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IN VITRO EVALUATION OF SILICON SOURCES AGAINST LATE BLIGHT (*Phytophthora infestans*) OF TOMATO

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ABSTRACT
The antifungal activity of different concentration of silicon sources *viz*., OSAB, Silixol plus, Greensil plus, Greensil, Silicon power and Pro-sil in inhibiting the mycelial growth of *P. infestans* was investigated by using poison food technique. The results from *in vitro* study revealed that, all the silicon sources tested at different concentrations showed a wide range of inhibitory effect (1.11 to 90.37 %) over untreated control. However, OSAB recorded highest mean mycelial growth inhibition (51.58 %) and it was followed by Greensil plus (49.84 %), Silixol plus (47.75 %), Greensil (29.87 %), Pro-sil (21.39 %) and Silicon power (21.35 %). However, different sources of silicon were inhibited mycelial growth of *P. infestans* with increasing concentration in the medium. The silicon sources used at 4.0 ml or g L^{-1} of medium were found to be significantly superior over other lower concentrations in inhibiting the mycelia growth of *P. infestans.*

KEYWORDS: *Phytophthora infestans*, Silicon, OSAB, Silixol plus, Mycelial growth.

INTRODUCTION

Tomato (*Solanum lycopersicum* L.) belongs to family solanaceae and is one of the most important and widely grown vegetable crop in both tropics and subtropics in the world. In India, it is fully fledged in different states under diverse climatic conditions. It is affected by several fungal, bacterial, viral and virus like diseases, known to contribute consistently too yield loss of the crop. Silicon (Si) is the second richest element found on the surface of the earth's crust (Liang *et al.*, 2007). While silicon is not regarded as an essential nutrient for most of the plants, many studies have shown that Si treatment progress the growth and yield of various plants, mainly when they are subjected to both biotic and abiotic stresses. The beneficial results of Si have been observed in an ample variety of plant species. The beneficial effects of Si are typically expressed more clearly in Si-accumulating plants under various abiotic and biotic stress conditions. Silicon is effective in managing various pests and diseases caused by both fungi and bacteria in different plant species. Silicon also exerts alleviative effects on various abiotic stresses including salt stress, metal toxicity, drought stress, radiation damage, nutrient imbalance, high temperature, freezing and so on. These beneficial results are chiefly attributed to the high accumulation of silica on the tissue surface although other mechanisms have also been suggested (Ma, 2004). Although direct antifungal activity of silicates against plant pathogens has been studied by Maekawa *et al*. (2003) who reported that hyphal growth of rice blast fungus was very slow on agar plates containing soluble silicon (liquid potassium silicate). Kaiser *et al.* (2005) examined the effect of liquid potassium silicate against several types of phytopathogenic fungi and determined *in-vitro* dose-responses towards soluble potassium silicate (20.7 % silicon dioxide) for *Phytophthora cinnamomi, Phytophthora capsici, Sclerotinia sclerotiorum, S. rolfsii, Pythium F-group,Mucor pusillus, Drechslera* spp, *Fusarium oxysporum, F. solani, A. solani, C. coccodes, Verticillium fungicola, C. lunata* and *Stemphylium herbarum.* They observed 100 per cent inhibition of mycelial growth at 40 and 80 ml of soluble silicon (20.7 % silicon dioxide) per litre of agar media. Also observed complete inhibition of *C. coccodes, M. pusillus, S. rolfsii, S. sclerotiorum* and *P. cinnamomi* at 5, 10 and 20 ml of soluble silicon per litre of agar. Bi *et al.* (2006) reported that, 100 mM sodium silicate was completely inhibited mycelial growth of *A. alternata, F. semitectum and Trichothecium roseum* which cause *Alternaria*, *Fusarium* and pink rots of Hami melon. Bekker *et al.* (2006) studied the effect of liquid potassium silicate against several phytopathogenic fungi *viz*., *A. solani, C. gloeosporoides, C. lunata, F. solani, P. capsici, S. rolfsii* and observed 100 per cent inhibition of mycelial growth at 40 ml (pH 11.5) and 80 ml (pH 11.7) soluble potassium silicate per litre of agar. Rachniyom and Jaenaksorn (2008) showed that all tested soluble silicon significantly reduced mycelial growth and sporangial production of *Pythium aphanidermatum.* Li *et al.* (2009) reported that, *F. sulphureum* spore germination was inhibited by sodium silicate (Aldrich, 27% SiO_2) at different concentrations (0, 25, 50, 100, 200 mM), with greater inhibitory effects at higher concentrations. Sodium silicate at different concentrations also markedly inhibited *F. sulphureum* mycelial growth, with greater inhibitory effects at higher concentrations. They also observed, morphological changes in sodium silicate-treated hyphae

