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Management of Late Blight with Alternative Products

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ABSTRACT

Controlling highly destructive plant diseases such as potato and tomato late blight, caused by the oomycete *Phytophthora infestans*, with non-fungicidal alternative products is a difficult but necessary task that needs to be accomplished. Given the rapid development of potato and tomato late blight epidemics, for many years control strategies relied solely upon the application of fungicides. There are many reports of alternative strategies for managing several plant diseases, however reports on late blight control with non-fungicidal products are recent and appeared in the last 20 years. The most commonly used strategy to control the disease has been the prevention of establishment of *P. infestans* in the host plant, mainly by using organisms capable of producing chemical compounds that inhibit spore germination. Nevertheless, the epidemiological characteristics of late blight makes the adoption of a "silver bullet approach" risky. To enhance the chances of success of alternative products, a combination of compounds and microorganisms with different modes of action should be employed beginning at the early stages of the host-pathogen interaction. Specifically, when effective options are available, one should consider the use of phylloplane and endophytic organisms combined with resistance induction mediated by plant growth promoting rhizobacteria. Late blight control with alternative products is likely to positively impact both conventional and organic production systems.

Keywords: biological control, disease, organic, *Phytophthora infestans*, potato, tomato
Abbreviations: PGPR, plant growth-promoting rhizobacteria

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INTRODUCTION

Late blight is a challenging plant disease to be tackled with alternative products. The major economic host crops – potato (*Solanum tuberosum* L.) and tomato (*S. lycopersicum* L.) – are considered the most important horticultural crops in the several countries and both are severely affected by the disease. The causal agent of late blight is an oomycete, *Phytophthora infestans* (Mont.) De Bary, which is well-known for its explosive development when environmental conditions are suitable and host plants susceptible to infection (Mizubuti and Fry 2006). Reports of complete field destruction due to late blight epidemics are relatively common in the literature and estimated worldwide economic losses due to the disease vary from 3 to 5 billion dollars annually (Raman *et al*. 2000; Guenthner *et al*. 2001; Judelson and Blanco 2005; Haldar *et al*. 2006).

What makes *P. infestans* such an aggressive pathogen and late blight such a difficult disease to manage? The genus *Phytophthora* literally translated from Greek means *Phyton* = plant and *Phthora* = destroyer; and in the case of *P. infestans* this name makes sense. The pathogen has a sophisticated weaponry, including effector molecules coded by avirulence genes that allow rapid infection and host tissue colonization (Kamoun and Smart 2005; Haldar *et al*. 2006). Once inside host tissue, a complex set of compounds such as metallopeptidase, cutinase, and other proteins with no identifiable function required for cell killing and nutrient uptake are promptly activated (Lee *et al*. 2006). To date, 10 avirulence (Avr) proteins are known to be involved in

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pathogenesis and act as effectors that are delivered inside the plant cells (Haldar *et al.* 2006). These effector proteins can either trigger resistance response or induce disease in the plants. In addition, different types of proteases inhibitors can be produced by the pathogen to avoid defense proteins formed by the plant. Recently, a new class of such protease inhibitors were described in *P. infestans* (Tian *et al.* 2007). Despite milestones achievements related to the understanding of the basic biology of *P. infestans*, the molecular mechanisms involved in the pathogenesis are not completely resolved yet (Lee *et al.* 2006). Nevertheless, it is well-known that the time required for *P. infestans* to complete its life cycle, i.e. from infection to sporulation (production of new propagules), can be as short as three days. After this short incubation period, a single lesion may give rise to thousands of spores.

Late blight management has been heavily based on fungicide application and in many areas fungicide applications have increased over the last decade due to the introduction of new, more aggressive genotypes of the pathogen (Kato *et al.* 1997). At the same time, two counter-balancing factors have also grown: societal pressure for reducing pesticide use on crops and acreage of organically-grown food crops, potato and tomato included (Bettiol *et al.* 2004; Ghorbani *et al.* 2004; Obach 2007). Innovative and effective control measures are needed if fungicide use is to be reduced or, as in the case of organic production, eliminated.

Organic production of potato and tomato has for many decades depended on the application of copper-based fungicides (e.g. Bordeaux mixture, fixed-copper hydroxide, copper oxide, and copper oxychloride) for control of late blight. In Brazil (Bettiol *et al.* 2004) as well as in the USA (Koenig and Baker 2002) and Japan (Notification No. 59 of the Ministry of Agriculture, Forestry and Fisheries 2000 - www.maff.go.jp/soshiki/syokuhin/hinshitu/organic/eng_yuki_how.pdf) these compounds can be used in certified organic fields. However, there has been increasing pressure to find substitutes for these products because of environmental contamination caused by copper residues. Currently in the European Union only 6 kg of elemental copper per ha per year is allowed in organic production (Ghorbani *et al.* 2004). As soon as reliable alternatives to manage late blight are available, a complete ban of copper compounds should take place (Duncan 2003). Thus, there is enormous interest in finding effective non-chemical alternatives to protect potato and tomato fields against their most threatening foliar disease.

The term “alternative” can have multiple meanings and its interpretation varies accordingly. In this review we will use it in a relatively “loose” sense that includes natural compounds produced by plants or by microorganisms and also synthetic substances that do not directly affect pathogen development *per se*. Other non-fungicidal alternatives to late blight management such as cultural (fertilization, planting density, irrigation, etc.) and physical (irradiation, heat, etc.) control methods will not be addressed. Rather, only practices that involve applications of non-fungicidal compounds or their potential producers will be discussed.

BIOLOGY AND POPULATION GENETICS OF *P. INFESTANS* – IMPLICATIONS FOR EPIDEMIOLOGY OF LATE BLIGHT

Important aspects of the biology and population genetics of *P. infestans* should be considered for thorough analysis and proper development of alternative procedures aiming at controlling late blight. From a historical perspective, late blight has devastated potato and tomato crops for more than 150 years (Turner 2005). The pathogen, *P. infestans*, belongs to the kingdom Stramenopila, phylum Oomycota, family Pythiaceae. These organisms are morphologically similar to fungi but phylogenetically related to brown algae (Sogin and Silberman 1998). *P. infestans* can infect foliage, stems, potato tubers and tomato fruits at all stages of plant development. Several wild solanaceous plants can also be infected by *P. infestans* (Forbes and Landeo 2006). Eggplant has also been identified as a host, but our efforts to find infection on foliage in South America have proven unsuccessful. Eggplant fruits appear to be easily infected (unpublished data). Initially lesions on potato or tomato foliage appear as irregular, water-soaked spots, which may enlarge rapidly into pale green to brown; covering extensive surface of large leaves and stems (Stevenson 1991; Fry *et al.* 2001). Tomato fruit lesions appear as dark olivaceous spots, which may enlarge and cover the whole fruit surface (Stevenson 1991). In potato tubers, lesions are reddish brown, dry, and granular, and can develop in inner tissue (Fry *et al.* 2001). When humidity is high, lesions can be covered with a gray to white moldy growth, which consists of specialized hyphae called sporangiophores that produce the asexual propagules known as sporangia (Stevenson 1991; Fry *et al.* 2001). Flagellate zoospores, formed inside sporangia, can swim, encyst (loose its flagella) and form a germ tube, which can penetrate host tissue. The pathogen can also produce oospores, which are thick-walled sexual spores capable of surviving in the absence of host plants. Both the A1 and A2 mating types are needed for production of oospores, although presence of both types does not always lead to sexual reproduction, indicating that there are levels of incompatibility.

P. infestans is extremely aggressive on potato and tomato plants under favorable weather conditions, which include average temperature between 18-22°C and high relative humidity (80-100%) (Stevenson 1991; Fry *et al.* 2001). Determining what is optimal in the complex variable temperatures of nature is more difficult. A study using a late blight forecast model within a geographic information system (GIS) presented a global map of late blight severity based on the number of fungicide sprays that would be needed to control the disease (Hijmans *et al.* 2000). Using global weather data bases, a number of areas with apparently different climates had high disease severity, including Northern Europe, parts of the Himalayas and many areas within the highland tropics. Nonetheless, disease simulation indicates that temperature regimes from the temperate summer (northern United States for example) cause disease to develop faster than do the lower temperature regimes of the highland tropics (unpublished data).

The pathogenesis process begins with the germination of oospores, sporangia or zoospores. Sporangia are the most abundantly dispersed propagules and germination can be either (i) direct by development of a germ tube or (ii) indirect, by the release of zoospores. Temperature regulates the mode of germination of sporangia. Usually, direct germination of sporangia occurs at 18 to 24ºC, while indirect germination takes place at temperature lower than 18ºC (Crosier 1934). The pathogen can penetrate plant tissue through the cuticle and epidermal cells and develops specialized structures that can extract nutrients from host cells. Necrosis follows host tissue colonization by *P. infestans*. In green tissue surrounding necrotic areas, sporangiophores are formed through the stomata and produce sporangia. These propagules are generally dispersed by the wind, but also by rain over a short distance.

In areas where sexual reproduction does not occur or is rare, the pathogen survives from season to season in infected tubers and to a lesser extent on crop debris. In areas where viable oospores are formed, this propagule can survive in soil for years (Zwankhuizen *et al.* 2000; Stromberg *et al.* 2001; Fernández-Pavía *et al.* 2004; Lehtinen and Hannukkala 2004). In the highland tropics, host tissue is available all year.

