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Effect of Soil Amendments with Different Chemical Inducers on the Pathogenicity of *Ralstonia solanacearum* to Tomato and Potato Plants

Hassan M. Emara[1*] • Safwat A. Elhadad[2] • Mahmoud A. Swelim[1] • Kamel M. Ata[2]

[1] Botany Department, Faculty of Science, Benha University, Benha, Egypt
[2] Potato Brown Rot Project, Agriculture Research Center, Dokki, Egypt

Corresponding author: * **hassanmemara@yahoo.com**

ABSTRACT

β-amino butyric acid (β-ABA), salicylic acid (SA), calcium oxide (CaO) + urea play important roles in inducing and accumulating phytoalexins in plants. Soil amended with these inducers influenced the presence of *Ralstonia solanacearum* population density in the soil rhizosphere of both tomato (*Lycopersicon esculentum* cv. 'Pinto') and potato (*Solanum tuberosum* cv. 'Spunta') plants. This led to a reduction in the percentage of infection and disease severity compared to the control treatment. Different concentrations of β-ABA and SA were effective in reducing the pathogenicity of *R. solanacearum* to tomato and potato plants as well as decreasing the population of bacteria invading their roots. Also, soil amended with CaO + urea influenced the presence of *R. solanacearum* population density in the soil rhizosphere of both tomato and potato plants.

Keywords: β-amino butyric acid, calcium oxide, potato, *Ralstonia solanacearum*, salicylic acid, tomato, urea
Abbreviations: β-ABA, β-amino butyric acid; **BW**, bacterial wilt; **CaO**, calcium oxide; **SA**, salicylic acid

INTRODUCTION

Tomatoes and potatoes are the most important crops in fresh-market production worldwide (Messiha *et al*. 2007). However, their yield and quality are threatened by bacterial wilt (BW) for which effective control measures are limited. BW is caused by *Ralstonia solanacearum* and is probably the most important bacterial disease of plants in different geographical zones worldwide (Elphinstone 2005). BW is a systemic disease that cannot be effectively managed with bactericides. Furthermore, it is also difficult to eliminate from fields since the pathogen persists in a wide range of crop and weed hosts and is associated with tomato, potato, and other Solanaceous hosts (Messiha *et al*. 2007). In addition to the loss of plants and yield, another serious impact of BW in Egypt (Messiha *et al*. 2009) and worldwide (Xue *et al*. 2009) is the abandonment of cultivation of potatoes and tomatoes on previously productive farms due to serious outbreaks of the disease (Li *et al*. 2004, 2005).

Application of synthetic bactericides is still the most effective management strategy on account of unavailability of suitable disease-resistant cultivars for commercial use. However, the use of potentially hazardous bactericides in agriculture has been the subject of growing concern because of their possible adverse effects on the environment and emergence of bactericide-resistant pathogens (Pal *et al*. 2001). Moreover, frequent application of bactericides leads to the development of tolerance in pathogens. Therefore, alternative control methods are deemed essential for the management of BW.

The use of soil amendments is a widespread means to control diseases caused by soil-borne plant pathogens (Huang and Huang 1993; Michel and Mew 1998). Islam and Toyota (2004) suggested that tomato BW was suppressed by certain amendments that promoted microbial activity due to their high contents of water-soluble C and N. Posas *et al*. (2007) used different sugars and amino acids as soil amendments to reduce the BW of tomato caused by *R. solanacearum* YU1Rif43. Michel *et al*. (1997) and Michel and Mew (1998) tested the efficacy of sugar, amino acids, urea and calcium oxide (CaO) as soil amendments in controlling tomato BW.

Okamura *et al*. (2002) reported that the density of *R. solanacearum* in soil was markedly reduced by adding simple organic compounds that are not utilized by the pathogens such as serine and glycine. In contrast, Matsuoka *et al.* (2005) found that the addition of glucose, utilized by the pathogen, to pumice culture increased its suppressiveness to tomato BW. The main objective of this study was to illustrate the efficiency of β-amino butyric acid (β-ABA), Salicylic acid (SA), and CaO + urea used as abiotic inducers for reducing the infection of *R. solanacearum* and its disease severity against potato and tomato plants.