such as mycelium sparsity and asymmetry, hyphal swelling, curling and cupped shape by scanning electron microscopy. Ultrastructural alterations were also observed under transmission electron microscopy, including thickening of the hyphal cell walls, cell distortion, cavity or electron-dense material in hyphal cells. Shen *et al.* (2010) investigated the effect of potassium silicate on the growth of five soil borne phytopathogenic fungi *in vitro* and showed that the growth of the fungal isolates (*Rhizoctonia solani, Pestalotiopsis clavispora* and *F. oxysporum* f. sp. *fragariae*) was significantly ($P < 0.05$) inhibited on potassium silicate amended PDA plates. Fayadh and Aledani (2011) reported that silicon elements were completely inhibited the growth of the pathogenic fungus *R. solani,* at 30, 200, 500 ppm respectively. Khan *et al.* (2013) reported silicon in the form of sodium silicate was found to be effective in reducing mycelial growth of *Macrophomina phaseolina* and also observed gradual reduction of mycelia growth with increase concentration of silicon. With a view to better characterizing the role of silicon in tomato late blight disease control, *in-vitro* experiment was performed to determine the effect of different silicon sources treatment on mycelial growth of *P. infestans* which cause a devastating late blight disease on tomato.

MATERIALS & METHODS

The poison food technique (Shravelle, 1961) was followed to evaluate the antifungal activity of different sources of silicon (silicic acid, orthosilicic acid, potassium silicate, SiO_2, silicon power and pro-sil) in inhibiting the mycelial growth of *P. infestans*. The required quantity *viz*., 0.1, 0.25, 0.5, 1.0, 2.0, 3.0 and 4.0 ml or g of different sources of silicon were added separately into one litre of sterilized molten V-8 agar media. Later, 20 ml of the poisoned media was poured into sterilized petri plates. Mycelium discs of five mm diameter from seven days old culture of the fungi were cut out by sterile cork borer and one such disc was placed at the centre of each plate. Media without any silicon sources served as control. Three replications were maintained for each concentration. The plates incubated at 18 $\pm 1^0$C for ten days and radial growth measured when fungus attained maximum growth in control plates.

Per cent inhibition of mycelia will be calculated by using formula:

$$\text{Inhibition rate (\%)} = \frac{C - T}{C} \times 100$$

C = Radial growth of fungi in control condition
T = Radial growth of fungi in silicon treated condition

Silicon sources used and their composition

The details of silicon sources used in the present study were presented below.

Sl. No.	Product name	Composition	Manufactured and Marketed by
Liquid formulation			
1	OSAB	Silicic acid-0.8% KCL-1.2% H_3BO_3-0.8 % HCl-1.0 % Peg $_{400}$- 48 % Demi water- 47%	Rexil agro BV Jamal Fazal Chambers, First Floor, "B" Block No. 26, Greams Road, Chennai (TN) – 600 006
2	Silixol plus	Orthosilicic acid-0.6 % Plant booster-2.0% Stabilizer-40 % Solvent-QS	Privi life sciences Pvt. Ltd A-71, TTC Thane Belapur Road, Kopar Khairane, Navi Mumbai- 400 709
3	Greensil plus	Silica-24 % Potassium-10 % QS- 66 %	Vedant Agrotech Sr. No. 32, Savitri Warehousing, Katraj-Kondhwa-Hadapsar bye-pass, village-Pisoli, Tq- Haveli, Dist- Pune
Powder formulation			
4	Greensil	Silicon as SiO_2-50 % Aluminium as Al-20 % QS-30 %	Vedant Agrotech 221, Mahalaxmi market, 2nd floor, Marketyard, Pune-411 037
5	Power silicon	Silicon-80 % Amino acid-15 % QS-5 %	Modern agro industries Factory No. 1, Lohiya compound, Survey No. 497, Naigaon Road, At. Post- Shinde MIDC, Tq and Dist- Nashik- 422 101
6	Pro-sil	Silicon Compound-60 %	Poorva Chemtech Pvt. Ltd. A-24 to 29 Samarth Co-Op, Industrial estate, Mukhed Road, Pimpalgoan (B) 422 209, Tq- Niphad, Dist- Nashik

