Effect of different concentration of nano-fungicides on mycelia radial growth of *Phytopthora infestans.*

ABSTRACT

Potato (*Solanum tuberosum* L.) popularly known as the "King of Vegetables" is a starchy, tuberous crop, belongs to family Solanaceae. Potatoes play an important role in the daily human diet because having a copious source of minerals, vitamins, antioxidants and micronutrient. The potato is a basically temperate crop, but it grows under a wide range of climatic conditions. Potato crop are generally affected by different types of biotic and abiotic diseases. Among all diseases, Late blight of potato, caused by the *Phytophthora infestans* (Mont) de Barry is one of the most devastating diseases of potato worldwide. In India, the losses caused by the disease are 10-20% in Uttar Pradesh, 10-15% in West Bengal and Punjab, 10-15% in Karnataka and Uttrakhand also have been reported during 2013-14. The management of the disease can be done through cultural, mechanical, use of resistant variety, biological and chemical *etc*. The efficacy of current strategies for management of disease is limited. Considering the destructive nature of pathogen and lack of efficient control measures, development of alternative or complementary approaches for management of this disease is highly desirable. However reported use of same chemicals may raise resistant strain among the pathogens. Therefore, use of Nano-fungicidal control of spot blotch disease cannot be overstated. Though, the disease can be managed with various nanofungicides under in vitro condition assessed for their efficacy against the disease. The effect of four nanofungicides *i.e.*, Agritec nanofungicide, Silver nanofungicide, Selenium nanofungicide and Nickel nanofungicide against *P. infestance* was evaluated at different concentrations (50, 75 and 100 ppm) by poison food technique. The in vitro experiment revealed a noticeable reduction in the radial mycelial growth of *P. infestance* as the concentration of nanofungicides increased. Specifically, at 7 days after inoculation (DAI), the following nanofungicides at a concentration of 100 ppm exhibited the highest levels of mycelial growth inhibition when compared to the control: Silver nanofungicide (72.45%), Agritec nanofungicide (70.21%), Selenium nanofungicide (68.77%), and Nickel nanofungicide (64.65%).

Introduction

Potato (*Solanum tuberosum* L.) popularly known as the "King of Vegetables" is a starchy, tuberous crop, belongs to family Solanaceae. It is the world's fourth-largest food crop following rice, wheat and maize. Potatoes play an important role in the daily human diet because having a copious source of minerals, vitamins, antioxidants and micronutrients (Ulger*et al*., 2018). The potato crop produces more edible energy per unit area and time as compared with wheat, rice and maize (Dutt, 2008). Potato as vegetable cultivation is an important constituent of Indian agriculture due to its high yield and ability to generate on-farm and in-farm employment (Ghosh, 2010). The potato crop is grown in almost all states of India, except Kerala, under a wide range of agro-climatic conditions (Kumar and Chandra,

2018). Out of 100 % total area of potatoes, 90 %area is located in subtropical plains, 6 % in the hills and the rest 4 % located in the plateauregion of peninsular India (Chadha, 2009).

Potato is the best source of dietary energy and some other beneficial micronutrients, and its protein and mineral content is very high when compared with other root and tuber crops Freshly harvested potato tubers contain around 80 % water and20 % dry matter (Wichrowska, 2022). More than 75 % of the dry matter is starch but they also contain protein, fiber and small amounts of fatty acids. In addition, potatoes contain the lower amount of fat but tubers are a rich source of minerals such as potassium, phosphorus, magnesium and various vitamins like B1, B3 and B6. Among vitamins, the amount of vitamin C is very high in the tuber.

Potato is considered as fourth most important food crop which is grown in more than 100 potato-growing countries around the world under different climatic conditions such as tropical, temperate and subtropical. More than one billion people around the world eat potatoes. According to the FAO data, a total of 376 million tonnes of potatoes were produced worldwide, with China (94 million tonnes) and India (54 million tonnes) the largest potato producing countries in 2021. FAO figures indicate that the total area harvested globally in 2021 was 18,132,694 hectares – a world average of about 21 tonnes per hectare, China contributes about shares 26.3 % of total potato production followed by (U.S. Potato production, 2020). Second Russian Federation in the world.