The occurrence of sexual reproduction also affects the population dynamics of the pathogen. Two contrasting scenarios for the genetic structure of populations of *P. infestans* can be envisioned: (1) a panmictic population is comprised of a large number of distinct genotypes of *P. infestans*; and (2) a clonal population structure with a limited number of clonal lineages is established across large areas. The impact of control measures can vary according to the predominant genetic structure of the population (Grünwald *et al*. 2006). For panmitic populations, high genotype diversity can provide greater potential for local adaptation to a changing environment, which makes disease management more difficult (Sujkowski *et al*. 1994). In areas where the population is clonal, the risk of new diversity being introduced grows as seed trade becomes more globalized. Ideally, strategies for using alternative products should also consider information about population biology of the pathogen; however, we are unaware of clear indications that this has happened. In most situations, research programs are oriented towards the search for products effective against all possible genotypes of *P. infestans*.

NATURAL PRODUCTS DERIVED FROM PLANTS

In recent years there has been an increased interest in using natural substances derived from plants for different industrial purposes, for example as food seasoning and natural medicine (Isman 2000; Cao *et al*. 2004), and also for pest management in agriculture (Mansingh 2004). In most cases, it would appear that natural products derived from plants are safer than fungicides because they have low acute toxicity and because they are readily biodegradable into non-toxic products (Tripathi and Dubey 2004). Consequently, these compounds can be considered as substances of reduced risk to environment and human health (Duke *et al*. 2003). For example, in the study of mammalian toxicity, some pure essential oils of plant origin that were fed in large quantities to rats did not cause mortality (Isman 2000).

The plant-derived products most commonly used to control plant diseases are essential oils and extracts. The two types of plant-based products have many similarities but also differ for some characteristics. Essential oils are oily liquids obtained from plants through fermentation, enfleurage, extraction, and steam distillation (Burt 2004). Plant extracts, in contrast, are dried plant products obtained by filtration, distillation, and evaporation (Wang *et al*. 2004). The main compounds that have been investigated to date include phenols, flavonoids, quinones, tannins, alkaloids, saponins, and sterols (Halama and van Haluwin 2004). These products can have fungicidal or fungistatic effects on plant pathogens, or they can provide conditions favorable to the establishment and increase of antagonistic microorganisms on host plants (Scheuerell and Mahaffee 2002).

Essential oils, aromatic compounds, and products of hydrolytic reactions with antagonism activity against several plant pathogens have been investigated for their antimicrobial properties, including anti-oomycete activity (Mari *et al*. 2003). The information about the use of natural products derived from plants to control late blight is limited (Soylu *et al*. 2006). The majority of reports are restricted to bioassays conducted under laboratory conditions and often recording a single, but not necessarily reliable, variable: inhibition of *P. infestans* mycelial growth on culture media.

Plant extracts

The search for plant extracts with anti-oomycete activity has increased over the last years, and efficacy of plant extracts against *P. infestans* has also been demonstrated. Several preliminary studies were conducted mainly in China and India but, unfortunately, these are brief reports of mostly *in vitro* tests (Jiang *et al*. 2001; Cohen *et al*. 2002; Cao *et al*. 2003; Deepa *et al*. 2004). Some of them have great potential to be used as alternative compounds to synthetic fungicide. Another interesting aspect is that in many studies conducted in controlled environment these compounds were as efficient as synthetic fungicides in retarding growth of *P. infestans in vitro* or in reducing late blight severity on host plants. The results of these more detailed studies are discussed below.

Extracts of 88 plant species, distributed among 44 botanical families, were tested for their capacity to inhibit zoospore formation or the *in vitro* growth of *P. infestans* (Wang *et al*. 2001). In that study, extracts of 19 species were effective in reducing both variables. One of the most effective was extract of garlic gloves, which at 1 or 2% completely inhibited zoospore formation (Cao and van Bruggen 2001; Wang *et al*. 2001) and colony growth of *P. infestans* (Cao and van Bruggen 2001). In another study, extract of long pepper (*Piper longum* L.) was assessed against *P. infestans* on tomato plants under greenhouse conditions (Lee *et al*. 2001). Dried fruits of long pepper were crushed and subjected to compound extraction with methanol. The extract was partitioned into hexan, chloroform, ethyl acetate, butanol, and water soluble portions, which were dissolved and diluted in dimethyl sulfoxide (DMSO) and water, respectively. The portion obtained from the hexan fraction at 1 mg/ml reduced mortality of inoculated plants by 60% (Lee *et al*. 2001). A similar experiment was conducted to assess the effect of curcumin, a polyphenol compound present in the rhizome of turmeric (*Curcuma longa* L.). Tomato plantlets were treated with a curcumin solution of 500 or 1000 mg/l (which was dissolved and diluted in DMSO and water, respectively) and later inoculated with *P. infestans*. All treated plants survived and the level of late blight control achieved with curcumin was similar to that in plants treated with the fungicide chlorothalonil (Kim *et al*. 2003).

These apparently promising results should be carefully evaluated since most tests were conducted under conditions not similar to those experienced by farmers in the field. For instance, *in vitro* tests suggested that extracts made of clove and garlic controlled late blight (Cao and van Bruggen 2001; Wang *et al*. 2001). Thus, based on previously reported results, field trials were set up to quantify the efficacy of three mixtures of plant extracts in the control of tomato late blight: chili pepper, black pepper, clove, turmeric, and garlic; black pepper, clove, and garlic; and clove, turmeric, and garlic. Variables related to late blight progress such as severity at half of epidemic duration (Y_{50}); final severity (Y_{max}); area under the disease progress curve (AUDPC); and disease progress rate (r) were quantified (Diniz *et al*. 2006). Unfortunately, under field conditions in Brazil, none of the plant extract combinations reduced any of the variables related to late blight intensity (Diniz *et al*. 2006). Low concentrations of the putative active ingredients allylsufites and dissulfite (allyldissulfites and allylmethyl dissulfites) (Cao and van Bruggen 2001), pipernonaline (Lee *et al*. 2001) and curcumin (Kim *et al*. 2003) in the extracts and/or their chemical and physical degradation products could have interfered with the tested compounds, reducing their efficacy in the field.

There is variation in the way compounds are extracted and also regarding the solvents used to obtain them. Plant extracts have been formulated as botanical fungicide dissolved in DMSO (Kim *et al.* 2003), ethanol benzene, diethylether, toluene (Deepa *et al*. 2004), acetone (Wang *et al.* 2004), water (Wang *et al.* 2004; Stephan *et al.* 2005), sodium citrate and sodium chloride (Burt 2004). However, there is limited information about the persistency and putative toxicity on plants sprayed with plant extracts obtained with these solvents. Temperature, ultraviolet light, pH on treated plant parts, rainfall, and other environmental factors may exert a more or less negative influence on the active principles (Schmutterer 1990). For instance, the residual effect of neem-based products is, in general, restricted to a few days, mostly around five to seven days (Schmutterer 1990). Nevertheless, the residual effect of some protective fungicides on plants is around one week, which does not compromise the use of neem compounds as natural fungicide.

Inula viscosa is a common medicinal plant native to the Mediterranean Basin. In addition to its therapeutic properties, this plant has antimicrobial activity against several plant pathogens. In a growth chamber study, extracts diluted in acetone or water at 1% (w/v) reduced late blight severity by more than 90% on potato and tomato plants (Wang *et al.* 2004). Several potentially active compounds were identified in paste samples: tomentosin, inuviscolide, costic acid, and isocostic acid. Furthermore, four thin-layer chromatography regions were highly inhibitory to *P. infestans*. However, compounds from these regions were not identified (Wang *et al*. 2004).

Medicinal plants native to China were evaluated against *P. infestans* in a detached potato leaf bioassay and in the field. Plant species included in this study were: *Terminalia chebula*, *Anemarrhena asphodeloides*, *Allium sativum*, *Galla chinensis*, and *Perilla frutenscens*. Based on the results of detached-leaf trials, only *T. chebula* and *G. chinensis* were selected for field experiments (Cao *et al*. 2004). The efficacy of these products to control late blight on potato plants in the field was low compared with a copper-based fungicide. The percentage of inhibition of late blight with the fungicide was around 60% and 80% in potato cvs. 'Agria' and 'Nicola', respectively. In potato cultivar 'Nicola' treated with *T. chebula* and *G. chinensis*, the percentages of inhibition were around 30% and 10%, respectively. There was no control of late blight in potato cv. 'Agria' treated with *G. chinensis*. However, *T. chebula* did control late blight (40%) in potato cultivar 'Agria'. Poor rainfastness of plant extracts was proposed as the main factor for the limited late blight control in the field (Cao *et al*. 2004). Protection of field-grown potato and tomato plants may depend on the association of effective plant extracts with adhesive adjuvants. On the other hand, use of extracts without adhesives might be suitable for greenhouse-grown tomatoes.

The commercial plant preparation Elot-Vis® (Prophyta GmbH, Germany) and extracts from *Rheum rhabarbarum*, *Solidago canadensis*, *Artemisia vulgaris*, *Impatiens parviflora*, and *Urtica dioica* reduced late blight severity on detached potato leaves (Stephan *et al*. 2005). These treatments were repeated in a second experiment, but only *S. canadensis* and Elot-Vis®, applied 24 h before inoculation with *P. infestans* reduced disease development on potato leaves. Nevertheless, *R. rhabarbarum*, applied 1 h after inoculation with the pathogen, was efficient in controlling late blight. When the plant extracts were tested at different concentrations on detached potato leaves *S. canadensis* reduced the infection of *P. infestans* at 0.1, 1, and 5% w/v, while *R. rhabarbarum* inhibited *P. infestans* at 1 and 5% w/v. Although extracts of *S. canadensis* and *R. rhabarbarum* are more efficient in controlling late blight at 5%, these products were phytotoxic at this higher concentration (Stephan *et al*. 2005).