The data obtained from this study were subjected to statistical analysis as per the procedure given by Sundaraj *et al.* (1972). Factorial complete randomized design used for to study and compare the treatments effects.

RESULTS & DISCUSSION

All the silicon sources tested at different concentrations by poison food technique showed a wide range of inhibitory effect (1.11 to 90.37 %) on *P. infestans* over untreated

control. Further, the per cent mycelial inhibition of *P. infestans* was found increased with increase in the concentration of silicon sources in the medium (Table 1, Fig. 1 and Plate 1).

Among the silicon sources tested at concentration of 4.0 ml or g L^{-1}, Silixol plus showed significantly higher inhibition of mycelial growth of *P. infestans* (90.37 %) followed by OSAB (89.81 %) and Greensil plus (89.63 %) and found on par with each other and Power silicon was found to be least effective (44.63 %) among the tested silicon sources. However, OSAB recorded highest mean mycelial growth inhibition (51.58 %) followed by Greensil plus (49.84 %), Silixol plus (47.75 %), Greensil (29.87 %), Pro-sil (21.39 %) and Power silicon (21.35 %). However, different sources of silicon were inhibited mycelial growth of *P. infestans* with increased concentration in the medium. However, silicon sources used at 4.0 ml or 4 g per litre of medium were found to be superior over other lower concentrations in inhibiting the mycelia growth of *P. infestans.*

TABLE 1: Antifungal activity of silicon sources against *Phytophthora infestans* under *in vitro* condition

Silicon sources	Per cent inhibition							Per cent mean inhibition
	Concentration (ml or g L^{-1})							
	0.1	0.25	0.5	1.0	2.0	3.0	4.0	
OSAB	10.74	16.67	26.85	66.48	72.59	77.96	89.81	51.58
Silixol plus	7.59	10.74	27.41	55.19	65.93	77.04	90.37	47.75
Greensil plus	11.67	16.11	26.48	57.59	70.56	76.85	89.63	49.84
Greensil	3.52	8.70	15.56	30.56	45.56	49.63	55.56	29.87
Power silicon	1.11	1.85	8.70	15.37	24.07	34.44	44.63	21.35
Pro-sil	1.48	6.11	10.37	21.67	28.52	35.19	46.11	21.39
Control	0.00	0.00	0.00	0.00	0.00	0.00	0.00	-
	Silicon Sources (S)				Concentrations (C)			S x C
SEm ±	0.02				0.02			0.05
CD at 5 %	0.05				0.06			0.15