In India the major potato growing states such as Uttar Pradesh, West Bengal, Bihar, Punjab, Haryana and Madhya Pradesh. About 85 % of potatoes are cultivated in Indo-gangetic plains of North India. Uttar Pradesh is the leading state of India in terms of area, production and productivity of potatoes. In India, Uttar Pradesh shares 31.36% of total potato production followed by West Bengal (23.29%), Bihar (13.22 %) and Gujarat (7.43 %).

Potato crop are generally affected by different types of biotic and abiotic diseases. Biotic diseases are mainly caused by fungi, bacteria and viruses (Ashraf *et al*., 2012), nematodes and mycoplasma which adversely affected the potato quality as well as production in almost all over the world (Gul *et al*., 2013), (Nazarov *et al*., 2020). Among all diseases, Late blight of potato, caused by the *Phytophthora infestans* (Mont) de Barry is one of the most devastating diseases of potato worldwide (Hardy *et al.*, 1995). Under favourable conditions Late blight can cause tuber losses of 100 per cent in susceptible germplasm and crop losses and crop protection costs are substantial (Henfling, 1987).

The disease is distributed all over the world like North & South America, Europe, Asian continents (Fry *et al*., 1993; Chowdappa *et al*., 2013). The annual economic losses caused by the disease worldwide have been about 170 billion US dollars (Haverkort*et al*., 2009; Wu and Gui, 2012). In India, the losses caused by the disease are 10-20% in Uttar Pradesh, 10-15% in West Bengal and Punjab, 10-15% in Karnataka and Uttarakhand also have been reported during 2013-14 (Lal *et al.*, 2016) but the amount of losses depends on the variety and plant protection measures adopted by the potato growers.

In India, late blight of potato is one of the important fungal diseases which appear in every year, in serious form. Disease development is favoured by cool, moist weather. Reducing initial amount of inoculum by using sanitation practices is an effective approach for disease management, but main emphasis should be to slow down disease progress. Hence management practices are very difficult and should be focused both on preventive as well as curative approaches. Host resistance, inter-cropping, time of planting and judicious use of fungicides are important components in management package (Shekhawat, 2000).

The management of the disease can be done through cultural, mechanical, use of resistant variety, biological and chemical *etc*. Cultural control measures such as eliminating cull piles and volunteer potato, soil solarization, crop rotation, using proper harvesting and storage practices, can be used to reduce the pathogen populations by reducing its survival, dispersal and reproduction (Garrett and Dendy 2001) but cannot control the disease in standing crop. Use of resistant variety is another alternative method for management of disease but It had been difficult to develop potato varieties with suitable long-lasting resistance to late blight because although specific genes were known they were overcome rapidly by mutants in local fungus populations (Fry and Goodwin 1997a). At present, there is no tolerant and/or resistant potato cultivar available against late blight pathogen in the world. Biological control by antagonists has attracted much attention because of being eco-friendly to environment and human health (Harmendez *et al.,* 2005). The efficacy of current strategies for management of disease is limited. Considering the destructive nature of pathogen and lack of efficient control measures, development of alternative or complementary approaches for management of this disease is highly desirable. New fungicides are introducing in the country every year against late blight pathogen but none of them found effective due to the dynamic changes of population structures of *P. infestans*.

Risks associated with pesticide use have received considerable and no single fungicide has consistently and effectively suppressed losses related to *P. infestans* epidemics. In contrast to conventional application of fungicides antibiotics, nanoparticles are most important strategy to manage plant diseases. Nanotechnology is the application of science and technology to control the matter at molecular level. The development of nanotechnology in conjunction with biotechnology has significantly expanded the application domain of nanomaterials in various fields. A variety of carbon-based, metal and metal oxide-based dendrimers (nano-sized polymers) and bio-composite nanomaterials (Nair *et al.,* 2010) are being developed. Nanoparticles are synthesized by physical, chemical and biological methods. Physical and chemical methods are energy intensive and cause toxic chemicals whereas, biogenic technique is eco-friendly, non-toxic and economically viable.