In the same study, the authors demonstrated that *R. rhabarbarum* and *S. canadensis* also reduced late blight severity on potato plants in a growth chamber. Extracts were more effective when applied up to 3 days before inoculation with *P. infestans*. Late blight severity in plants treated with *R. rhabarbarum* and *S. canadensis* was around 40% and 50%, respectively, while severity in control plants, inoculated only with the pathogen, was above 60%. According to the authors, these products will be tested in combination with non-chemical agents and forecasting models in field experiments (Stephan *et al*. 2005).

Water-based extracts of fermented soil and plant composts, generally referred to as compost teas, have been tested as control products for many plant diseases including late blight. Compost teas purportedly improve soil fertility and at the same time control pests and plant pathogens. Microbial populations in compost tea are likely to be the "active ingredients" responsible for the efficacy of the compound (Scheuerell and Mahaffee 2002). The effects of a commercially available compost tea (Jolly Farmer®) and a foliar feed extract (Acadian SeaPlants Inc.) derived from powdered kelp (*Ascophylum nodosum*) on the bacterial community of potato foliage was assessed in field trials (Sturz *et al*. 2006). These products were applied on potato plants at 5- to 10-day intervals throughout the 2003 and 2004 growing seasons. Epiphytic bacteria were sampled and identified by fatty acid analysis and 16S ribosomal RNA genes. Population densities of bacterial community were greater in plants treated with the compost tea than in powdered-kelp treated plants. Bacterial density was lowest on potato foliage treated with the fungicide mancozeb. In another bioassay, bacterial isolates recovered from plants treated with both compost tea and a foliar feed extract inhibited growth of *P. infestans* on culture media (Sturz *et al*. 2006). These alternative products are less toxic to the microbial community than fungicides. Nevertheless, more studies on the biology and ecology of microorganisms associated with these compounds are required to evaluate their potential role in late blight control.

Galls caused by an aphid, *Schlechtendalia chinensis*, are commonly found in nutgall sumac tree (*Rhus javanica*). Interestingly, these galls can be a source of several antimicrobial compounds, which have attracted interest from the chemical and pharmaceutical industries. Several of these compounds have high fungicidal activity. Methanolic extracts obtained from these galls reduced late blight severity on tomato plants by more than 90% (Ahn *et al*. 2005).

Other compounds also reported to have inhibited growth of *P. infestans* under laboratory conditions are extracts derived from *Pseudarthria viscida* (Deepa *et al*. 2004), *Cassia tora* (Kim *et al*. 2004) and *Catalpa ovata* (Cho *et al*. 2006). Bryophyte extracts from *Bazzania trilobata* and *Diplophyllum albicans* also reduced late blight severity by more than 70% on tomato plants in growth chamber (Mekuria *et al*. 2005). Extracts obtained from lichens, such as *Evernia prunastri*, *Hypogymnia physodes*, and *Cladonia portentosa* were also reported as capable of inhibiting *P. infestans in vitro* (Halama and van Haluwin 2004).

Essential oils

Neem (*Azadirachta indica* L.) is a widely used and well-known plant from which seed extracts and oils are commonly used to control insects and pathogens. A high content of azadirachtin, its active ingredient, can be found both in the oil and in the extract (Mordue and Nisbet 2000). In tomato crops, neem oil and extract have been used to control whiteflies (*Bemisia tabaci*) (Kumar and Poehling 2006), nematodes, fungi (Abbasi *et al*. 2005), and also *P. infestans* (Diniz *et al*. 2006; Rani *et al*. 2006). Late blight severity at Y_{50} was similar on plants treated with neem oil (0.5%) and Bordeaux mixture (1%), but Y_{max} on neem-treated plants (44%) was higher than on plants treated with Bordeaux mixture (14%). The disease progress rate "r" (0.16) and AUDPC (533) values were lower in plots treated with neem oil than in the controls (r = 0.21 and AUDPC = 1186) and similar to the Bordeaux mixture plots (r = 0.16 and AUDPC = 130) (Diniz *et al*. 2006). Neem oil is potentially useful and its efficacy may be improved at higher concentrations or in combination with other compounds or microorganisms.

Essential oils from 19 plants were evaluated for their efficacy in controlling *P. infestans* on potato in both *in vitro* bioassays and greenhouse experiments. The most inhibitory compounds *in vitro* were essential oils derived from thyme (*Thymus vulgaris*), peppermint (*Mentha piperita*), dill (*Anethum graveolens*), caraway (*Carum carvi*), and hyssop (*Hyssopus officinalis*) (Quintanilla *et al*. 2002). In the greenhouse experiments, these essential oils reduced late blight severity on potato plants, although the essential oil from hyssop was the most effective. Maximum late blight severity values on treated plants were 30% and 15% for cultivars 'Mandel' and 'Kerrs pink', respectively. In the control treatment, where plants were inoculated with *P. infestans*, severity was above 65% in both potato varieties. Phytotoxicity was observed on potato plants treated with compounds derived from caraway and thyme (Quintanilla *et al*. 2002). We are not aware of any trials designed to assess the efficacy of any of these particular essential oils under field conditions.

The anti-oomycete activity of essential oils derived from *Origanum syriacum* var. *bevanii*, *Thymbra spicata* subsp. *spicata*, *Lavandula stoechas* subsp. *stoechas*, *Rosmarinus officinalis*, *Foeniculum vulgare*, and *Laurus nobilis*, was assessed against *P. infestans* in assays designed to test both contact and volatile effects. The contact effect was evaluated by amending V8 juice agar medium with the compound of interest. The volatile phase effect was assessed with sterile filter paper moistened with essential oils and placed on the inner surface of the inverted lid of Petri plates (Soylu *et al*. 2006). All essential oils reduced mycelial growth of *P. infestans* in both the volatile and contact tests. The compounds caused degenerative changes in sporangiophore morphology and inhibition of sporangia formation. Components of essential oils used in these experiments were identified as carvacrol, camphor, borneol, 1,8-cineole, and anethole. Although all tests had been conducted in a laboratory, essential oils putatively can control late blight under field conditions and the authors proposed the development of a new class of fungicides based on these compounds (Soylu *et al*. 2006).

Compounds such as: **1.** (3′-chlorophenyl)propan-1-ol, **2.** (4′-chlorophenyl)propan-1-ol, **3.** 1-(4′-chlorophenyl)-2-cyclopropylethanol, **4.** 1-(4′-chlorophenyl)propan-2-phenylethanol, **5.** 2β-(2′nitroethoxy)cloven-9α-ol, **6.** N-(9α-hydroxyclovan-2β-yl)-N″-p-bromophenylacetamidine, and **7.** 2β-(2′-pseudothiouretoxy) cloven-9α-ol have been reported as capable of preventing growth of *P. infestans* in culture medium (Fernandez-Acero *et al*. 2006). Compounds **1** to **4** are derivatives of phytoalexins, and the others are based on clovane structures. Compounds derived from phytoalexins inhibited growth by more than 31% at 100 ppm. However, compounds **1** and **2** were more efficient at 200 ppm, where pathogen growth was reduced by 48%. Compounds derived from clovane were more efficient than phytoalexin derivatives, and inhibition was near 46% at 100 ppm. These compounds could be an alternative to synthetic fungicides because of low persistency in the ecosystem (Fernandez-Acero *et al*. 2006).

Currently, there are commercial compounds derived from lavender, oregano, thyme borneal, and majoram (Aromaland, Inc., Santa Fe, NM), which are used to control insects and plant pathogens. The anti-oomycete activity of these compounds was evaluated against isolates of *P. infestans* represented by genotypes US-8, US-14, and US-1 (Olanya and Larkin 2006). Essential oils from these compounds were added to culture media at concentrations of 100 and 1000 ppm to evaluate the inhibition of pathogen growth. All compounds affected growth at 1000 ppm, except that from oregano, which completely suppressed the development of *P. infestans* at both concentrations. Efficacy of the essential oil derived from oregano was further investigated on potato plants in a growth chamber. Oregano oil reduced late blight severity by 28-30%. Bioassays were also done at higher doses to increase disease control, however, the compound was strongly phytotoxic when applied at 2 and 20% (Olanya and Larkin 2006).

ANTAGONISTIC MICROORGANISMS AND CLASSICAL BIOLOGICAL CONTROL APPROACHES

The market for non-conventional fungicide products, specifically biological control agent formulations, is growing at a rapid pace. For some diseases, biological control is a routine part of disease management (e.g. Crown gall of Rosaceae; Citrus tristeza; Powdery mildew; etc.). Several commercial formulations of biocontrol agents are available and many others are currently being developed (Fravel 2005). Nevertheless, there are relatively few reports of biological control as a potentially successful alternative for management of highly destructive epidemics such as potato and tomato late blight, rice blast, apple fire blight, and black Sigatoka of banana. Two factors, probably among others, that make biocontrol difficult for these diseases are rapid establishment of infection and explosive disease development. It is reasonable to assume that many attempts to use biocontrol for potato and tomato late blight have been unsuccessful and this may be the reason why the literature in this field is so scarce. In fact, most of literature in this area is rather recent, having appeared in the last 20 years.

Antagonism by microorganisms

Preparations made of saprophytic, epiphytic, and endophytic organisms have been assessed as potential biocontrol agents to late blight management and a compilation of the studies will be presented. We define direct antagonism as the suppressive effects of biological control agents against *P. infestans* due to antibiosis, hyperparasitism, and competition for space and nutrients. Although in many cases the mechanisms of action of biocontrol agents were not elucidated, there are several evidence that suggests the involvement of direct antagonism.

