Agronomy Research **4**(Special issue), 341–344, 2006

The question of metalaxyl resistance on late blight fungus in Estonia

E. Runno and M. Koppel

Jõgeva Plant Breeding Institute, Aamisepa St. 1, 48309 Jõgeva alevik, Estonia
(Phone:+3727762790; Fax: +3727766902; E-mail: Eve.Runno@jpbi.ee)

Abstract. Metalaxyl containing fungicides are commonly used by Estonian potato growers because of their efficiency in control of potato late blight. Metalaxyl-resistant isolates have become an important part of the Estonian population of *Phytophtora infestans*. The aim of the current study was to measure the frequency of metalaxyl resistance among the Estonian population of *Phytophtora infestans* and to test the efficacy of metalaxyl-based fungicides in controlling potato late blight. 138 isolates of *Phytophtora infestans* were collected from conventional, experimental and small scale growers' fields in different parts of Estonia in 2003-2005 and were assessed for resistance to metalaxyl. Metalaxyl-resistant, intermediate and sensitive isolates were recorded at frequencies of 37.1, 45.4 and 15.1%, respectively. There were differences in frequency of metalaxyl resistance between different locations, years and different types of potato cultivations. Metalaxyl-resistant isolates were predominant (66.7%) on conventional potato fields in 2004 and 2005. Metalaxyl- resistant strains were not detected among isolates collected from the field where fungicide containing metalaxyl was not used. The increased frequency of metalaxyl-resistant isolates was detected in 2003-2005. Trials of late blight control with fungicides containing metalaxyl were performed in the same period. Metalaxyl showed lowered efficacy only in conditions of extreme late blight pressure in 2004. Therefore metalaxyl could be effectively used for control of potato late blight a maximum of two times during the growing period.

Key words: metalaxyl resistance, *Phytophtora infestans*, potato late blight, late blight control

INTRODUCTION

Phenylamide fungicides are a class of systemic compounds including metalaxyl (Ridomil, oxadixyl, benalaxyl and ofurace) that show excellent protective, curative and eradicative antifungal activity and exclusively control diseases caused by Peronosporales (Schwinn & Staub, 1987). An increase of metalaxyl-resistant isolates of *Phytophtora infestans* was observed between 1981 and 1984, the period during which phenylamide fungicides were suspended in Ireland and The Netherlands. The proportion of metalaxyl-resistant isolates fluctuates from year to year and within seasons (Gisi & Cohen, 1996). In single years the values varied significantly (between 0-100%), with maximum levels at 1987–90 in most countries. In 1990s, resistance levels remained more or less stable in all European countries (Gisi & Cohen, 1996).

Fungicides containing Metalaxyl are commonly used by Estonian potato growers because of their efficiency in the control of potato late blight. Metalaxyl-resistant isolates have become an important part of the Estonian population of *P. infestans*.

The aim of the current study was to measure the frequency of metalaxyl resistance among the Estonian population of *P. infestans* and to test the efficacy of metalaxyl-based fungicides in controlling potato late blight.

MATERIALS AND METHODS

Collection and culture of isolates

Potato leaves naturally infected by *P. infestans* were collected during 2003–2005 from different parts of Estonia. Samples originated from conventional fields (Ingliste), small-scale farm fields (different locations) and experimental plots of Jõgeva Plant Breeding Institute. Blighted leaves were collected starting from the emergence of disease until the end of growing season. Isolation was carried out from one typical, single lesion per sample. Isolations were attempted by transferring a fragment of infected plant tissue at first to potato slices or tubers of susceptible cultivars (Bintje or Berber) without known R-genes. Rye agar was used for long term preservation of isolates of *P. infestans.*

Response to metalaxyl

In total, 138 samples were analysed for resistance to metalaxyl. Leaves of greenhouse grown 4-5 week-old plants of susceptible cultivars Bintje or Berber were used. Leaflets were floated in distilled water or in solutions of technical metalaxyl of 10,0 and 100,0 mg L^{-1} concentrations. Twenty microlitres of sporangial suspension was placed in the centre of each leaflet. Inoculated leaflets were incubated at 15°C for 7 days. Each isolate was tested in **3** replications. After incubation, the leaves were observed using a stereomicroscope to estimate fungal growth and sporulation. The isolates were rated as resistant if they sporulated on leaflets in 100 mg L^{-1} metalaxyl. Those sporulating in a metalaxyl concentration of 10 mg L^{-1} were rated intermediate, and those sporulating only in water (?) metalaxyl were rated sensitive.