Somalraju *et al.* (2022) reported that treatment of potato seed with different Se rates, and applying different foliar Se rates on potato plants subsequently inoculated with the late blight pathogen in a greenhouse setting, and found that foliar Se application alone or combined with Se seed treatment reduced the severity and incidence of late blight, and activated the production of secondary metabolites in potato leaves and tubers. Further, *in vitro* studies showed that Se directly inhibited the mycelial growth of *P. infestans* and other

important plant pathogens, including *Fusarium graminearum* and *Sclerotinia sclerotiorum*. Various forms of silver ions and nanoparticles were tested to examine the antifungal activity on two plant-pathogenic fungi, *Bipolaris sorokiniana* and *Magnaporthe grisea*. *In vitro* assays indicated that silver ions and nanoparticles had a significant effect on the colony formation of two pathogens at concentration of 50 ppm (Jo *et al*., 2009).

Elamawi *et al*. (2013) studied the effect of silver nanoparticles (20-30 nm) against rice leaf blast caused by *Magnaporthae grisea*. Under laboratory conditions, the application of four concentrations *viz.*, 25, 50, 100 and 200 ppm of silver nanoparticles showed significant inhibition of both hyphal growth and number of colonies formed in a dose-dependent manner (Shantamma, 2016).

Ouda *et al.*, (2014) evaluated the efficacy of AgNPs at 15 ppm against *Alternaria alternate* and *Botrytis cinerea*, and recorded mycelial inhibition of 59.30 and 52.90 per cent, respectively. Silver nanoparticle supplemented medium showed 12, 36 and 41 per cent hyphal growth of soil borne pathogens *viz*., *Rhizoctonia solani*, *Sclerotinia sclerotiorum* and *S. minor* respectively (Min *et al*., 2009).

Cruz-Luna *et al.* (2021) studied Ag nanoparticles have been the most investigated nanoparticles due to their good antifungal activities, followed by Cu nanoparticles. It was also found that other metal nanoparticles have been investigated as antifungal agents, such as Se, Ni, Mg, Pd, and Fe, showing prominent results. Different synthesis methods have been used to produce these nanoparticles with different shapes and sizes, which have shown outstanding antifungal activities.

Abdelmalek *et al.* (2016) synthesized Silver nanoparticles with 10 ± 5 nm average diameter size and *In vitro* assay was carried out on Potato Dextrose Agar (PDA) media treated with 50, 100 and 150ppm of silver nanoparticles in addition to two control traditional fungicides products, namely Iprodione and Difenoconazole at @ 150 ppm concentrations. They found that Silver nanoparticle, 150 ppm, showed potent antifungal activity against the isolated fungi that will open the gate of new generation of less harmful fungicides replacing the cytotoxic dangerous currently used fungicides.

MATERIALS AND METHODS

Experimental Site:

All experiments were carried out in the laboratory of the Department of Plant Pathology at C.S.A. University of Agriculture and Technology in Kanpur during the period from 2021 to 2022.

Isolation and Purification of the Pathogen:

Samples of potato leaves that showed symptoms of blight were collected and kept in clean nylon bags (polyethylene) and transferred to the laboratory of the Plant Pathology Department. It was washed with distilled water to remove dust, a small piece of infected leaf from border of sporulating lesion along with some healthy green tissue was cut and

dipped in mercuric chloride solution (0.1%) for 30 seconds followed by rinse in sterilized distilled water thrice and dried offwith sterilized filter paper. The small pieces were then placed on tomato extractbased media which was previously pour in sterilizedPetri plates. The plates were then incubated at 18 ± 1^0C. The Petri plates were observed daily to find out thepresence of mycelium around the leave bits. As soon as the mycelia growth isnoticesaroundthebits, thepathogenwaspurifiedbyhyphal tipculturemethod.