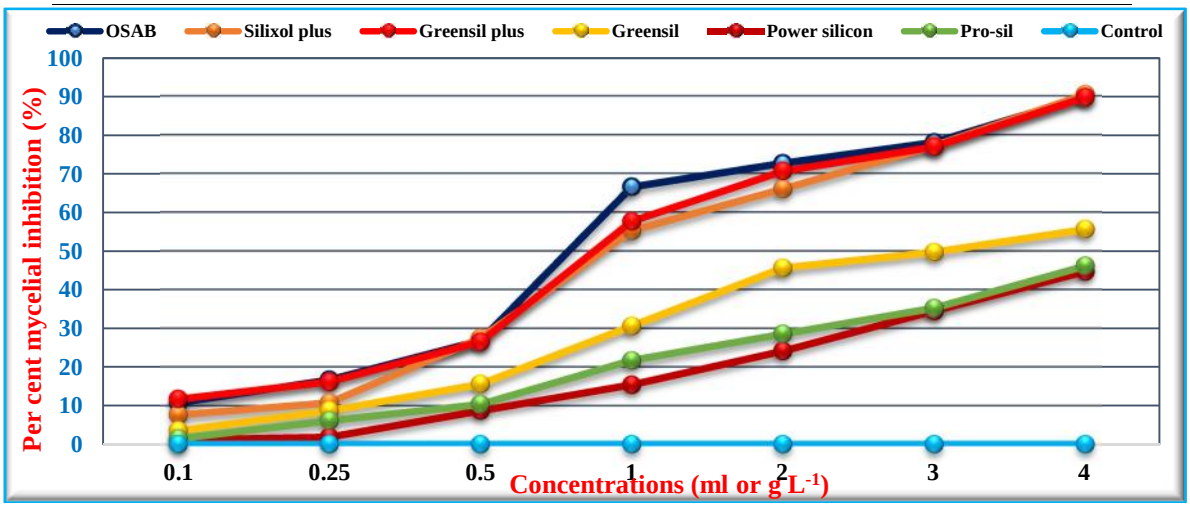

FIGURE 1: Antifungal activity of silicon sources against *Phytophthora infestans* under *in vitro* condition

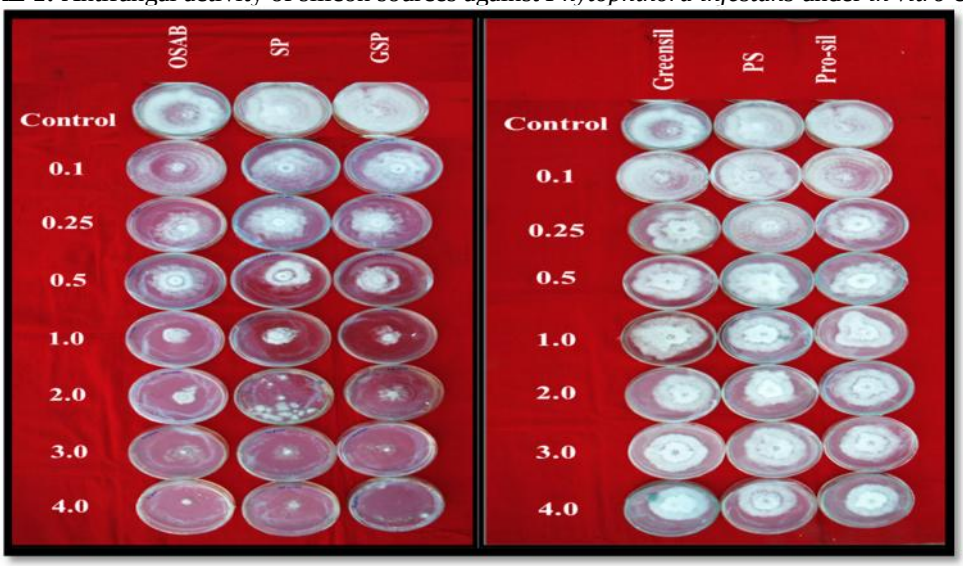

PLATE 1: Inhibitory effect of silicon sources against *Phytophthora infestans* under *in vitro* condition

* **SP**: Silixol plus **GSP**: Greensil plus **PS**: Power silicon

In the present study, silicon sources tested at different concentrations directly inhibited the *in vitro* mycelial growth of *P. infestans*, and these results showed that silicon sources had fungitoxic activity against *P. infestans.* These result are in line with the observations of Bi *et al.* (2006) who found complete inhibition of the mycelial growth of *Alternaria alternata, Fusarium semitectum* and *Trichothecium roseum* with 100-mM sodium silicate. Postharvest pathogens *Pencillium expansum, Monilinia fructicola* (Qin and Tian, 2005), *Colletotrichum* spp. (Biggs, 1999), *Botryosphaeria dothidea* (Biggs, 2004) mycelia growth was inhibited by different silicon sources tested under *in vitro.* Silicon amended PDA @ 0.6 per cent or more completely inhibited the growth of *P. expansum* (Ebrahimi *et al.*, 2012). Li *et al.* (2009) also observed morphological changes in sodium silicate-treated hyphae such as mycelium sparsity and asymmetry, hyphal swelling, curling and cupped shape by scanning electron microscopy. Ultrastructural alterations *viz*., thickening of the hyphal cell walls, cell distortion, cavity or electron-dense material in hyphal cells were also observed under transmission electron microscopy. These findings strongly suggest that silicon sources have direct fungitoxic activity against the pathogen.