MATERIALS AND METHODS

Tomato plants

An experiment was set up in a glasshouse (Potato Brown Rot Project, Central Administration of Plant Quarantine, Dokki, Egypt) at 25°C in the first growth stage then at 22°C till the end of the experiment with 75% relative humidity. Tomato (*Lycopersicon esculentum* cv. 'Pinto') seedlings were obtained from the Agricultural Research Center, Vegetables Research Institute, Giza, Egypt. Tomato seedlings (3-5 leaf stage) were grown in pots 15-cm in diameter containing 1.5 kg sandy soil with 3 replicates for each treatment. Soil was infested with *R. solanacearum*. After 48 h from culturing on nutrient agar media, a suspension of strain was prepared in sterile distilled water (SDW) and adjusted to optical density at 600 nm (OD_{600}) = 0.3 (approximately 6×10^8 CFU/ml). The inoculum: soil ratio was 1: 10 (v/v). Soil and inoculum was mixed thoroughly, placed in polyethylene bags, and incubated at 30°C. The plants were irrigation daily.

Different concentrations of β–ABA (0, 20, 40, 80 and 100

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μg/l), SA (0, 3, 4, 5, 6, 7 and 8 mM), and CaO + urea (0, and 20 g CaO + 8.7 g urea/l) were prepared and used for irrigating the tested plants. The control plants were irrigated by SDW without the tested inducers (β-ABA, SA, or CaO + urea). Wilting of tomato plants was recorded after 5 and 10 days (the time needed for symptoms appearing) as follows:

% infection = (No. of infected plants/total No. of seedlings) × 100

Disease severity was recorded using the scale adapted by Kempe and Sequeira (1983): 0 = no symptoms, 1 = 1-10% of the foliage wilted, 2 = 11-30% of the foliage wilted, 3 = 31-60% of the foliage wilted, 4 = > 60% but < 100% of the foliage wilted, and 5 = all leaves wilted.

About 10 g of tomato root tissue, which the pathogen can penetrate, colonize and concentrate, was cut. Surfaces were sterilized by 70-90% ethyl alcohol, then gently-flamed for few seconds. The sterilized cut roots were macerated with a mortar and pestle in 0.05 M phosphate buffer (4.26 g Na_2HPO_4 and 2.72 g KH_2PO_4 in 1.0 L SDW; pH = 7.2) and were shaken for 30 min at 15°C and at 100 rpm (Innova 4330 Refrigerated Incubator Shaker, USA). Thereafter, root extracts were left to settle and 1.0 ml of the supernatant was pipetted, serially diluted from the original concentration up to 10^{-6}, then cultured on SMSA media (Elphinstone *et al.* 1996) in Petri dishes using a sterilized glass spreader, and incubated at 28°C for 48 h. Then, the total number of single typical pure colonies of *R. solanacearum* was recorded per g of root according to the following equation:

Total count of bacterial colonies = colony count/sample weight × sample dilution.

Potato plants

Another experiment was designed and conducted with the same conditions as previously mentioned for tomato, except that potato plants (*Solanum tuberosum*) commercial cultivar ('Spunta') were used (15-20-day old seedlings). Symptoms of potato wilt disease were recorded after 7 and 14 days, the time needed for symptoms appearing.

RESULTS

Table 1 represents the effect of different concentrations of β-ABA on the pathogenicity of *R. solanacearum* to tomato. Both percentages of infection and disease severity as well as the bacterial count decreased as the concentration of β-ABA increased up to 20 μg/ml; the infection percentage reduced from 50 to 20%, and disease severity from 3 to 2 after 5 and 10 days from soil infestation, respectively. Increasing the β-ABA concentration to 40 μg/mL reduced the percent of infection from 13.3% at 5 days to 6.67% infection after 10 days and disease severity from 2 to 1. However, 60, 80, and 100 μg β-ABA/ml protected tomato roots against infection by bacteria where infection and disease severity were zero at 10 days after soil infestation in the case of the water control. The lowest concentration able to completely protect tomato plants was 80 μg/mL. The bacterial count in tomato roots was also affected by increasing β-ABA concentration compared to the control.