The white mycelial bits of *P. infestans* was removed from the margin offungal colony and then transferred to another Petri-plate which was previouslypoured with sterilized tomato extract based medium. After purification, the pureculture of *P. infestans* was transferred on slant medium and incubated at 15-18^0Cin darkness till full growth. The culture was then transferred into the incubator at10-12^0Cforfurtheruse. Additionally, the pathogenicity of *P. infestans* was confirmed by conducting Koch's postulates on healthy potato plants.

Identification of the pathogen

The isolated pathogen was identified on the basis of its morphological, cultural characters and pathogenic behaviour towards the host. The mycelium is characterized by the absence of cross walls, along with both asexual and sexual reproduction occurs. The sporangiophores and sporangia emerge at asexual reproduction phase. The sporangia are lemon shape, measurement of 21-38µm×12-23µm. Sporangia develop at the end of these sporangiophores.

Maintenance of the culture

After confirmation of isolated pathogen as *P. infestans*, the pure culture was transferred on media slant and maintain in the BOD at 10-12°C for further study.

Effect of different nano-fungicides on radial mycelia growth of *Phytopthora infestans.*

The four nano- fungicides *viz*. Agritec, Silver, Selenium and Nickel at various concentrations (50, 75, and 100 ppm) were screened against the pathogen under laboratory conditions to find out their relative efficacy in inhibiting the growth of the pathogen with the help of the poisoned food technique (Schmitz, 1930). First of all, a stock solution of 1000 ppm of each nano-fungicide was made. The desired concentrations were obtained by adding the appropriate amount of stock solution of nano fungicides to tomato dextrose agar (TDA) medium in a separate flask and thoroughly mixed them by shaking prior to pouring in sterilized Petri plates. The medium was allowed to solidify and then 5 mm bits of fungal culture from seven days old culture of *P. infestans* were placed at the centre of Petri plates. The fungal disc was reversed so that the pathogen could come in direct contact with the medium. Three replications were kept for each treatment. The inoculated Petri plates were sealed with paraffin wax strips and incubated at 18±2°C in B.O.D. incubator. One set of control was maintained in which the medium was not mixed with any fungicide but simply

inoculated with the pathogen. The data on radial growth of the fungal colony was measured in mm after seven days. The percent inhibition over control was calculated by the following formula:

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

Where:

I = Percentage inhibition of mycelium
C = Colony diameter (mm) in the control
T = Colony diameter (mm) in the treatment

Treatments details: T1-Agritec nano-fungicide @ 50 ppm; T2-Agritec nano-fungicide @ 75 ppm; T3-Agritec nano-fungicide @ 100 ppm; T4- Selenium nano-fungicide @ 50 ppm; T5-Selenium nano-fungicide @ 75 ppm; T6- Seleniu mnano-fungicide @ 100 ppm; T7- Silver nano-fungicide @ 50 ppm; T8- Silver nano-fungicide @ 75 ppm; T9- Silver nano-fungicide @ 100 ppm; T10- Nickel nano-fungicide @ 50 ppm; T11- Nickel nano-fungicide @ 75 ppm; T12- Nickel nano-fungicide @ 100 ppm; T13-Control.