Trials of late blight control

Field experiments were arranged at the Jõgeva Plant Breeding Institute in 2003–2005. Two potato varieties differing in resistance to late blight were used: moderately resistant Anti in 2003 and 2004 and susceptible Asterix in 2005. In all three years two different treatment regimes were used: untreated, where fungicides were not applied, and routine treatment where the first two sprayings used protectant fungicide Ridomil Gold MZ 68 WG (metalaxyl-M 40, mancozeb 640) 2.5 kg ha^{-1} at 10–12 day intervals starting from row closing, and the next treatments with eradicant fungicide Shirlan (fluazinam 500) 0.4 l ha^{-1} at 10 day intervals. Trials were laid out according to randomized block design in three replications. The plot size was four rows (7,5*0,7 m). To avoid possible neighbouring effects in border rows, late blight was estimated and the crop was harvested only from the two middle rows. Late blight infection was assessed twice a week according to 0–100% scale.

Statistics

The statistical analysis of frequencies of *P. infestans* in different categories was performed using MS Excel. The results of the analyses are expressed as probability

values (p) related to chi-square values. Late blight infection was expressed as an average percentage of infected leaf area.

RESULTS AND DISCUSSION

Metalaxyl-resistant, intermediate and sensitive isolates were recorded as 37.1%, 45.4% and 15.1% respectively. Statistically significant differences ($P < 0.001$) in the frequency of resistance to metalaxyl were observed between locations (Fig. 1), potato growing field types and years. The proportion of metalaxyl-resistant isolates from different locations ranged from 0 to 67% (Fig. 1). Metalaxyl-resistant isolates were predominant among isolates collected from the experimental field in 2004 (66,7%) and where metalaxyl was used on conventional fields in 2004 and 2005 (60%).In 2004 no metalaxyl-resistant strains were detected among isolates collected from the field (Naha) where fungicide containing metalaxyl was not used. As might be expected, insensitivity was more often found in conventional fields, where metalaxyl was commonly applied, than in small-scale growers` fields where fungicides were rarely used. The increase of frequency of metalaxyl-resistant isolates was detected from 2003-2005. Metalaxyl-resistant isolates were less frequent among isolates collected in 2003 than in 2004 and 2005. Even though the proportion of metalaxyl-resistant isolates fluctuates from year to year, there is strong evidence- that the quantity of metalaxyl-resistant isolates has increased compared with previous results (Runno et al., 2005). The reason for the increase could be the more consistent use of fungicides containing metalaxyl. As long as metalaxyl is used according to the recommendations in Estonia it is unlikely that metalaxyl-resistant isolates will completely displace sensitive isolates.

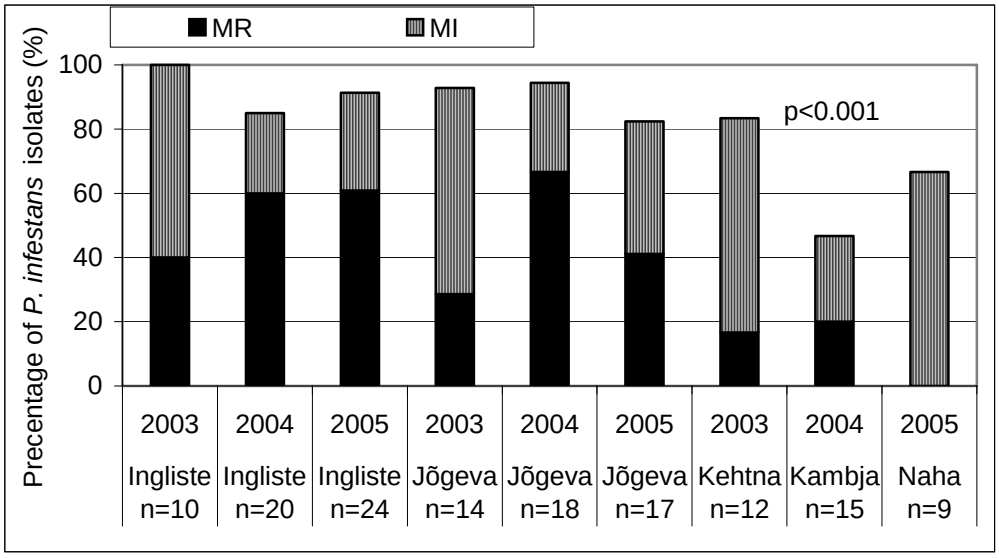

Fig. 1. Metalaxyl resistance[a] among isolates of *Phytophthora infestans* from different locations in Estonia during 2003–2005.
[a]MR, metalaxyl resistant; MI, intermediate metalaxyl sensitive.