Different concentrations of β-ABA decreased both percentage of infection and disease severity of *R. solanacearum* in potato plants and bacterial count of invading potato roots (**Table 2**). Disease severity and bacterial count reduced as the concentration of β-ABA increased up to 20 μg/mL; with the percentage infection reducing from 43.3 to 23.3% and disease severity from 3 to 2 after 7 and 14 days from soil infestation, respectively. Increasing the β-ABA concentration up to 40 μg/mL reduced the percentage of infection from 12.4 to 8.6% and disease severity from 2 to 1. However, 60, 80 and 100 μg/mL reduced the percentage of infection and disease severity to zero after 7 and 10 days from soil infestation, resembling the water control. The lowest concentration that was able to completely protect potato plants was 60 μg/mL. The bacterial count in tomato roots was also affected by increasing the β-ABA concentration compared to the control.

Tables 3 and **4** show that different concentrations of SA played an important role in reducing bacterial infection and disease severity percentages in tomato and potato plants as well as the bacterial count in tomato and potato roots. The potato plants showed different resistance against bacteria with different SA concentrations. Increasing the concentration of SA decreased the ability of bacteria to colonize inside potato roots (**Table 4**).

Tables 5 and **6** show that soil amended with CaO + urea influenced the presence of *R. solanacearum* population density in the soil rhizosphere of both tomato and potato plants, leading to the reduction of the infection percentage and disease severity compared to the control. The percentage of infection, disease severity, and bacterial count were recorded for both tomato and potato plants after wilting appeared on leaves (**Table 5**). A reduction in bacterial population in amended soil with CaO + urea was observed by increasing

Table 1 Effect of different concentrations of β-amino butyric acid (β-ABA) on pathogenicity of *R. solanacearum* pathogenicity to tomato plants after 5 and 10 days after soil infestation and count of bacteria in tomato roots on SMSA media.

β-ABA concentrations	**After 5 days from soil infestation**		**After 10 days from soil infestation**		**Count/g root × 10^2**
(μg/ml)	**% infection**	**Disease severity**	**% infection**	**Disease severity**	**After 10 days from soil infestation**
0	52	3	100	5	136.2×10^7
20	50	3	20	2	14.6
40	13.3	2	6.67	1	0.5
60	4.2	1	0	0	0
80	0	0	0	0	0
100	0	0	0	0	0
β-ABA control	0	0	0	0	0
Water control	0	0	0	0	0

Table 2 Effect of different concentrations of β-amino butyric acid (β-ABA) on *R. solanacearum* pathogenicity to potato plants after 7 and 14 days after soil infestation.

β-ABA concentration	**After 7 days from soil infestation**		**After 14 days from soil infestation**		**Count/g root × 10^2**
(μg/ml)	**% infection**	**Disease severity**	**% infection**	**Disease severity**	**after 14 days from soil infestation**
0	56.3	4	97.8	4	129.3×10^6
20	43.3	3	23.3	2	126.3
40	12.4	2	8.6	1	0.26
60	0	0	0	0	0
80	0	0	0	0	0
100	0	0	0	0	0
β-ABA control	0	0	0	0	0
Water control	0	0	0	0	0

Table 3 Effect of different concentrations of salicylic acid (SA) on the pathogenicity of *R. solanacearum* to tomato plants after 10 days of soil infestation.

SA concentration (mM)	% infection	Disease severity	Count CFU/g root × 10^2
0	92.4	4	48.3 × 10^6
3	44.2	3	6.5
4	26	2	0.6
5	18	2	0.02
6	8	1	0.005
7	2	1	0.002
8	0	0	0
SA control	0	0	0
Water control	0	0	0

Table 4 Effect of different concentrations of salicylic acid (SA) on the pathogenicity of *R. solanacearum* to potato plants after 14 days of soil infestation.