RESULTS AND DISCUSSION

The activity of different nano fungicides likes Agritec nano-fungicides, Silver nano-fungicides, Selenium nano-fungicides and Nickel nano-fungicides at different concentrations (50,75 and 100ppm) against *P. infestans* was determined *in vitro* through Poison Food Technique. The data present in Table 1 and Fig. 1showed that among all the nano-fungicides, Silver nano fungicide at 100 ppm was significantly superior over others in inhibiting mycelial growth of *P. infestans* representing the value 10.68, 13.10, 15.89, 18.38, 20.13 and 24.32 mm at 2, 3, 4, 5, 6 and 7 days after inoculation, respectively against 21.89, 33.85, 43.20, 58.50, 74.90 and 88.30 mm in case of control which was 72.45 percent inhibition over control. The Selenium nano fungicide at 100 ppm was showing 11.19, 14.02, 17.70, 20.80, 4.59 and 27.54mm radial growth of mycelium at 2, 3, 4, 5, 6 and 7 days after inoculation, respectively, representing second minimum radial mycelial growth among all the treatments. The maximum radial growth of mycelium was recorded in case of Nickel Nano fungicide at 50 ppm which is 56.17 per cent inhibition over control. From the table, it is cleared that all the tested nano fungicides were statistically significant in reducing the mycelial growth of pathogen as compared with control up to 7 days. Similar observation was reported previously by several workers (Abdel-Hafez *et al.,* 2016; Lamsal *et al.,* 2011; Thirumurugan *et al.,* 2011; Mohamed, 2015) found that silver nano fungicide effective against plant disease causing pathogens *i.e., Alternaria solan,* pepper anthracnose, *Rhizoctonia solani* and *Sclerotinia sclerotiorum*. Park *et al.* (2006) also studied the effective concentration of nanosized silica-silver on suppression of growth of many fungi and found that, *Pythium ultimum, Magnaporthe grisea, Colletotrichum gloeosporioides, Botrytis cinererea* and, *Rhizoctonia solani*, showed 100% growth inhibition at 10 ppm of the nanosized silica-silver. Silver nanoparticles with 10 ± 5 nm average diameter and *in vitro* assay was carried out with 50, 100 and 150 ppm of silver nanoparticles. Results revealed that Silver nanoparticle, 150 ppm, showed potent antifungal activity against the *Alternaria alternate, Alternaria citri* (Abdelmalek, 2016). Kim *et al.* (2012) conducted experiment to quantify the antifungal

effects of silver nanoparticles (AgNPs) against various plant pathogenic fungi. They used 10, 25, 50, and 100 ppm concentrations of AgNPs. Eighteen different plant pathogenic fungi were treated with these AgNPs on potato dextrose agar (PDA), malt extract agar, and corn meal agar plates. The results indicated that AgNPs possess antifungal properties against these plant pathogens at various levels.

Table 1: Effect of different concentration of nano-fungicides on radial mycelial growth (mm) of *Phytophthora infestans*

Comment [a1]: Add Statistical mean values (Tukey mean test)

Treatment	Concentration (ppm)	Radial mycelial growth (mm)						Percent Inhibition over control
		2 DAI	3DAI	4DAI	5DAI	6DAI	7DAI	
Agritecnano-fungicide	**50**	16.40	21.42	26.56	30.56	33.59	37.89	57.12
	75	13.90	20.25	23.35	25.35	27.25	31.53	64.29
	100	12.18	16.19	18.75	21.76	24.23	26.30	70.21
Selenium nano-fungicide	**50**	15.30	20.15	23.56	27.90	30.70	34.40	61.04
	75	13.24	16.89	19.13	23.62	26.67	30.69	65.24
	100	11.19	14.02	17.70	20.80	24.59	27.54	68.77
Silver nano-fungicide	**50**	13.55	16.20	19.70	23.75	27.60	31.50	64.32
	75	12.12	15.30	18.03	20.07	24.22	28.80	67.38
	100	10.68	13.10	15.89	18.38	20.13	24.32	72.45
Nickel Nano-fungicide	**50**	17.26	21.05	27.75	31.80	34.56	38.70	56.17
	75	15.45	19.87	24.75	28.13	31.51	35.69	59.58
	100	14.70	17.69	21.75	25.55	28.69	31.21	64.65
Control		21..89	33.85	43.20	58.50	74.90	88.30	-
CD		2.30	2.64	3.29	4.44	4.65	4.97	
SE (m)±		0.78	0.90	1.12	1.51	1.72	1.86	