Microorganism preparations applied to protect plant tissues

One of the first published reports of biocontrol of late blight was on the use of conidial suspensions (10^{4-5} spores/ml) of *Penicillium aurantiogriseum* and *Stachybotrys atra* (= *S. chartarum*) applied to leaflets of greenhouse-grown potato plants 12 h prior to the inoculation with *P. infestans* (Jindal *et al*. 1988). Late blight intensity was reduced by 93% and 84%, respectively. Simultaneous application of the biocontrol agents with pathogen inoculum also resulted in late blight control (86% and 65% reduction for *P. aurantiogriseum* and *S. atra*, respectively). When culture filtrates of both organisms were applied 12 h before or at the time of inoculation, late blight severity was also reduced. Possible explanations for the observed results were antibiosis and competition for space and nutrients (Jindal *et al*. 1988), however, no detailed studies were conducted to investigate the mechanisms of control. *S. atra* is a cellulose-decaying fungus (Chapman 2003), thus its cellulolytic activity could affect the integrity of *P. infestans* cell wall. One negative aspect of using *S. atra* as a biocontrol agent is the well-known capability of this species to produce trichothecene, a mycotoxin that can affect human health (Chapman 2003).

Durable and consistent control of late blight was reported from a series of experiments carried out under field conditions in Germany (Weltzien 1991). Compost tea made from horse or cow manure, amended or not with seven microorganisms, was applied to potato foliage to control late blight. In plants treated with compost tea amended with microorganisms, final late blight severity was 11% and did not differ significantly from that recorded in plants treated with a mixture of the fungicides metalaxyl and mancozeb. Adding microorganisms to the compost tea significantly enhanced disease control (Weltzien 1991).

Apparently variation in the composition of compost microflora can contribute to inconsistency in late blight control. In The Netherlands, the two most effective isolates among more than 200 microorganisms isolated from compost extract (a mixture of compost + water 1:9 w/w) or from the phyllosphere of potato plants were used in field trials designed to assess their biological control capabilities. The two bacterial isolates, one fluorescent *Pseudomonas* sp. and one *Bacillus* sp., did not control late blight under field conditions either when applied alone or in combination with compost extracts (Jongebloed *et al*. 1993). Thus, addition of effective antagonists to the compost extract did not increase efficacy of the extract. The interactions among organisms "native" to the compost with those amended to it seem to play a role in disease control. However, for practical purposes, this is likely to be a difficult factor to be taken into account and controlled.

Biological control of tuber blight (infection of tubers by *P. infestans*) has also been investigated (Culow *et al*. 1995). Tubers grown in wet substrates (soil or compost) were less susceptible than those formed in dry conditions (Stewart *et al*. 1993). Inhibition of tuber blight was not due to *Streptomyces* sp., *Penicillium* sp., *Trichoderma* sp., *Gliocladium* sp., or *Rhizoctonia* sp., however, bacteria isolated from the surface of tubers and then cultivated in compost were capable of inhibiting growth of *P. infestans in vitro*. Higher numbers of bacterial isolates were recovered from tubers kept in wet conditions, mainly for the more resistant cultivar (Clulow *et al*. 1995). Unfortunately, no characterization of the bacterial isolates was done and we are not aware of any further developments towards using these agents under field conditions.

Fungal and bacterial isolates from the phylloplane and rhizoplane of cultivated and wild tomatoes were able to reduce late blight lesion size on detached leaflets and in whole tomato plants (Garita *et al*. 1998). One bacterial isolate of *Serratia* sp. and isolates of *Trichoderma* sp., *Fusarium* sp., and *Penicillium* sp. were selected as potential biocontrol agents, but none were effective in reducing late blight severity in the field (Garita *et al*. 1999).

Among the many trials conducted so far, the most consistent results of biological control of late blight have been achieved with the application of *Xenorhabdus* spp. (Li *et al*. 1995; Ng and Webster 1997; Yang *et al*. 2001). The antagonist is a Gram-negative member of the Enterobacteriaceae, commonly found in mutual relationship with entomopathogenic nematodes (Kaya *et al*. 2006). Formulations based on the complex of nematode and bacterial symbiont are available commercially and used against soil pests. Metabolites produced by species of *Xenorhabdus* have been evaluated against *Phytophthora* spp., including *P. infestans* (Kaya *et al*. 2006). Different types of antibiotics can be produced by *Xenorhabdus* spp.: indole derivatives; xenorhabdins, which are organically soluble dithiolopyrrolones; and the xenocoumacins, which are water soluble benzopyran-1-one derivatives (Li *et al*. 1995). The organic fraction of the supernatant of a tryptic soy broth culture of *X. bovienii* coming from the nematode *Steinernema feltiae* was tested against potato late blight. Both *in vitro* and *in vivo* trials were conducted and phytotoxicity of the metabolites to potato plants was evaluated. The organic fraction at 0.1 and 1.0 mg/ml completely prevented mycelial growth of *P. infestans in vitro* and the size of late blight lesions was reduced when detached leaflets were treated with 10 or 50 mg/ml (Ng and Webster 1997). Leaflets treated with the compounds had alterations and slight phytotoxic effects were detected in those treated with 10 mg/ml and, more intensively, with 50 mg/ml. In whole plants, application of the metabolites of *X. bovienii* at 10 mg/ml resulted in control level similar to that achieved with the protectant fungicide chlorothalonil (Ng and Webster 1997). A summary of the results is presented in **Fig. 1**.

Aminoglycoside antibiotic compounds have been reported to reduce the intensity of diseases caused by oomycetes (Jones and Samac 1996; Xiao *et al*. 2002), including *P. infestans* (Lee *et al*. 2005). Four purified commercial aminoglycoside antibiotics: neomycin, paromomycin, ribostamycin, and streptomycin were tested against *P. infestans*. Paromomycin was the most active compound against the pathogen *in vitro* and the estimated effective dose to reduce mycelial growth by 50% was approximately 10 μg/ml (Lee *et al*. 2005). Subsequent tests were done on tomato plants inoculated with *P. infestans* and then treated with either commercially available paromomycin or with the culture filtrate of a paromomycin-producing actinomycete *Streptomyces* sp. (strain AMG-P1). Outstanding tomato late blight control was reported when the concentration of paromomycin was adjusted to 100 μg/ml. Strain AMG-P1 produced this antibiotic at a rate of 25 mg/l. A freeze-dried culture extract used at the rate of 125 μg/ml gave effective disease control against tomato late blight. Nevertheless, even though no phytotoxicity was mentioned, tomato plants treated with 125, 250 and 500 μg/ml apparently had prominent chlorosis (see Fig. 3 in Lee *et al*. 2005).

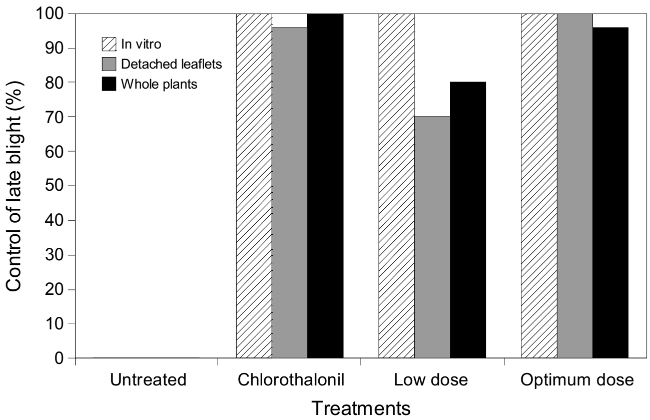

Fig. 1 Control of *P. infestans* mycelial growth *in vitro* or of late blight on detached leaflets or whole plants with metabolites produced by *X. bovienii*. Low doses of the metabolite correspond to 0.1, 2.0, and 5.0 mg/ml for the *in vitro*, detached leaflet and whole plants, respectively. Optimum doses in each of these experiments correspond to 1.0; 10.0; and 10.0 mg/ml, respectively. Modified from Ng and Webster (1997).

In another study, metabolites from the culture broth of *X. nematophilus* isolated from the nematode *S. carpocapsae* were also tested for the control of late blight in potted potato plants. Metabolites (in this study, the whole supernatant of culture broth) at 25 and 50 mg/l were effective in reducing late blight intensity (Yang *et al*. 2001). However, plants were inoculated with sporangial suspension of *P. infestans* as shortly as 2h after metabolite application. It would be interesting to assess longer time intervals between metabolite application and pathogen inoculation; also an assessment of the efficacy of treatment with bacteria, instead of the metabolites, could provide useful insights.

An interesting study demonstrated that a yeast-like fungus *Pseudozyma flocculosa* inhibited growth of *P. infestans in vitro* by means of *cis*-9 heptadecenoic acid (CHDA), a fatty acid molecule. Detailed biochemical analyses indicated that CHDA was incorporated in the membrane of *P. infestans* and affected its permeability. The authors postulated that altered membrane permeability would lead to an increase in electrolyte and protein loss and even cytoplasmic disintegration of cells (Avis and Bélanger 2001).

Fourteen of 83 bacterial isolates, mostly *Pseudomonas* spp. and *Bacillus* spp., were capable of preventing growth of *P. infestans in vitro* (El-Sheikh *et al*. 2002). Overall, *Pseudomonas* spp. isolates were more effective than *Bacillus* spp. isolates and both were more effective when applied preventively. Three isolates (2 *Bacillus* sp. and 1 *Pseudomonas* sp.) had good antagonistic properties, but caused tuber soft rot and were discarded. High levels of late blight control were reported in this study for application of antagonists, but no fungicide treatment was included for comparison.