SA concentration (mM)	% infection	Disease severity	Count * (CFU/g root × 10^2)
0	100	5	60 × 10^6
5	29.5	2	590
7	9	1	21.2
9	0	0	0
SA control	0	0	0
Water control	0	0	0

the time. However, soil amended by CaO + urea showed lower bacterial count than that infested without soil amendments (**Table 6**).

DISCUSSION

These results show that β-ABA played an effective role in reducing pathogenicity of *R. solanacearum* to tomato and potato plants as well as the population of bacteria invading tomato and potato roots. This effect is because the application of β-ABA in soil leads to the induction of both physical and biochemical defense mechanisms in both plants. β-ABA induction causes the accumulation of pathogenesis-related (PR) protein before challenge (Hwang *et al*. 1997; Ovadia 2001). In similar studies, β-ABA played an important role in accumulating phytoalexins (Hwang *et al*. 1997), which is considered as specific mechanism of host defense. β-ABA is thought to have the ability to break or cut chemical signals between tomato and potato roots by changing the chemical composition of root exudates secreted by both plants and increasing the amount of phenolic compounds secreted in plant root exudates (Bais *et al*. 2004). A single foliar spray on tomato with an effective dose of β-ABA induces a variety of physical and biochemical defense mechanisms in plants (Baysal *et al*. 2007). Pathogen–specific defense mechanisms induced by β-ABA may depend not only on the plant species but also on the elicitor(s) released by the specific pathogen (Hwang *et al*. 1997).

In recent years, evidence has been accumulated, in *Arabidopsis* mutants, showing that β-ABA possesses a large spectrum of activity against many disease-causing organisms such as virus, bacteria, oomycetes, fungi, and nematodes (Jakab *et al.* 2001), as well as multiple forms of plant activation against disease (Siegrist *et al*. 2000; Ovadia 2001; Zimmerli *et al.* 2001, 2008). β-ABA induces the formation of pinpoint necrotic spots which were considered by some researchers to be involved in systemic acquired resistance (SAR) (Siegrist *et al.* 2000; Zimmerli *et al.* 2000, 2008). *Arabidopsis* plants treated with β-ABA showed callus formation and papillae after a challenged with *Peronospora parasitica*. In tomato challenged with *Phytophthora infestans*, callus (Jeun *et al*. 2000; Jeun and Buchenauer 2001) and both callus and lignin (Raviv 1994) were observed as a result of treating with β-ABA. It thus appears that a physical barrier(s) induced by β-ABA is pathosystem-specific. β-ABA is also effective when incorporated (as a powder) into the soil, injected into the stem, or applied as a solution to bare roots, cut stems, or cut leaves. The concentration of β-ABA required to achieve effective (~90%) resistance under controlled conditions depends on the host, the pathogen, and the mode of application. Normally, higher doses are required in foliar sprays (250-1000 μg/ml) than in soil drenches (20-100 μg/ml), probably because of a more efficient uptake through the roots. Higher concentrations (0.5-1% solutions) are needed in seed soaking (Cohen 1994, 1996; Shailasree *et al.* 2001).

In the present study, soil amended with SA influenced the presence of *R. solanacearum* population density in soil rhizosphere of both tomato and potato plants. This may be because SA played an effective role in the induction signals in the mechanism of SAR leading to the reduction of both the percentage of infection and disease severity in tomato and potato plants as well as the bacterial counts in their roots (Raskin 1992; Sticher *et al.* 1997). SA is initially proposed to bind to catalase and ascorbate peroxidase, which might lead to the formation of free radicals involved in lipid peroxidation, which can activate defense gene expression (Meraux 2001). SA and acetyl salicylic acid (aspirin) was considered as a candidate for exogenous applications as SAR activator because they are inexpensive and non-phytotoxic products (Lopez 1993).

SA is a key hormone of stress defense responses induced by biotrophic pathogens in plants (Durrant and Dong 2004; Loake and Grant 2007) involved in the activation of defense genes. Besides activating stress defenses, SA is

Table 5 Effect of soil amended by calcium oxide (CaO) + urea on *R. solanacearum* pathogenicity to tomato and potato plants after different periods from soil infestation.