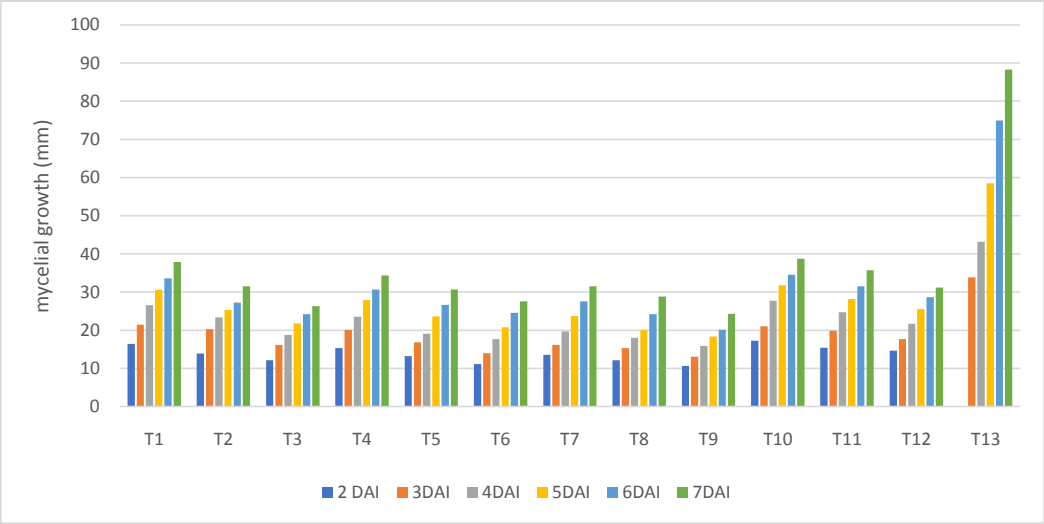

Fig. 1: Effect of different concentration of nano-fungicides on radial mycelial growth (mm) of *Phytophthora infestans*

Comment [a2]: Error bars should be added

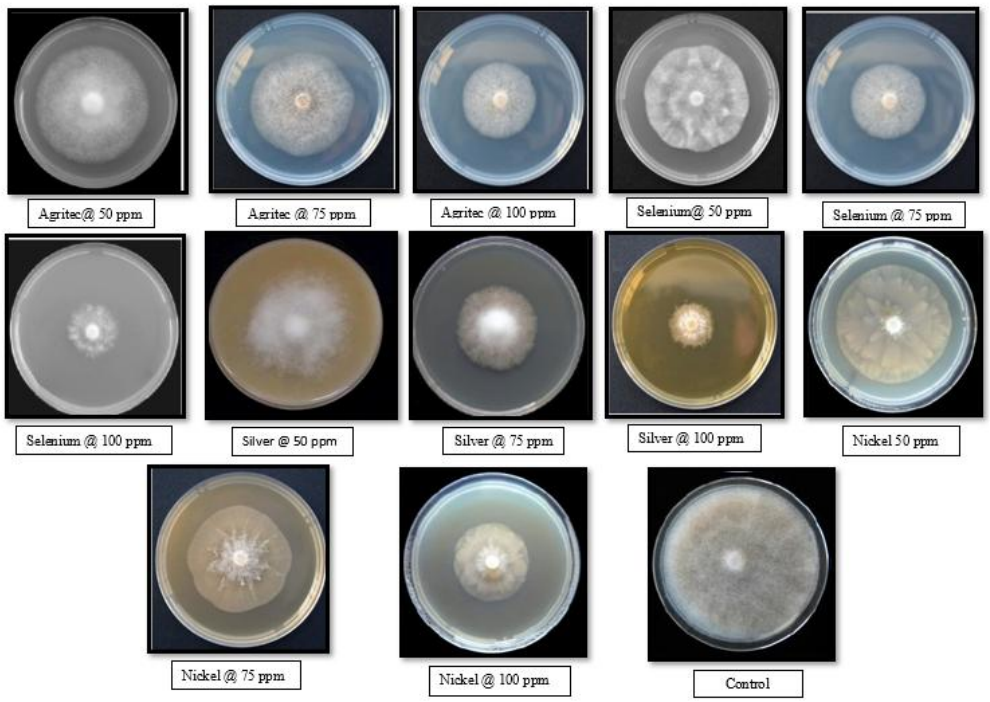

Plate 1: Effect of different concentration of nano fungicides on radial mycelial growth (mm) of *Phytophthora infestans*

