Kenyan cut flowers as a pathway for the introduction of plant pests to the UK

M G Palmieri, K Dehnen-Schmutz, J M Touza Montero, C Perrings
Environment Department, University of York, YO10 5DD, UK
Email: gianna_palmieri@hotmail.com

INTRODUCTION

The cut flower trade has followed the general trends in international trade in terms of increasing volumes (AIPH, 2004), and this has exacerbated the risk of introducing plant pests to importing countries (Kiritani & Yamamura, 2003; Work *et al.*, in press). The aim of this study is to enhance the understanding of the factors that favour the introduction of plant pests as a subcategory of alien invasive species through the cut flower trade. The analysis is applied to the case of the cut flower trade between Kenya (the exporting country) and the UK (the importing country) for the last eight years. Kenya is one of the largest suppliers of cut flowers to the EU (CPI, 2002), and the UK is the second-largest importer of Kenyan cut flowers (Thoen *et al.*, 1999).

DATA

Monthly data on plant pests detected by Her Majesty's Customs and Excise (HMCE) on cut flowers imported into the UK from Kenya were provided by Central Science Laboratory (CSL), UK for the period January 1996 to December 2003. Observations on *Chrysanthemum*, *Dendranthema* and *Euphorbia* were excluded because it was not possible to differentiate between imported cuttings and cut flowers. Data on volumes and values of cut flowers imported were obtained from Eurostat; information on exchange rates were also collected. Precipitation rates and temperatures in Kenya were obtained from the National Oceanic and Atmospheric Administration (NOAA). Finally, data on the monthly number of inspections in cut flowers by HMCE were also collected. Here, we explore the potential relationship between the number of pest interceptions in the importing country, trade-related variables (imported volume, prices and exchange rate), local environmental conditions in the exporting country and detection effort.

RESULTS AND DISCUSSION

The volume of cut flowers imported to the UK from Kenya has tripled in the period analysed. However, the inspection effort has not increased commensurately; in fact, it has declined from more than 500 in 1996 to just over a 100 inspections seven years later (Figure 1). There was a negative relationship between volume and detection effort (Spearman correlation coefficient $= -0.74$, $P < 0.001$) (Figure 1). The number of interceptions per year has remained relatively steadily during the period studied. Thrips (Thysanoptera) accounted for approximately 30% of the total interceptions. A stepwise regression analysis showed that only rainfall in Kenya and detection effort significantly influence the number of interceptions, although their explanatory power is very small ($R^2 < 0.15$). This seems to imply that local environmental conditions in Kenya may affect the likelihood of imports being infested by plant pests. This variable may also capture the seasonal character of the

interceptions as these increase during the European winter when UK cut-flower production is low. The relationship between inspections and interceptions should be considered when deciding upon the optimal level of inspection effort. These results are merely exploratory, and further statistical analysis is necessary to investigate, in greater depth, the links between socio-economic variables and the introduction of plant pests.
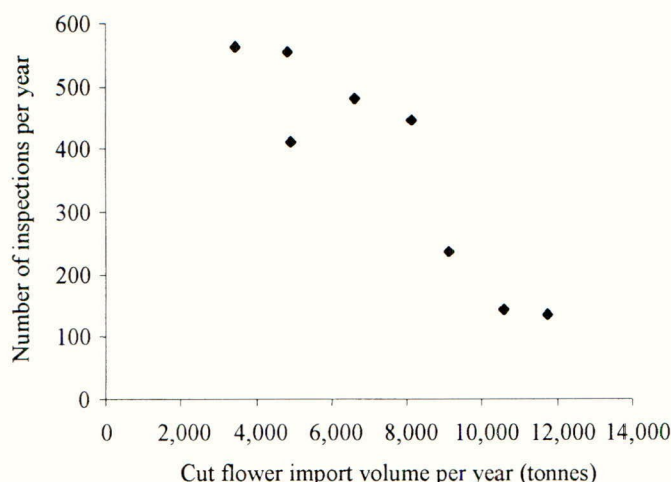

Figure 1. Relationship between cut flower import volume and number of inspections per year, from 1996 to 2003.

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Alien insect and mite pests introduced to Italy in sixty years (1945-2004)

G Pellizzari, L Dalla Montà
Dip. Agronomia Ambientale, Viale Università 16, 35020 Legnaro, Italy
Email: giuseppina.pellizzari@unipd.it

V Vacante
Dip. Agrochimica-Agrobiologia, Piazza S. Francesco di Sales 4, 89061 Gallina, Italy

INTRODUCTION

The incidental introduction of exotic phytophagous insects and mites has became quite a common event in Italy, owing to intensive commercial exchanges of plants and goods and an ever-increasing tourist traffic. Furthermore, the range of Italian climatic parameters allows the establishment of subtropical species in the southern regions and of northern species in the temperate climate of North and Central Italy. Several alien species are recorded only in greenhouses. To find out how many phytophagous exotic species of phytophagous insects and mites have been recorded as incidentally introduced to Italy, and how many of these have succeeded in acclimatizing to Italian conditions, a selected bibliography over a period of 60 years (from 1945 to 2004) has been checked (Pellizzari & Dalla Montà, 1997; Pellizzari & Vacante, 2005). This work has led to the compilation of chronological lists in which the exotic, introduced species are grouped according to their host plants.