Treatments	Tomato plants				Potato plants			
	After 5 days from soil infestation		After 10 days from soil infestation		After 7 days from soil infestation		After 14 days from soil infestation	
	% infection	Disease severity	% infection	Disease severity	% infection	Disease severity	% infection	Disease severity
Infested soil with CaO + urea	20	2	26	2	12	2	22	2
Infested soil without CaO + urea	94	4	96	4	100	5	100	5
CaO + urea control	0	0	0	0	0	0	0	0
Water control	0	0	0	0	0	0	0	0

Table 6 Count of *R. solanacearum* bacteria in soil amended by calcium oxide (CaO) + urea cultivated with potato plants after time course on SMSA media.

Treatments	Log_{10} Count of bacteria on SMSA media (CFU/g dry soil)						
	0 days	3 days	7 days	10 days	14 days	21 days	28 days
Infested soil with CaO + urea	10.4	8.01	7.1	5.9	5.2	3.4	2
CaO + urea control	0	0	0	0	0	0	0
Water control	0	0	0	0	0	0	0
Infested soil without CaO + urea	10.4	10.1	9.02	8.3	9.5	9.1	8.4

known to inhibit jasmonic acid and auxin-mediated responses enhancing the disease-resistance mechanism (Fujita *et al.* 2006; Ndamukong *et al.* 2007; Wang *et al.* 2007). These responses allow plants not only to survive pathogen infection, but also to acquire a long-lasting SAR responsible for the protection from further infections by a broad range of pathogens (Grant and Lamb 2006). SA is an important natural component in signal transduction pathways leading to the activation of defense responses and SAR of plant against viral, fungal, and bacterial pathogens (Alvarez 2000; Faravardeh and Rabbani 2006). SA plays a key role in mediating disease resistance in plants, where the endogenous SA concentration increases at the site of HR and acts as a signal transducer for activation of defense response (Delaney *et al.* 1994). Marfa *et al.* (2001) reported that induced resistance in plants can be triggered either by localized infection with pathogens or by treatment with certain chemicals such as SA and acetylsalicylic acid (ASA).

Höper and Alabouvette (1996) reported that soil suppressiveness can be due to soil physico-chemical characteristics such as texture, structure, pH, and Ca content. The obtained results herein showed that soil amended with CaO + urea influenced the presence of *R. solanacearum* population density in the soil rhizosphere of both tomato and potato plants. This amendment leads to the reduction of both percentage infection and disease severity compared to the control. Amended urea may be transformed to other nitrogenous compounds such as nitrite and ammonia that affect the occurrence of bacteria. The accumulation of nitrite against time leading to an increase in soil pH and nitrous acid, which considered the non-ionized form of nitrite was accumulated (Tsao and Oster 1981; Tenuta and Lazarovits 2002; Cohen *et al.* 2005). Reduction in bacterial population may be due to the accumulation of nitrite which is responsible for the decline of several soil-borne pathogens (Smiley *et al.* 1970; Walker 1971). Also, ammonia was reported to be accumulated (Gilpatrick 1969; Rush and Lyda 1982) and may play an important role in suppression process due to its toxicity. On the other hand, Persson and Olsson (2000) found a correlation between the degree of soil suppressiveness against root rot of vining pea, *Pisum sativum* L. and soil Ca, pH and vermiculite-smectite content of clay minerals. Elevated Ca levels have been associated with suppression of infection by a range of different pathogenic fungi (Heyman *et al.* 2007). Likewise, the increase of Ca uptake in tomato shoots was correlated with lower levels of disease severity of *R. solanacearum* (Yamazaki *et al.* 1996; Yamazaki 2001; Messiha *et al.* 2007).