Comment [a3]: Include this in figures

CONCLUSION

The antifungal activity of different nano fungicides was evaluated *in vitro* through poison food technique. The experimental finding showed that radial growth of mycelium of *P. infestans* was inhibited by nano fungicides over control. At 100 ppm, the minimum radial growth of mycelium was found in Silver nano-fungicides treatment as 10.68, 13.10, 15.89, 18.38, 20.13 and 24.32 mm over control against 21.89, 33.85 43.20, 58.50, 74.90 and 88.30 mm at 2, 3, 4, 5, 6 and 7 days after inoculation respectively. Similarly, 50, 75 and 100 ppm the minimum radial growth of mycelium was found in Silver nano-fungicides treatment. Among the different concentrations 100 ppm was found most effective than 50 and 75 ppm.

REFERENCES

Abdelmalek GAM, Salaheldin TA (2016) Silver Nanoparticles as a Potent Fungicide for Citrus Phytopathogenic Fungi. J Nanomed Res 3(5): 00065. DOI: 10.15406/jnmr.2016.03.00065.

Ashok Somalraju, Jason L. Mccallum, David Main, Rick D. Peters & Bourlaye Fofana (2021): Foliar selenium application reduces late blight severity and incidence in potato and acts as a pathogen growth inhibitor and elicitor of induced plant defence, Canadian Journal of Plant Pathology, DOI: 10.1080/07060661.2021.1954093

Ashraf, A., Rauf, C.A., Abbas, M.F. and Rehman, R. (2012). Isolation and identification of Verticillium dahliae causes wilt on potato in Pakistan. Pakistan Journal of Phytopathology, 24(2): 112-116.

Chadha, K.L. (2009). Potato. Handbook of Horticulture. ICAR, New Delhi, pp. 494-498.

Chowdappa, P., Kumar, N.B.J., Madhura, S., Kumar, M.S.P., Myers, K.L., Fry, W.E., Squires, J.N. and Cooke, D.E.L. (2013). Emergence of 13_A2 blue lineage of Phytophthora infestans was responsible for severe outbreaks of late blight on tomato in south west India. J. Phytopathol., 161: 49-58.

Cruz-Luna, Aida R., Heriberto Cruz-Martínez, Alfonso Vásquez-López, and Dora I. Medina. 2021. "Metal Nanoparticles as Novel Antifungal Agents for Sustainable Agriculture: Current Advances and Future Directions" *Journal of Fungi* 7, no. 12: 1033. https://doi.org/10.3390/jof7121033.

Dutt, S. (2008). Potato food: an antidote to hunger. Indian Horticulture, 53:2.

Elamawi RM, Al-Harbi RE, Hendi AA.Biosynthesis and characterization of silver nanoparticles using Trichoderma longibrachiatum and their effect on phytopathogenic fungi. Egyptian Journal of Biological Pest Control. 018;28(1):28.

Fry, W. E., Goodwin, S.B., Dyer, A.T., Matuszak, J.M., Drenth, A., Tooley, P.W., Sujkowski, L.S. Koh, Y.J., Cohen, B.A., Spielman, L.J., Deahl, K.L., Inglis D.A. and Sandlan, K. P., (1993). Historical and recent migrations of

Phytophthora infestans: Chronology, pathways, and implications. Plant Dis., 77: 653-661.

Fry, W.E. and Goodwin S.B. (1997a). Resurgence of the Irish Potato Famine Fungus. Bioscience, 47: 363-371.

Garrett, K.A. and Dendy, S.P. (2001). Cultural Practices in Potato Late Blight Management. In: Complementing Resistance to Late Blight in the Andes, February 13-16, Cochabamba.

