J. (2019). Distribution and stability of quinone outside inhibitor fungicide resistance in populations of potato pathogenic Alternaria spp. in Wisconsin. *Plant Disease*, *103*(8), 2033-2040.

Ellis J B and Martin G B. 1882. Macrosporium solani E&M. American Naturalist.16: 1003

FAOSTAT. 2017. Food and Agricultural Organization Statistical database, Crop production. http://faostat3.fao.org/download /Q/QC/ E (accessed on June 22, 2023).

Galloway B T. 1891. The new potato disease. Gardenand Field, Adelaide, Australia. 16: 158.

Geliebter, A., Lee, M. I., Abdillahi, M., and Jones, J. 2013. Satiety following intake of potatoes and other carbohydrate test meals. Annals of Nutrition and Metabolism, 62(1), 37-43.

Gilman JC, Joseph C. 1998. A manual of soil fungi. Daya Books.

Hijmans, R.J. and Spooner, D.M. 2001. "Geographic distribution of wild potato species". Am. J. Bot., (Botanical Society of America) 88 (11): 2101–12.

Horsfield, A., Wicks, T., Davies, K., Wilson, D., & Paton, S. (2010). Effect of fungicide use strategies on the control of early blight (Alternaria solani) and potato yield. *Australasian Plant Pathology*, *39*, 368-375.

Jones JB, Jones JP, Stall RE, Zitter TA. 1993. Compendium of tomato diseases. St. Paul (MN): American Phytopathological Society, 73.

Kumari, S. 2012. Influence of drip irrigation and mulch on leaf area maximization, water use efficiency and yield of potato (*Solanum tuberosum L.*). Journal of Agricultural Science, 4(1), 71.

Meena M, Swapnil P, Upadhyay RS. 2017. Isolation, characterization and toxicological potential of Alternariamycotoxins (TeA, AOH and AME) in different Alternaria species from various regions of India. Scientific Reports. 7(1):1-19.

Mulder A, Turkensteen LJ. 2005. Early blight. Potato diseases. Den Haag: PlantijnCasparie; p. 280.

Pscheidt, J. W. 1986. Early blight of potato and tomato: a literature review. Univ. Wis. Ext. Coop. Ext. Serv. 3376. Research Division of the College of Agriculture and Life Sciences University of Wisconsin-Madison, Madison, WI.

Rotem J. 1966. Variability in Alternaria porri f. solani. Israel J Bot. 15:48–57.

Russel, D.F. and S.P. Eisensmith. 1983. MSTAT-C. Crop Soil Science Department, Michigan State Uni. East Lancing, Michigan.

Saha, P., & Das, S. 2017. Temporal and spatial epidemic development of early blight (Alternaria solani Ell. and Mart) in tomato as a function of different soil nutrition sources. New Agriculturist, 28(1), 171-185.

Schmitz, H. (1930). Poisoned food technique. *Industrial and Engineering Chemistry-Analytical Edition*, *2*(4), 361-363.

Subramanian CV. 1971. Hyphomycetes. An account of Indian species, except Cercosporae,

Sundar, A.R., N.D. Das and D. Krishnaveni. 1995. In vitro antagonism of *trichoderma spp*. against two fungal pathogens of castor. Ind. J. Plant Protec. 23:152-155.

Wei, M., Wang, H., Ma, T., Ge, Q., Fang, Y., and Sun, X. 2021. Comprehensive utilization of thinned unripe fruits from horticultural crops. *Foods*, *10*(9), 2043.

Yellareddygari, S. K., Pasche, J. S., Taylor, R. J., Hua, S., & Gudmestad, N. C. (2016). Beta regression model for predicting the development of pink rot in potato tubers during storage. *Plant Disease*, *100*(6), 1118-1124.

Yellareddygari, S., Pasche, J. S., Taylor, R. J., & Gudmestad, N. C. (2016). Individual participant data meta-analysis of foliar fungicides applied for potato early blight management. *Plant Disease*, *100*(1), 200-206.