REFERENCES

Alvarez ME (2000) Salicylic acid in the machinery of hypersensitive cell death and disease resistance. *Plant Molecular Biology* **44**, 429-442

Bais H, Park S, Weir T (2004) How plants communicate using the underground information superhighway. *Trends in Plant Science* **9**, 26-32

Baysal Ö, Gursoy YZ, Örnek H, Çetinel B, Teixeira da Silva JA (2007) Enhanced systemic resistance to bacterial speck disease caused by *Pseudomonas syringae* pv. *tomato* by DL-β-amino butyric acid (BABA) under salt stress. *Physiologia Plantarum* **129**, 493-506

Cohen Y (1994) Local and systemic control of *Phytophthora infestans* in tomato plants by DL-3-amino-n-butanoic acids. *Phytopathology* **84**, 55-59

Cohen Y (1996) Induced resistance against fungal diseases by aminobutyric acids. In: Lyr H, Russel PE, Sisler HD (Eds) *Modern Fungicides and Antifungal Compounds*, Intercept, Andover, pp 461-466

Cohen MF, Yamasaki H, Mazzola M (2005) *Brassica napus* seed meal soil amendment modifies microbial community structure, nitric oxide production and incidence of *Rhizoctonia* root rot. *Soil Biology and Biochemistry* **37**, 1215-1227

Delaney T, Uknes S, Vernooji B, Friedrich L, Weymann K, Negrotto D, Gaffney T, Gut-Rella M, Kessmann H, Ward E, Ryals J (1994) A central role of salicylic acid in plant disease resistance. *Science* **266**, 1247-1250

Durrant WE, Dong X (2004) Systemic acquired resistance. *Annul Review of Phytopathology* **42**, 185-209

Elphinstone JG (1996) Survival and possibilities for extinction of *Ps. Solanacearum* (Smith) in cool climates. *Potato Research* **39**, 403-410

Elphinstone JG (2005) The current bacterial wilt situation: a global overview. In: Allen C, Prior P, Hayward AC (Eds) *Bacterial Wilt Disease and the Ralstonia solanacearum Species Complex*, APS Press, St. Paul, pp 9-28

Faravardeh L, Rabbani A (2006) Studies on the superoxide anion production and peroxidase activity in potato leaf cell suspension exposed to salicylic acid. *Acta Physiologiae Plantarum* **28**, 237-243

Fujita M, Fujita Y, Noutoshi Y, Takahashi F, Narusaka Y, Yamaguchi-Shinozaki K, Shinozaki K (2006) Crosstalk between abiotic and biotic stress responses: a current view from the points of convergence in the stress signaling networks. *Current Opinion in Plant Biology* **9**, 436-442

Gilpatrick J (1969) Role of ammonia in the control of avocado rootrot with alfalfa meal soil amendment. *Phytopathology* **59**, 973-978

Grant M, Lamb C (2006) Systemic immunity. *Current Opinion in Plant Biology* **9**, 414-420

Hayward AC (1991) Biology and epidemiology of bacterial wilt caused by *Pseudomonas solanacearum*. *Annual Review of Phytopathology* **29**, 65-87

Heyman F, Lindahl B, Persson L, Wikström M, Stenlid J (2007) Calcium concentrations of soil affect suppressiveness against *Aphanomyces* root rot of pea. *Soil Biology and Biochemistry* **39**, 2222-2229

Höper H, Alabouvette C (1996) Importance of physical and chemical soil properties in the suppressiveness of soils to plant diseases. *European Journal of Soil Biology* **32**, 41-58

Huang H, Huang J (1993) Prospects for control of soilborne plant pathogens by soil amendment. *Current Topics of Botanical Research* **1**, 223-235

Hwang B, Sunwoo J, Kim Y, Kim B (1997) Accumulation of beta-1,3- glucanase and chitinase isoforms, and salicylic acid in the DL-beta-amino-n-butyric acid-induced resistance response of pepper stems to *Phytophthora capsici*. *Physiology of Molecular Plant Pathology* **51**, 305-322

Islam TMD, Toyota K (2004) Suppression of bacterial wilt of tomato by *Ralstonia solanacearum* incorporation of composts in soil and possible mechanisms. *Soil Microorganisms* **19**, 53-90

Jakab G, Cottier V, Toquin V, Rigoli G, Zimmerli L, Metraux J, Mauch-Mani B (2001) β-amino butyric acid-induced resistance in plants. *European Journal of Plant Pathology* **107**, 29-37