Isolates of the bacterial genera *Bacillus*, *Pseudomonas*, *Rahnella*, and *Serratia* contributed to a reduction in late blight severity in potato plants in controlled conditions. This study is noteworthy because a highly aggressive isolate of the US-8 clonal lineage of *P. infestans* was used for inoculations. Several mechanisms of inhibition were thought to jointly act to reduce late blight intensity (Daayf *et al*. 2003). An isolate of *Pseudomonas putida* did not inhibit *in vitro* growth of *P. infestans*, but induced systemic resistance in potato plants, whereas an isolate of *Serratia plymuthica* inhibited *in vitro* growth of *P. infestans* by antibiosis, but did not induce systemic resistance to the pathogen. However, both bacteria were effective in controlling late blight (Daayf *et al*. 2003).

Use of endophytic organisms

Another line of investigation in the general area of biocontrol is the use of endophytic organisms to control pathogen development. Control of late blight was attempted with arbuscular mycorrhizal fungi (AMF) (O'Herlihy *et al*. 2003). Potato plantlets originating from tissue culture were transplanted to the field and commercial inoculum of AMF was applied in-furrow at planting. The authors claimed that the late blight epidemic on AMF treated potato plants was delayed, but careful analysis of the disease progress curves revealed that the major epidemiological effect of AMF application was a reduction of the disease progress rate. For polycyclic diseases such as late blight this is the most effective strategy to reduce crop losses. In this experiment, even though final late blight severity was high (around 80%), tuber yield in AMF-treated plots did not differ from the most effective treatment: application of fungicide plus chitosan (see O'Herlihy *et al*. 2003).

Rhizobacteria

Crop growth, yield, and disease resistance can be enhanced by plant growth promoting rhizobacteria (PGPR) (Pieterse *et al*. 2003). Two PGPR, *Bacillus pumilus* and *Pseudomonas fluorescens*, induced resistance to *P. infestans* and there was reduced zoospore formation and germination (Yan *et al*. 2002). The *in vitro* and *in vivo* tests have shown that species of *Bacillus*, *Pseudomonas*, *Rahnella*, and *Serratia* can lessen late blight symptoms by a combination of antibiosis and induced resistance against *P. infestans* (Daayf *et al*. 2003). When phylloplane isolated organisms were tested in combination with *B. cereus*, a PGPR, late blight severity was significantly reduced compared with application of epiphytes alone. Curiously, application of the PGPR alone was not effective in reducing tomato late blight intensity, indicating an apparent synergistic effect (Lourenço Jr. *et al*. 2006).

Combination of potential antagonists

As noted previously, combining antagonists with different modes of action can lead to better control (Punja 1997; van Lenteren 2000). With the objective of developing several strategies to manage late blight in both conventional and organic production, potentially useful biocontrol agents for late blight management were isolated in Brazil. Many phylloplane microorganisms and rhizobacteria isolated from conventional or organically grown tomato plants were tested for antagonistic activity against *P. infestans*. Based on *in vitro* inhibition of sporangia germination and detached leaflet bioassays, four phylloplane microorganisms *Aspergillus* sp., *Cellulomonas flavigena*, *Candida* sp., and *Cryptococcus* sp. were selected (Lourenço Jr. *et al*. 2006).

A strategy of selecting antagonists that could hamper distinct stages of *P. infestans* pathogenesis was implemented at the screening stage of candidate microorganisms. *C. flavigena* and *Cryptococcus* sp. inhibited sporangia germination, but did not reduce late blight severity in detached leaflets (Lourenço Jr. *et al*. 2006). *Aspergillus* sp. and *Candida* sp. reduced both sporangia germination and disease severity, probably, through reduced infection frequency due to low sporangial germination and/or inhibited zoospore germ tube formation.

The observation of limited infection of *P. infestans* on potato tubers from some susceptible varieties in the Tolluca Valley incited researchers to search for microorganisms as possible antagonists to this pathogen. Isolates of *Pseudomonas* spp., *Burkholderia* spp., *Streptomyces* spp., and *Trichoderma* spp. were obtained from stems, leaves, tubers, and rhizoplane of potato plants. The suppressive activity of these microorganisms to A1 and A2 mating type isolates of *P. infestans* was assessed on potato leaves kept in a moist chamber, and also plants grown in a greenhouse and in the field (Lozoya-Saldaña *et al*. 2006). In the first experiment, the microorganisms were evaluated individually or in combinations on detached potato leaves inoculated with zoospores and sporangia of *P. infestans*. Reduction of late blight severity occurred with *Burkholderia* spp., *Streptomyces* spp., and *Pseudomonas* spp., applied individually and in combination within and among species (Lozoya-Saldaña *et al*. 2006).

Strains of *Pseudomonas* spp. selected in the detached-leaf assay were tested, along with several isolates of *Trichoderma* spp. for late blight control on potato plants in a greenhouse. The value of the area under disease progress curve (AUDPC) in the control plants, which were inoculated only with the pathogen, was 770, while that of the treatments with mixed bacterial strains, combined isolates of *Trichoderma* spp., and a commercial formulation of *Trichoderma* spp. (Biopack-F), were 313.3, 373.3, and 366.3, respectively (Lozoya-Saldaña *et al*. 2006).

Results obtained under greenhouse conditions were not repeatable in the field. In two field experiments done 2001 and 2002, only the treatments with mixed strains of *Pseudomonas* were efficient in reducing disease. In 2001, a mix of strains of *Pseudomonas* spp. was the best treatment and in 2002 the most effective treatment was a mix of *Pseudomonas* spp. and *Burkholderia*. The AUDPC values of the control plants were 1698.2 and 716.1, while those of the treatment with bacterial strains were 1172.5 and 520.8, during 2001 and 2002, respectively. Preparations based on the antagonists used in these experiments could be implemented as a control measure in greenhouses or in areas where the late blight epidemics are less severe, mainly when combined with other available methods of controlling *P. infestans* (Lozoya-Saldaña *et al*. 2006).

Under controlled conditions, tomato late blight severity was reduced when a combination of antagonists was used. The best results were achieved when roots of tomato plants were treated with the rhizobacterium *B. cereus* concomitantly with the treatment of foliage with epiphytic microorganisms (Silva *et al*. 2004; Lourenço Jr. *et al*. 2006). *B. cereus* is postulated to have induced systemic resistance in tomato to *P. infestans*, since it has been shown to induce non specific resistance to other pathogens in tomatoes (Silva *et al*. 2004).

The impacts of a potentially effective biocontrol agent against late blight would enhance disease management in organic cropping systems and its contribution would be of great relevance. Bordeaux mixture and other copper-based fungicides are still used to control late blight in organic crops in most countries. However, it is expected that copper-based fungicides will be reduced or banned in the near future. A combination of biocontrol agents with products such as neem oil, which was demonstrated to be effective in reducing tomato late blight severity (Diniz *et al*. 2006), could be another option to reduce crop losses caused by the disease.

Use of commercial products based on biological control agents

Several commercial formulations of biocontrol agents have been tested for efficacy against late blight. Of many trials involving different microorganisms, including *Trichoderma harzianum*, *Bacillus subtilis*, *Streptomyces* sp., *Coniothyrium minitans* and a pool of undetermined effective microorganisms (EM 5), the most effective was the *B. subtilis* based-product Serenade®. Curiously, bacterial cells were not directly responsible for the inhibition of *P. infestans*. A cell-free culture extract contained metabolites that were active against *P. infestans* (Stephan *et al*. 2005). Caution must be exerted when using biocontrol agents capable of producing metabolites with antibiotic activity. *B. subtilis*, an ubiquitous bacteria, can produce antibiotic compounds (Romero-Tabarez *et al*. 2006) and little is known about the persistence of these molecules on plant products or in the environment.

In a similar study, commercial formulations of three well-known antagonist species, *Trichoderma harzianum* (Plant Shield HC®), *Gliocladium virens* (G41), and *Bacillus subtilis* (Rhapsody AS®) were tested for the control of late blight on tomatoes and petunias, under greenhouse conditions. Biocontrol agents were not effective in controlling late blight on either host, but on petunias, which is less susceptible to late blight than tomato, the results were more promising (Becktell *et al*. 2005). These results highlight the potential of integrated management by adjusting biocontrol agent applications as a function of host resistance and environmental conditions.

NON-FUNGICIDAL CHEMICAL INDUCERS

When extract of dried mycelium of the penicillin-producing fungus *Penicillium chrysogenum* was sprayed on tomato plants in a greenhouse and on potato plants in the field, late blight severity was reduced by 71% on tomatoes but there was no reduction on potatoes (Thuerig *et al*. 2006). No penicillin residue was present in the extract (mycelium was dried for 3 h at 140°C and the antibiotic is thermo-labile) and the extract had no direct effect on *in vitro* growth of *P. infestans*. The authors proposed that differences in response were plant-mediated, in that dried mycelium induced resistance in tomato, but not on potato. In a complementary study, an extract of *P. chrysogenum* was used to induce resistance against *P. infestans* in tomato plants (Unger *et al*. 2006). A high level of late blight control (>90%) was achieved with two foliar sprays of the extract. The induction of resistance was positively correlated with activity of peroxidase, an enzyme known to participate in resistance response processes in several plant-pathogen interactions (Unger *et al*. 2006).

Chitosan, a linear polysaccharide composed of randomly distributed β-(1-4)-linked D-glucosamine and N-acetyl-D-glucosamine, is reported to act as an antimicrobial compound and also boost the ability of plants to defend against infections (Rabea *et al*. 2003). Late blight epidemics were delayed on plots that received 8 sprays of 0.1% (w:v) chitosan (N, O-carboximethyl chitosan) compared to the control and the treatment was at least as effective as the fungicide metalaxyl. When chitosan application was alternated with metalaxyl, there was an apparent synergistic effect and the number of metalaxyl sprays was reduced by 50% (from 8 to 4) (Rabea *et al*. 2003). This promising result should be further evaluated under different environmental conditions to assure consistency.