Several reports suggest that resistance is unstable in the population and is selected anew each year, increasing steadily during the season and declining over winter (Gisi & Cohen, 1996).

Trials of late blight control with fungicide containing metalaxyl Ridomil Gold MZ 68 WG were performed in the same period. Metalaxyl-continaing fungicide was applied 2 times in all three years. The foliage of untreated plots was completely destroyed by the end of the growing season in all trial years. Metalaxyl provided high protection in 2003 and 2005, but showed lowered efficacy only in conditions of extreme late blight pressure in 2004 when the foliage was infected while metalaxyl was in use (Table 2). The trial results show that in spite of the occurrence of resistant strains the use of fungicides consisting of metalaxyl is still effective. Therefore metalaxyl could be used effectively for control of potato late blight a maximum of two times at the beginning of the fungicide treatments. By following the application rules for metalaxyl fungicides it is possible to restrict the development of metalaxyl-resistant strains and thus avoid the reduction of efficacy of the fungicide.

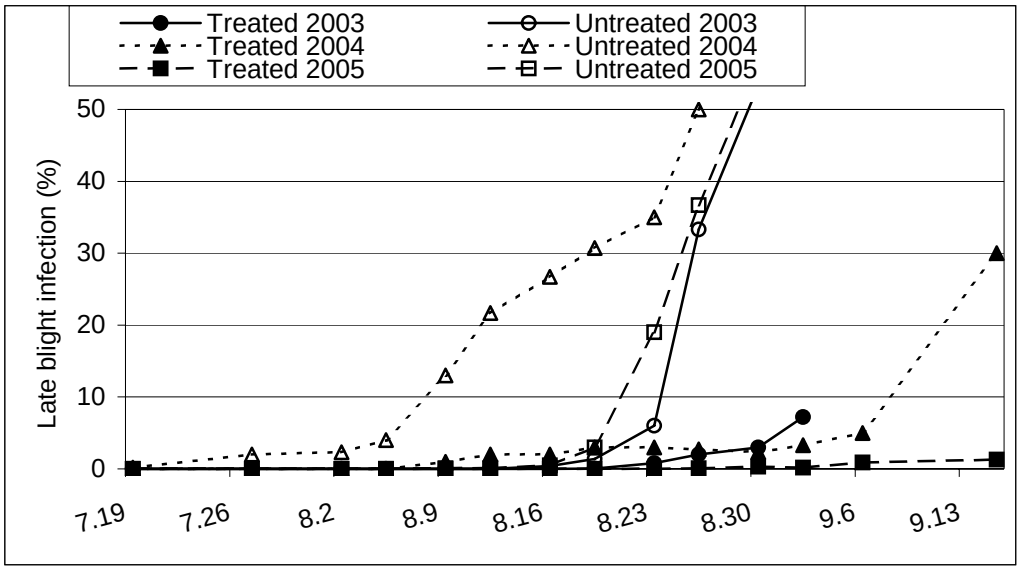

Fig. 2. Potato late blight infection assessments during 2003–2005.

ACKNOWLEDGEMENTS. The study was supported by the Estonian Science Foundation grants No 4734 and 6098.

REFERENCES

Gisi, U. & Cohen, Y. 1996. Resistance to phenylamide fungicides: a case study with *Phytophthora infestans* involving mating type and race structure. *Annual Review of Phytopathology* **34**, 549–72.

Schwinn, F.J. & Staub, T. 1987. Phenylamides and other fungicides against Oomycetes. In Lyr, H. (ed.): *Modern Selective Fungicides*. Jena: VEB Gustav Fischer**,** pp. 259–73.

Runno, E., & Koppel, M. 2005. *Phytophthora infestans* Eesti isolaatide metalaksüülitundlikkus ja paarumistüübid. *Agronoomia 2005. Teadustööde kogumik* **220**, 168–170 (in Estonian).