Jeun Y, Buchenauer H (2001) Infection structures and localization of the pathogenesis- related protein AP24 in leaves of tomato plants exhibiting systemic acquired resistance against *Phytophthora infestans* after pre-treatment with 3-aminobutyric acid or tobacco necrosis virus. *Journal of Phytopathology* **149**, 141-153

Jeun Y, Siegrist J, Buchenauer H (2000) Biochemical and cytological studies on mechanisms of systemically induced resistance to *Phytophthora infestans* in tomato plants. *Journal of Phytopathology* **148**, 129-140

Kempe J, Sequeira L (1983) Biological control of bacterial wilt of potatoes attempts to induce resistance by treating tubers with bacterial. *Plant Disease* **67**, 499

Li GC, Jin LP, Xie KY, Qu DY (2004) Advances in research on bacterial wilt of ginger in China. *Chinese Potato* **18**, 350-354

Liu M, Zhang M, Ji JC, Yin FQ, Zhang Y, Tu Y, Ye YJ (2005) Advances in research on bacterial wilt of ginger in China. *Chinese Agricultural Science Bulletin* **21**, 337-341

Loake G, Grant M (2007) Salicylic acid in plant defense – the players and protagonists. *Current Opinion in Plant Biology* **10**, 466-472

Lopez M (1993) Mecanismos de resistencia inducidos por compustos quimicos frente a la podredumbre blanda en tuberculos de patata. PhD thesis, Universidad Complutense de Madrid, Spain, 215 pp

Marfa M, Marfa J, Javier Z, Jesus L, Sanchez M, Ramona B (2001) effect of acetylsalicylic acid on soft rot produced by *Erwinia Carotovora* subsp. *Carotovora* in potato tubers under green house conditions. *Potato Research* **44**, 197-206

Matsuoka T, Toyota K, Sato K, Masuda K, Kuroda T (2005) Suppressive mechanisms of used pumice to bacterial wilt of tomato. *Soil Microorganisms* **59**, 15-20

Mertaux J, Ryals J, Wards E, Wyss-Benz M, Gaudin J, Rashdorf K, Schmid E, Blum W, Inverardi B (1990) Increase in salicylic acid at the onset of systemic acquired resistance in cucumber. *Science* **250**, 1004-1006

Messiha NAS, van Bruggen AHC, van Diepeningen AD, de Vos OJ, Termorshuizen AJ, Tjou-Tam-Sin NNA, Janse JD (2007) Potato brown rot incidence and severity under different management and amendment regimes in different soil types. *European Journal of Plant Pathology* **119**, 367-381

Messiha NAS, van Bruggen AHC, Franz E, Janse JD, Schoeman-Weerdesteijn ME, Termorshuizen AJ, van Diepeningen AD (2009) Effects of soil type, management type and soil amendments on the survival of the potato brown rot bacterium *Ralstonia solanacearum*. *Applied Soil Ecology* **43**, 206-215

Michel VV, Mew TW (1998) Effect of a soil amendment on the survival of *Ralstonia solanacearum* in different soils. *Phytopathology* **88**, 300-305

Michel VV, Wang J-F, Midmore DJ, Hartman GL (1997) The effects of intercropping and soil amendment with urea and calcium oxide on the incidence of bacterial wilt of tomato and survival of soilborne *Pseudomonas solanacearum* in Taiwan. *Plant Pathology* **46**, 600-610

Ndamukong I, Abdallat AA, Thurow C, Fode B, Zander M, Weigel R, Gatz C (2007) SA-inducible Arabidopsis glutaredoxin interacts with TGA factors and suppresses JA-responsive PDF1.2 transcription. *The Plant Journal* **50**, 128-139

Okamura Y, Murakami N, Yagi W (2002) Hyoutekibiseibutsu no yokuseihouhou. International patent No. WO99/44422

Ovadia A (2001) Mode of action of 3-amino butyric acid in systemic acquired resistance. PhD thesis, Bar-Ilan University, Ramat-Gan, Israel, total pp

Pal KK, Tilik KVBR, Saxena AK, Dey R, Singh CS (2001) Suppression of maize root disease caused by *Macrophomina phaseolina*, *Fusarium moniliforme* and *Fusarium graminearum* by plant growth promoting rhizobacteria. *Microbiological Research* **156**, 209-223