Cao and Forrer (2001) classified different antagonists/compounds for their efficacy in controlling late blight. Two organisms were classified as the most effective: A *Pseudomonas* strain used in a product identified as Immunofit M. biomonas (Filippov and Kuznetsova 1994) and *Fusarium equisetii* (Jindal *et al*. 1988). Among the chemical inducers, arachidonic acid, eicosapentaetaenoic acid, linoleic acid, and salicic acid were ranked as the most effective compounds. The activity of synthetic amino-*n*-butanoic acids (BABA) were assessed against *P. infestans* in tomato plants and *in vitro* tests (Cohen 1994). Generally, BABA did not reduce sporangia germination and mycelial growth of *P. infestans*. Nevertheless, these compounds, applied at 2000 ppm, reduced the late blight severity in tomato plants. The levels of control provided with the application of 3-ABA, 2-ABA, and 4-ABA, were 84%, 36%, and 15%, respectively. Nevertheless, apparently no additional studies were carried with these synthetic amino acids regarding late blight control.

OTHER PRODUCTS

Diluted cow milk (10 or 20%) was reported as an effective alternative product to control powdery mildew on zucchini (Bettiol 1999) and raw milk was effective in reducing the severity of sorghum downy mildew (*Peronosclerospora sorghi*) (Arun *et al*. 2004). It was postulated that diluted cow milk (20%) could also control *P. infestans*, another oomycete, however at this concentration it did not reduce the severity of tomato late blight. Values of r and AUDPC on plots treated with milk were similar to those in the control plots (Diniz *et al*. 2006).

Homeopathic preparations have been tested for the control of various diseases and pests on different crops. In tomatoes, post-harvest fruit rot caused by *Fusarium roseum* was reduced with preparations made from *Arsenicum album* (C1), *Kali iodatum* (C149), *Phosphorus* (C35), and *Thuja occidentalis* (C87). The latter inhibited spore germination, while fungal colony growth was inhibited by *Kali iodatum* (C149) and *Thuja occidentalis* (C87) (Khanna and Chandra 1976). The incidence of tomato powdery mildew (*Oidiopsis siculae*) was 46.4% in plants treated with *Kali iodatum* (C100) whereas it reached 58% in untreated control plants (Rolim *et al*. 2001). To date there is no report of successful late blight control with homeopathic preparations. In the early 1990's a homeopathic product was tested for the control of potato late blight with no success (van Bol *et al*. 1993). Recently, a nosode preparation made of *P. infestans*-infected tomato leaflet was tested for control of tomato late blight. There was no reduction of late blight severity at either the midpoint or the end of the epidemic. Also, no reduction in AUDPC and r values were observed when plants were treated with the homeopathic product (Diniz *et al*. 2006). Given the peculiar characteristics of pathosystems involving *P. infestans* and potato or tomato, the potential for using homeopathic preparations to reduce epidemics under field conditions would appear unlikely.

CONCLUSIONS

As stated in the beginning of this review, the way *P. infestans* infects and kills host tissue and its very high multiplication rate present major barriers for late blight management with alternative compounds. Most strategies used so far focused in preventing pathogen establishment in the host plant by reducing germination/infection processes; most of them combining organisms with similar modes of action; e.g. antibiosis (Ng and Webster 1997; Garita *et al*. 1998). For many pathosystems this might be appropriate; but the epidemiological characteristics of late blight makes the adoption of a "silver bullet approach" risky. *P. infestans* propagules are produced in high numbers and promptly dispersed by wind. Additionally, inoculum deposition on plant surface is a stochastic process (Aylor 1986). For these reasons, active antagonists should be present in sufficient number and well distributed on host tissue. Under field conditions, however, the combination of these conditions is unlikely to be always met. Thus, a safer approach to late blight biocontrol would be to use a combination of compounds and microorganisms with different modes of action beginning at the early stages of the host-pathogen interaction. For example, a potentially useful scheme would be protection conferred by antagonists applied on the phylloplane with effective endophytic treatments that could help prevent pathogen establishment combined with resistance induction mediated by PGPR before pathogen infection takes place.

Other important aspects related to the biological control of late blight are:

- More effort should be devoted to the development of formulations that can supply nutrients and protection to biocontrol agents applied to either the host plant or another environment where the pathogen is colonizing or surviving.
- Biocontrol can be enhanced by implementation of cultural practices, which create environmental conditions favorable to antagonists, and by activation of plant resistance with chemical or biological inducers.
- Under highly disease-conducive conditions, i.e. favorable environmental conditions and host susceptibility, more effective treatments should be applied. On the other hand, under less conducive conditions, complementation with biological control preparations may contribute to reduce the number of fungicides applications.
- There is a knowledge gap regarding biocontrol agents capable of reducing viability of survival structures of the pathogen. For regions where sexual reproduction occurs and oospores contribute to the epidemics, acceleration of the decomposition of survival structures would reduce the potential of primary inoculum.

Preventing the establishment of infection is perhaps the most interesting strategy for biocontrol of late blight. Detaining pathogen development after infection has occurred would be difficult, at least based on results presented so far. Application of control measures that could reduce the survival period, the effectiveness of the source of initial inoculum; prevent sporangia germination (germ tube formation) and/or zoospore germination should be evaluated in more detail. As a complement to biocontrol, activation of plant resistance should be effective.

Biological control can become an integral part of management programs aimed at controlling late blight in both conventional and organic cropping systems. In conventional cropping system, the association of biocontrol agents with fungicides may be an option for late blight control and also contribute to reduce selection pressure exerted by fungicides to increase populations of fungicide-resistant individuals. Another possibility is to combine biocontrol, fungicide, and forecast system to schedule applications of the biocontrol agent, fungicide or a mixture of the two. This approach was shown to be valid for management of *B. cinerea* in cucumber and tomato cultivated in greenhouses in Israel (Shtienberg and Elad 1997). Late blight forecast systems have been validated in many areas of the world and adapting their usage to a biocontrol agent should not be complicated. For organic production it seems risky to rely solely upon biological control and application of alternative products. To date, apparently no alternatives can assure effective late blight control under favorable conditions and a combination with copper-based fungicides is still required to manage the disease in many regions.

Regardless of the cropping system, alternative control of potato and tomato late blight should be implemented under a holistic approach. However, a continued search and screening of potential biocontrol agents is needed. We hope that the recent advancements in the field of metagenomics will contribute to the search for antagonists, especially those that are not easily cultivated under laboratory conditions but that could be capable of producing compounds effective against the pathogen. Finally, there should be more support for research oriented towards alternative methods to control tomato and potato late blight.

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REFERENCES

Abbasi PA, Riga E, Conn KL, Lazarovits G (2005) Effect of neem cake soil amendment on reduction of damping-off severity and population densities of plant-parasitic nematodes and soilborne plant pathogens. *Canadian Journal of Plant Pathology* **27**, 38-45

Ahn Y, Lee H, Oh H, Kim H, Lee Y (2005) Antifungal activity and mode of action of *Galla rhois*-derived phenolics against phytopathogenic fungi. *Pesticide Biochemistry and Physiology* **81**, 105-112

Arun K, Bhansali RR, Mali PC (2004) Raw cow's milk and *Gliocladium virens* induced protection against downy mildew of pearl millet. *International Sorghum and Millets Newsletter* **45**, 64-65

Avis T, Bélanger R (2001) Specificity and mode of action of the antifungal fatty acid cis-9-heptadecenoic acid produced by *Pseudozyma flocculosa*. *Applied and Environmental Microbiology* **67**, 956-960

Aylor DE (1986) A framework for examining inter-regional aerial transport of fungal spores. *Agricultural and Forest Meteorology* **38**, 263-288

Becktell MC, Daughtrey ML, Fry WE (2005) Epidemiology and management of petunia and tomato late blight in the greenhouse. *Plant Disease* **89**, 1000-1008

Bettiol W (1999) Effectiveness of cow's milk against zucchini squash powdery mildew (*Sphaerotheca fuliginea*) in greenhouse conditions *Crop Protection* **18**, 489-492

Bettiol W, Ghini R, Galvão JAH, Siloto RC (2004) Organic and conventional tomato cropping systems. *Scientia Agricola* **61**, 253-259

Burt S (2004) Essential oils: their antibacterial properties and potential applications in foods - a review. *International Journal of Food Microbiology* **94**, 223-253

Cao KQ, Forrer HR (2001) Current status and prosperity on biological control of potato late blight (*Phytophthora infestans*). *Journal of Agricultural University of Hebei* **24**, 51-58

Cao KQ, van Bruggen AHC (2001) Inihibitory efficacy of several plant extracts and plant products on *Phytophthora infestans*. *Journal of Agricultural University of Hebei* **24**, 91-99

Cao KQ, Wang ST, Forrer HR, Fried PM (2004) Inhibitory effects of Chinese medicinal plants against *Phytophthora infestans* on potatoes. *Regional Workshop on Potato Late Blight for East and Southeast Asia and the Pacific*, International Potato Center and the Ministry of Agriculture and Irrigation, Myanmar, Yezin Agricultural University, pp 45-53

Cao KQ, Wang ST, Kessler P, Fried PM, Forrer HR (2003) Potato late blight: A new approach for an outdoor screening of substitutes for copper fungicides. Krautfaulebekampfung im Bio-Kartoffelanbau ohne Kupfer? *Agrarforschung* **10**, 182

Chapman J (2003) *Stachybotrys chartarum* (*chartarum = atra = altemans*) and other problems caused by allergenic fungi. *Allergy and Asthma Proceedings* **24**, 1-7