Ghosh, B.K. (2010). Determinants of farm mechanization in modern agriculture: A case study of Burdwan districts of west Bengal. International Journal of Agricultural Research, 5(12): 1107-1115.

Gul, Z., Khan, A.A., Khan, A.U.R. and Khan, Z.U. (2013). Incidence of potato viruses in different districts of Khyber Pakhtunkhawa, Pakistan. International Journal of Phytopathology, 2(1): 32-36.

Hardy BB, Trognit Z, Forbes GA (1995) Late blight breeding at CIP. Circular(Lima-Peru) 21: 2-5.

Harmendez, C.J., Andrew, R. and Ferre, Y.B.J. (2005). Eval-uation of foliage resistance in potato to Phytophthora infestans based on international field network. Plant Pathol., 88: 8-16.

Haverkort, A.J., Struik, P.C., Visser, R.G.F., Jacobsen, E. (2009). Applied Biotechnology to Combat Late Blight in Potato Caused by Phytophthora Infestans. Potato Res., 52: 249-264.

Henfling, J.W. 1987. Late blight of potato. In: CIP, eds. Phytophthora Technical Bulletin, 2nd edn. Lima, Peru: International Potato Centre (CIP).

Kumar, U. and Chandra, G. (2018). A brief review of potash management in potato (Solanum tuberosum L.). Journal of Pharmacognosy and Phytochemistry SP1, 7: 1718-1721.

Lal, M., Arora, R.K., Maheshwari, U., Rawal, S. and Yadav, S. (2016). Impact of late blight occurrence on potato productivity during 2013-14. Int. J. Agricult. Stat. Sci., 12 (1): 187-192.

Lamsal, K., Sang, W. K., Jin, J., Yun, K., Kyong, S and Youn, S. L. (2011). Application of Silver Nanoparticles for the Control of *Colletotrichum* Species *in vitro* and Pepper Anthracnose Disease in Field. *Mycobiology*, *39*(3): 194-199.

Min, J. S., Kim, K. S., Kim, S. W., Jung, J. H., Lamsal, K., Kim, S. B., Jung, M. and Lee, Y. S. 2009. Effects of colloidal silver nanoparticles on Sclerotium forming phytopathogenic fungi. Pl. Pathol. J. 25 4: 376-380.

Nair, r., varghese s. H., nair, b. G., maekawa, t., yoshida, y. And Kumar, d. S., 2010, Nanoparticulate material deliver to plants. Pl. Sci., 179:154-163.

Nazarov, P.A., Baleev, D.N., Ivanova, M.I., Sokolova, L.M. and Karakozova, M.V. (2020). Infectious plant diseases: etiology, current status, problems and prospects in plant protection. Acta Naturae, 12(3): 46.

Ouda, S. M. 2014. Antifungal activity of silver and copper nanoparticles on two plant pathogens, Alternaria alternate and botrytis cinerea. Res. J. Microbiol. 9(1): 34- 42.

Shekhawat, N.S. Singh, R.P., Deora, N.S., Kaur,G., Kotwal. R.C. and Choudhary, N. (2000). Micropropagation of plants of stressed ecosystems. In: Plant tissue culture and molecular biology: application and prospects. Srivastava, P.S. (ed). Narosa Publishing House, New Delhi, pp: 579-586.

Ulger, T.G., Songur, A.N., Çırak, O. and Çakıroğlu, F.P. (2018). Role of vegetables in human nutrition and disease prevention. Vegetables—Importance of Quality Vegetables to Human Health; IntechOpen: London, UK, 7-32.

Wichrowska, D. (2022). Antioxidant Capacity and Nutritional Value of Potato Tubers (Solanum tuberosum L.) as a Dependence of Growing Conditions and Long-Term Storage. Agriculture, 12(1): 21.

Wu YJiang, J. and Gui, C. (2012). Low genetic diversity of Phytophthora infestans population in potato in north China. African J. Biotech., 11(90): 15636-15642.