Persson L, Olsson S (2000) Abiotic characteristics of soils suppressive to *Aphanomyces* root rot. *Soil Biology and Biochemistry* **32**, 1141-1150

Posas MB, Toyota K, Islam TMD (2007) Inhibition of bacterial wilt of tomato caused by *Ralstonia solanacearum* by sugars and amino acids. *Microbes and Environments* **22**, 290-296

Raskin I (1992) Role of salicylic acid in plants. *Annual Review of Plant Pathology and Plant Molecular Biology* **43**, 439-463

Raviv A (1994) The mode of action of β-amino butyric acid in inducing resistance in tomato plants against late blight. PhD thesis, Bar-Ilan University, Ramat-Gan, Israel, total pp

Rush C, Lyda S (1982) Effects of anhydrous ammonia onmycelium and sclerotia of *Phymatotrichum omnivorum*. *Phytopathology* **72**, 1085-1089

Shailasree S, Sarosh B, Vasanthi N, Shetty H (2001) Seed treatment with β-aminobutyric acid protects *Pennisetum glaucum* systemically from *Sclerospora graminicola*. *Pest Management Science* **57**, 721-728

Siegrist J, Orober M, Buchenauer H (2000) Beta-aminobutyric acid-mediated enhancement of resistance in tobacco to tobacco mosaic virus depends on the accumulation of salicylic acid. *Physiology of Molecular Plant Pathology* **56**, 95-106

Smiley R, Cook R, Papendick R (1970) Anhydrous ammonia as a soil fungicide against *Fusarium* and fungicidal activity in the ammonia retention zone. *Phytopathology* **60**, 1227-1232

Sticher L, Mauch-Mani B, Mertaux P (1997) Systemic acquired resistance. *Annual Review of Plant Pathology* **35**, 235-270

Tenuta M, Lazarovits G (2002) Ammonia and nitrous acid from nitrogenous amendments kill microsclerotia of *Verticillium dahliae*. *Phytopathology* **92**, 255-264

Tsao P, Oster J (1981) Relation of ammonia and nitrous acid to suppression of *Phytophthora* in soils amended with nitrogenous organicsubstances. *Phytopathology* **71**, 53-59

Walker J (1971) Populations of *Pratylenchus penetrans* relative to decomposing nitrogenous soil amendments. *Journal of Nematology* **3**, 43-49

Wang D, Pajerowska-Mukhtar K, Culler AH, Dong X (2007) Salicylic acid inhibits pathogen growth in plants through repression of the auxin signaling pathway. *Current Biology* **17**, 1784-1790

Xue Q-Y, Chen Y, Li S-M, Chen L-F, Ding G-C, Guo D-W, Guo J-H (2009) Evaluation of the strains of *Acinetobacter* and *Enterobacter* as potential biocontrol agents against *Ralstonia* wilt of tomato. *Biological Control* **48**, 252-258

Yamazaki H (2001) Relation between resistance to bacterial wilt and calcium nutrition in tomato seedlings. *Japan Agricultural Research Quarterly* **35**, 163-169

Yamazaki H, Ishizuka O, Hoshina T (1996) Relationship between resistance to bacterial wilt and nutrient uptake in tomato seedlings. *Soil Science and Plant Nutrition* **42**, 203-208

Zimmerli L, Hou B-H, Tsai C-H, Jakab G, Mauch-Mani B, Sommerville S (2008) The xenobiotic beta-aminobutyric acid enhances *Arabidopsis* thermotolerance. *The Plant Journal* **53**, 144-156

Zimmerli L, Jakab C, Metraux J, Mauch-Mani B (2000) Potentiation of pathogen-specific defense mechanisms in *Arabidopsis* by β-aminobutyric acid. *Proceedings of the National Academy of Sciences USA* **97**, 12920-12925

Zimmerli L, Metraux J, Mauch-Mani B (2001) β-aminobutyric acid-induced protection of *Arabidopsis* against the necrotrophic fungus *Botrytis cinerea.* *Plant Physiology* **126**, 517-523