Cho JY, Kim HY, Choi GJ, Jang KS, Lim HK, Lim CH, Cho KY, Kim JC (2006) Dehydro-alpha-lapachone isolated from *Catalpa ovata* stems: activity against plant pathogenic fungi. *Pest Management Science* **62**, 414-418

Clulow SA, Stewart HE, Dashwood EP, Wastie RL (1995) Tuber surface microorganisms influence the susceptibility of potato tubers to late blight. *Annals of Applied Biology* **126**, 33-43

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, Baider A, Ben-Daniel B, Ben-Daniel Y (2002) Fungicidal preparations from *Inula viscosa*. *Plant Protection Science* **38**, 629-630

Crosier W (1934) Studies in the biology of *Phytophthora infestans* (Mont.) de Bary. *Cornell University Agricultural Experiment Station Memoir* **155**, 40

Daayf F, Adam L, Fernando WGD (2003) Comparative screening of bacteria for biological control of potato late blight (strain US-8), using *in-vitro*, detached-leaves, and whole-plant testing systems. *Canadian Journal of Plant Pathology* **25**, 276-284

Deepa MA, Bai VN, Basker S (2004) Antifungal properties of *Pseudarthria viscida*. *Fitoterapia* **75**, 581-584

Diniz LP, Maffia LA, Dhingra OD, Casali VWD, Santos RHS, Mizubuti ESG (2006) Avaliação de produtos alternativos para controle da requeima do tomateiro. *Fitopatologia Brasileira* **31**, 171-179

Duke SO, Baerson SR, Dayan FE, Rimando AM, Scheffler BE, Tellez MR, Wedge DE, Schrader KK, Akey DH, Arthur FH, Lucca AJD, Gibson DM, Harrison Jr. HF, Peterson JK, Gealy DR, Tworkoski T, Wilson CL, Morris JB (2003) United States Department of Agriculture - Agriculture research service on natural products for pest management. *Pest Management Science* **59**, 708-717

Duncan JM (2003) Breeding to tackle blight without copper or GM. *Nature* **425**, 15

El-Sheikh M, El-Korany A, Shaat M (2002) Screening for bacteria antagonistic to *Phytophthora infestans* for the organic farming of potato *Alexandria Journal of Agricultural Research* **47**, 169-178

Fernandez-Acero F, Carbu M, Garrido C, Collado I, Cantoral J, Vallejo I (2006) Screening study of potential lead compounds for natural product-based fungicides against *Phytophthora* species. *Journal of Phytopathology* **154**, 616-621

Fernández-Pavía SP, Grünwald NJ, Díaz-Valasis M, Cadena-Hinojosa M, Fry WE (2004) Soilborne oospores of *Phytophthora infestans* in Central Mexico survive winter fallow and infect potato plants in the field. *Plant Disease* **88**, 29-33

Filippov A, Kuznetsova M (1994) Different influence of some biofungicides on dynamics of potato plant susceptibility to *Phytophthora infestans* (Mont) de Bary. *Mikologiya I Fitopatologiya* **28**, 64-69

Forbes GA, Landeo JA (2006) Late blight. In: Gopal JPKSM (Ed) *Handbook of Potato Production, Improvement, and Postharvest Management*, Haworth Press Inc., Binghamton, NY, pp 279-320

Fravel DR (2005) Commercialization and implementation of biocontrol. *Annual Review of Phytopathology* **43**, 337-359

Fry WE, Thurston HD, Stevenson WR (2001) Late blight In: Stevenson WR, Loria R, Franc GD, Weingartner DP (Eds) *Compendium of Potato Diseases* (2nd Edn), The American Phytopathological Society, St. Paul, USA, pp 22-23

Garita VS, Bustamante E, Shattock R (1998) Selección de antagonistas para el control biológico de *Phytophthora infestans* en tomate. *Manejo Integrado de Plagas (Costa Rica)* **48**, 25-34

Garita VS, Bustamante E, Shattock R (1999) Control microbiologico de *Phytophthora infestans* en tomate. *Manejo Integrado de Plagas (Costa Rica)* **51**, 47-58

Ghorbani R, Wilcockson SJ, Giotis C, Leifert C (2004) Potato late blight management in organic agriculture. *Outlooks on Pest Management* **15**, 176-280

Grünwald NJ, Sturbaum AK, Montes GR, Serrano EG, Lozoya-Saldaña H, Fry WE (2006) Selection for fungicide resistance within a growing season in field populations of *Phytophthora infestans* at the center of origin. *Phytopathology* **96**, 1397-1403

Guenthner JF, Michael KC, Nolte P (2001) The economic impact of potato late blight on US growers. *Potato Research* **44**, 121-125

Halama P, van Haluwin C (2004) Antifungal activity of lichen extracts and lichenic acids. *BioControl* **49**, 95-107

Haldar K, Kamoun S, Hiller NL, Bhattacharje S, van Ooij C (2006) Common infection strategies of pathogenic eukaryotes. *Nature Reviews Microbiology* **4**, 922-931

Hijmans RJ, Forbes GA, Walker TS (2000) Estimating the global severity of potato late blight with GIS-linked disease forecast models. *Plant Pathology* **49**, 697-705

Isman M (2000) Plant essential oils for pest and disease management. *Crop Protection* **19**, 603-608

Jiang J, Shi J, Zhao L, Zheng X, Lu J (2001) Resistance to *Phytophthora infestans* in potato induced with plant extracts. *Acta Phytopathologica Sinica* **31**, 144

Jindal KK, Singh H, Meeta M (1988) Biological control of *Phytophthora infestans* on potato. *Indian Journal of Plant Pathology* **6**, 59-62

Jones C, Samac DA (1996) Biological control of fungi causing alfalfa seedling damping-off with a disease-suppressive strain of *Streptomyces*. *Biological Control* **7**, 196-204

Jongebloed PHJ, Kessel GJT, Molhoek WML, Van der Plas CH, Fokkema NJ (1993) Biological control of *Phytophthora infestans* with compost extracts and selected bacterial antagonists In: Fokkema NJ, Köhl J, Elad Y (Eds) *Biological Control of Foliar and Post-Harvest Disease*, Proceedings of a workshop, IOBC/WPRS Bulletin, Wageningen, The Netherlands, pp 16-20

Judelson HS, Blanco FA (2005) The spores of *Phytophthora*: weapons of the plant destroyer. *Nature Reviews Microbiology* **3**, 47-58

Kamoun S, Smart CD (2005) Late blight of potato and tomato in the genomics era. *Plant Disease* **89**, 692-699

Kato M, Mizubuti ESG, Goodwin SB, Fry WE (1997) Sensitivity to protectant fungicides and pathogenic fitness of clonal lineages of *Phytophthora infestans* in the United States. *Phytopathology* **87**, 973-978

Kaya HK, Aguillera MM, Alumai A, Choo HY, de la Torre M, Fodor A, Ganguly S, Hazir S, Lakatos T, Pye A, Wilson M, Yamanaka S, Yang H, Ehlers R (2006) Status of entomopathogenic nematodes and their symbiotic bacteria from selected countries or regions of the world. *Biological Control* **38**, 134-155

Khanna KK, Chandra S (1976) Control of tomato fruit rot caused by *Fusarium roseum* with homeopathic drugs. *Indian Phytopathology* **29**, 269-272

Kim M, Choi G, Lee H (2003) Fungicidal property of *Curcuma longa* L. rhizome-derived curcumin against phytopathogenic fungi in a greenhouse. *Journal of Agricultural and Food Chemistry* **51**, 1578-1581

Kim Y, Lee C, Kim H, Lee H (2004) Anthraquinones isolated from *Cassia tora* (Leguminosae) seed show an antifungal property against phytopathogenic fungi. *Journal of Agricultural and Food Chemistry* **52**, 6096-6100

Koenig RL, Baker B (2002) U. S. National Organic Program Standards: implications for researchers. Published online by The American Phytopathological Society, St. Paul, MN. Available online: http://www.apsnet.org/online/feature/organic/

Kumar P, Poehling HM (2006) Persistence of soil and foliar azadirachtin treatments to control sweetpotato whitefly *Bemisia tabaci* Gennadius (Homoptera: Aleyrodidae) on tomatoes under controlled (laboratory) and field (netted greenhouse) conditions in the humid tropics. *Journal of Pest Science* **79**, 189-199

Lee HB, Kim Y, Kim JC, Choi GJ, Park SH, Kim CJ, Jung HS (2005) Activity of some aminoglycoside antibiotics against true fungi, *Phytophthora* and *Pythium* species. *Journal of Applied Microbiology* **99**, 836-843

Lee S, Park B, Kim M, Choi W, Kim H, Cho K, Lee S, Lee H (2001) Fungicidal activity of pipernonaline, a piperidine alkaloid derived from long pepper, *Piper longum* L., against phytopathogenic fungi. *Crop Protection* **20**, 523-528

Lee SJ, Kelley BS, Damasceno CMB, St. John B, Kim B, Kim BD, Rose JKC (2006) A functional screen to characterize the secretomes of eukaryotic pathogens and their hosts in planta. *Molecular Plant-Microbe Interactions* **19**, 1368-1377

Lehtinen A, Hannukkala A (2004) Oospores of *Phytophthora infestans* in soil provide an important new source of primary inoculum in Finland. *Agricultural and Food Science in Finland* **13**, 399-410

Li J, Chen G, Webster JM (1995) Antimicrobial metabolites from a bacterial symbiont. *Journal of Natural Products* **58**, 1081-1086

Lourenço Jr. V, Maffia LA, Romeiro RS, Mizubuti ESG (2006) Biocontrol of tomato late blight with the combination of epiphytic antagonists and rhizobacteria. *Biological Control* **38**, 331-340