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REFERENCES

Bekker, T.F., Kaiser, C., Van-Der-Merwe, R., Labuschagne, N., (2006) *In-vitro* inhibition of mycelial growth of several phytopathogenic fungi by soluble potassium silicate. *South African J. Plant Soil.*, **23**: 169-172.

Bi, Y., Tian, S.P., Guo, Y.R., Ge, Y.H. and Qin, G.N., (2006) Sodium silicate reduces postharvest decay on *Hami melons*: Induced resistance and fungistatic effects. *Plant Dis.*, **90**: 279-283.

Biggs, A.R. (1999) Effects of calcium salts on apple bitter rot caused by two *Colletotrichum* spp. *Plant Dis.*, **83**:1001-1005.

Biggs, A.R. (2004) Effect of inoculum concentration and calcium salts on infection of apple fruit by *Botryosphaeria dothidea*. *Plant Dis.*, **88**:147-51.

Fayadh, M.A. and Aledani, M.A. (2011) Effect of some microelements and biological control agents in control of tomato seedling damping-off caused by *Rhizoctonia solani* Kuhn. *Basra. J. Agric. Sci.*, **24**(1): 53-69.

Kaiser, C., Merwe, V., Bekker, T.F. and Labuschagne, N., (2005) *In-vitro* inhibition of mycelial growth of several phytopathogenic fungi, including *Phytophthora cinnamomi* by soluble silicon. *South African Avocado Growers' Association Yearbook*, **28**: 70-74.

Khan, M.O., Azizuddin and Shahzad, S. (2013) Use of silicon in inhibiting the growth of *Macrophomina phaseolina. Pak. J. Bot.*, **45**(4): 1469-1472.

Li, Y.C., Bi, Y., Ge, Y.H., Sun, X.J. and Wang, Y. (2009) Antifungal activity of sodium silicate on *Fusarium sulphureum* and its effect on dry rot of potato tubers. *J. Food Sci.*, **74**(5): 213-217.

Liang, Y., Sun, W., Zhu, Y.G. and Christie P. (2007) Mechanisms of silicon-mediated alleviation of abiotic stresses in higher plants: A review. *Environ. Poll*., **147**: 422-428.

Ma, J.F. (2004) Role of silicon in enhancing the resistance of plants to biotic and abiotic stresses. *Soil Sci. Plant Nutri.*, **50**(1): 11-18.

Maekawa, K., Watanabe, K., Kanto, T., Aino, M. and Saigusa, M. (2003) Effect of soluble silicic acid on suppression of rice leaf blast. *Japanese J. Soil Sci. Plant Nutri.*, **74**: 293-299.

Qin, G.Z. and Tian, S.P. (2005) Enhancement of biocontrol activity of *Cryptococcus laurentii* by silicon and the possible mechanisms involved. *Phytopathology*, **95**: 69-75.

Rachniyom, H. & Jaenaksorn, T. (2008) Effect of soluble silicon and *Trichoderma harzianum* on the *in vitro* growth of *Pythium aphanidermatum. J. Agri. Tech.*, **4**(2): 57-71.

Shen, G.H., Xue, Q.H., Tang, M., Chen, Q., Wang, L.N. and Duan, C.M. (2010) Inhibitory effects of potassium silicate on five soil-borne phytopathogenic fungi *in vitro*. *J. Plant Dis. Prot.*, **117**(4): 180-184.

Sunderaraj, H., Nagaraj, S., Venkataramu, M.N. and Jagannath, M.K. (1972) *Design and analysis of field experiments.* UAS, Misc. Series No. 22. *Univ. Agri. Sci., Bangalore*, India, 255pp.