Lozoya-Saldaña H, Coyote-Palma MH, Ferrera-Cerrato R, Lara-Hernandez ME (2006) Microbial antagonism against *Phytophthora infestans* (Mont) de Bary. *Agrociencia* **40**, 491-499

Mansingh A (2004) Limitation of natural pesticides: a challenge to scientists. In: Tan BK-H, Bay B-H, Zhu Y-Z (Eds) *Novel Compounds from Natural Products in the New Millenium* (Vol I) World Scientific, Singapore, pp 87-101

Mari M, Bertolini P, Pratella G (2003) Non-conventional methods for the control of post-harvest pear diseases. *Journal of Applied Microbiology* **94**, 761-766

Mekuria T, Steiner U, Hindorf H, Frahm JP, Dehne HW (2005) Bioactivity of bryophyte extracts against *Botrytis cinerea*, *Alternaria solani* and *Phytophthora infestans*. *Journal of Applied Botany and Food Quality* **79**, 89-93

Mizubuti ESG, Fry WE (2006) Potato late blight. In: Cooke BM, Jones DG, Kaye B (Eds) *The Epidemiology of Plant Diseases* (2nd Edn), Springer-Verlag, Dordrecht, pp 445-471

Mordue AJ, Nisbet AJ (2000) Azadirachtin from the neem tree *Azadirachta indica*: its action against insects. *Anais da Sociedade Entomológica do Brasil* **29**, 615-632

Ng KK, Webster JM (1997) Antimycotic activity of *Xenorhabdus bovienii* (Enterobacteriaceae) metabolites against *Phytophthora infestans* on potato plants. *Canadian Journal of Plant Pathology* **19**, 125-132

O'Herlihy EA, Duffy EM, Cassells AC (2003) The effects of arbuscular mycorrhizal fungi and chitosan sprays on yield and late blight resistance in potato crops from microplants. *Folia Geobotanica* **38**, 201-207

Obach BK (2007) Theoretical interpretations of the growth in organic agriculture: Agricultural modernization or an organic treadmill? *Society and Natural Resources* **20**, 229-244

Olanya O, Larkin R (2006) Efficacy of essential oils and biopesticides on *Phytophthora infestans* suppression in laboratory and growth chamber studies. *Biocontrol Science and Technology* **16**, 901-917

Pieterse CMJ, van Pelt JA, Verhagen BWM, Ton J, van Wees SCM, Leon-Kloosterziel KM, van Loon LC (2003) Induced systemic resistance by plant growth-promoting rhizobacteria. *Symbiosis* **35**, 39-54

Punja ZK (1997) Comparative efficacy of bacteria, fungi, and yeasts as biological control agents for diseases of vegetable crops. *Canadian Journal of Plant Pathology* **19**, 315-323

Quintanilla P, Rohloff J, Iversen TH (2002) Influence of essential oils on *Phytophthora infestans*. *Potato Research* **45**, 225-235

Rabea EI, Badawy MET, Stevens CV, Smagghe G, Steurbaut W (2003) Chitosan as antimicrobial agent: Applications and mode of action. *Biomacromolecules* **4**, 1457-1465

Raman KV, Grünwald NJ, Fry WE (2000) Promoting international collaboration for potato late blight disease management. *Pesticide Outlook* **11**, 181-185

Rani A, Bhat MN, Singh BP (2006) Efficacy of neem formulations against late blight of potato in sub-tropical plains. *Crop Research* **31**, 179-180

Rolim PRR, Brignani Neto F, Souza JM (2001) Ação de produtos homeopáticos sobre oídio (*Oidium lycopersici*) do tomateiro *Summa Phytopathologica* **27**, 129

Romero-Tabarez M, Jansen R, Sylla M, Lunsdorf H, Haubler S, Santosa DA, Timmis KN, Molinari G (2006) 7-*O*-Malonyl macrolactin A, a new macrolactin antibiotic from *Bacillus subtilis* active against methicillin-resistant *Staphylococcus aureus*, vancomycin-resistant enterococci, and a small-colony variant of *Burkholderia cepacia*. *Antimicrobial Agents and Chemoterapy* **50**, 1701-1709

Scheuerell S, Mahaffee W (2002) Compost tea: principles and prospects for plant disease control. *Compost Science and Utilization* **10**, 313-338

Schmutterer H (1990) Properties and potential of natural pesticides from the neem tree, *Azadirachta indica*. *Annual Review of Entomology* **35**, 271-297

Shtienberg D, Elad Y (1997) Incorporation of weather forecasting in integrated, biological-chemical management of *Botrytis cinerea*. *Phytopathology* **87**, 332-340

Silva HSA, Romeiro RS, Carrer R, Pereira JLA, Mizubuti ESG, Mounteer A (2004) Induction of systemic resistance by *Bacillus cereus* against tomato foliar diseases under field conditions. *Journal of Phytopathology* **152**, 371-375

Sogin ML, Silberman JD (1998) Evolution of the protists and protistan parasites from the perspective of molecular systematics. *International Journal for Parasitology* **28**, 11-20

Soylu EM, Soylu S, Kurt S (2006) Antimicrobial activities of the essential oils of various plants against tomato late blight disease agent *Phytophthora infestans*. *Mycopathologia* **161**, 119-128

Stephan D, Schmitt A, Carvalho SM, Seddon B, Koch E (2005) Evaluation of biocontrol preparations and plant extracts for the control of *Phytophthora infestans* on potato leaves. *European Journal of Plant Pathology* **112**, 235-246

Stevenson WR (1991) Late blight. In: Jones JB, Jones JP, Stall RE, Zitter TA (Eds) *Compendium of Tomato Diseases*, The American Phytopathological Society, St. Paul, USA, pp 17-18

Stewart HE, Wastie RL, Bradshaw JE (1993) The effect of moisture level of potting compost on the susceptibility of glasshouse-grown tubers to late blight. *Potato Research* **36**, 29-33

Stromberg A, Bostrom U, Hallenberg N (2001) Oospore germination and formation by the late blight pathogen *Phytophthora infestans in vitro* and under field conditions. *Journal of Phytopathology-Phytopathologische Zeitschrift* **149**, 659-664

Sturz AV, Lynch DH, Martin RC, Driscoll AM (2006) Influence of compost tea, powdered kelp, and Manzate (R) 75 on bacterial-community composition, and antibiosis against *Phytophthora infestans* in the potato phylloplane. *Canadian Journal of Plant Pathology-Revue Canadienne de Phytopathologie* **28**, 52-62

Sujkowski LS, Goodwin SB, Dyer AT, Fry WE (1994) Increased genotypic diversity via migration and possible occurrence of sexual reproduction of *Phytophthora infestans* in Poland. *Phytopathology* **84**, 201-207

Thuerig B, Binder A, Boller T, Guyer U, Jimenez S, Rentsch C, Tamm L (2006) An aqueous extract of the dry mycelium of *Penicillium chrysogenum* induces resistance in several crops under controlled and field conditions. *European Journal of Plant Pathology* **114**, 185-197

Tian M, Win J, Song J, van der Hoorn R, van der Knaap E, Kamoun S (2007) A *Phytophthora infestans* cystatin-like protein targets a novel tomato papain-like apoplastic protease. *Plant Physiology* **143**, 364-377

Tripathi P, Dubey NK (2004) Exploitation of natural products as an alternative strategy to control postharvest fungal rotting of fruit and vegetables. *Postхar-*

vest Biology and Technology **32**, 235-245

Turner RS (2005) After the famine: plant pathology, *Phytophthora infestans*, and the late blight of potatoes, 1845-1960. *Historical Studies in the Physical and Biological Sciences* **35**, 341-370

Unger C, Wilhelm I, Junger R, Thalmann R (2006) Evidence of induced resistance of tomato plants against *Phytophthora infestans* by a water extract of dried biomass of *Penicillium chrysogenum*. *Journal of Plant Diseases and Protection* **113**, 225-233

van Bol V, Decamps C, Maraite H, Peeters A (1993) Lutte contre *Phytophthora infestans* en culture de pomme de terre. Test des methodes utilisables en agriculture biologique. *Mededelingen van de Faculteit Landbouwwetenschappen, Universiteit Gent* **58**, 1315-1320

van Lenteren JC (2000) A greenhouse without pesticides: fact or fantasy? *Crop Protection* **19**, 375-384

Wang S, Wang X, Liu J, Cao K (2001) Screening of Chinese herbs for the fungitoxicity against *Phytophthora infestans*. *Journal of Agricultural University of Hebei* **24**, 101-107

Wang W, Ben-Daniel BH, Cohen Y (2004) Control of plant diseases by extracts of *Inula viscosa*. *Phytopathology* **94**, 1042-1047

Weltzien HC (1991) Biocontrol of foliar fungal diseases with compost extracts. In: Andrews JH, Hirano SS (Eds) *Microbial Ecology of Leaves*, Springer-Verlag, New York, pp 430-450

Xiao K, Kinkel LL, Samac DA (2002) Biological control of Phytophthora root rots on alfalfa and soybean with *Streptomyces*. *Biological Control* **23**, 285-295

Yan Z, Reddy MS, Ryu C, McInroy JA, Wilson M, Kloepper JW (2002) Induced systemic protection against tomato late blight elicited by plant growth-promoting rhizobacteria. *Phytopathology* **92**, 1329-1333

Yang X, Zhang Z, Yang H, Jian H (2001) Inhibition of metabolites from *Xenorhabdus nematophilus* against *Phytophthora infestans*. *Journal of Hebei Agricultural University* **24**, 65-68

Zwankhuizen M, Govers F, Zadoks J (2000) Inoculum sources and genotypic diversity of *Phytophthora infestans* in Southern Flevoland, the Netherlands. *European Journal of Plant Pathology* **106**, 667-680