RESULTS AND DISCUSSION

In total, from 1945 to 2004, 162 exotic pests have been introduced to Italy. Of these, about 130 are presently acclimatized and exhibit a different distributional range according to their climatic requirements and host plants. Most are pests of ornamentals (79 species), woody plants (38 species), *Citrus* (16 species), horticultural crops (15 species), fruit trees and grapevine (14 species). Most of the introduced species are Hemiptera (64%) (mainly aphids and scale insects), followed by Coleoptera (12%), Lepidoptera (7%), Diptera (6%), Thysanoptera (3%), Hymenoptera (2%); mites make up 6% of the introduced pests. The majority of these alien pests have come from the Americas (37%), Asia (29%), Africa (14%) and Australia (6%). In some cases Italy has been the first European focus of an exotic pest that has later expanded towards neighbouring countries and, in some cases, throughout Europe: e.g. the psyllid *Acizzia jamatonica*, the wax scale *Ceroplastes japonicus*, the lace bug *Corythucha ciliata*, the planthopper *Metcalfa pruinosa*, and the moths *Parectopa robiniella* and *Phyllonorycter robiniellus*. In other cases, some of the aliens have reached Italy from previously infested European countries: e.g. the moth *Cameraria ohridella* from Austria and Slovenia. Further, several alien insects that could represent a threat for other European countries have recently become established in Italy. These are: the Asian ambrosia beetle *Xylosandrus crassiusculus* (a pest of forest trees), the oriental chestnut gall wasp (*Dryocosmus kuriphilus*), the locust gall midge (*Obolodiplosis robiniae*), the mealybug *Pseudococcus comstocki* (a pest of fruit trees), the oak lace bug (*Corythucha arcuata*) (currently spreading from its first focus by about 10 km/year) and the

longhorn beetle *Anoplophora malasiaca* (a quarantine species unfortunately established in North Italy). A list of the exotic insects introduced to Italy in the last six years is provided in Table 1, together with information on their present distribution.

Table 1. Alien insects accidentally introduced to Italy (1999–2004).

Species	Order/Superfamily	Host plant	Origin	Year
Echinothrips americanus (g)	Thysanoptera	polyphagous	N. America	1999
Lopholeucaspis japonica (i)	Coccoidea	polyphagous	Far East	1999
Bradinothrips musae (g)	Thysanoptera	*Spathiphyllum*	C. America	1999
Monelliopsis pecanis (al)	Aphdoidea	*Carya*	N. America	1999
Phenacoccus solani (g)	Coccoidea	Cycadaceae	?	1999
Entaspidiotus lounsbouryi (al)	Coccoidea	*Mesembryant*	S. Africa	1999
Scyphophorus acupunctatus (g)	Coleoptera	*Beaucarnea*	C. America	2000
Ophelinus eucalypti (aw)	Hymenoptera	*Eucalyptus*	Australia	2000
Corythucha arcuata (al)	Hemiptera	*Quercus*	N. America	2000
Neotoxoptera formosana (g)	Aphidoidea	Liliaceae	Far East	2000
Platypus mutatus (al)	Coleoptera	*Populus*	S.America	2000
Leptoglossus occidentalis (al)	Hemiptera	Coniferae	N. America	2001
Ceroplastes ceriferus (al)	Coccoidea	polyphagous	Far East	2001
Anoplophora malasiaca (al)	Coleoptera	polyphagous	Far East	2001
Cerodontha unisetiorbita (aw)	Diptera	bamboos	Japan	2001
Illinoia liriodendri (aw)	Aphidoidea	*Liriodendron*	N. America	2001
Stephanitis takeyai (aw)	Hemiptera	polyphagous	Japan	2001
Acizzia jamatonica (aw)	Psylloidea	*Albizzia*	Far East	2002
Dryocosmus kuriphilus (al)	Hymenoptera	*Castanea*	China	2002
Josephiella microcarpae (al)	Hymenoptera	*Ficus*	Asia	2002
Paysandisia archon (aw)	Lepidoptera	palms	S. America	2003
Cacopsylla fulguralis (al)	Psylloidea	*Elaeagnus*	Far East	2003
Obolodiplosis robiniae (aw)	Diptera	*Robinia*	N. America	2003
Lissorhoptrus oryzophilus (al)	Coleoptera	*Oryza sativa*	N. America	2004
Acanalonia conica (al)	Hemiptera	polyphagous	N. America	2004
Xylosandrus crassiusculus (al)	Coleoptera	trees	Far East	2004
Pseudococcus comstocki (al)	Coccoidea	polyphagous	Far East	2004
Fiorinia pinicola (al)	Coccoidea	*Pittosporum*	Far East	2004

al = acclimatised, localised; aw = acclimatised, widespread;
g = greenhouse; I = interception.

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Gone with the wind: is it always true for invasive plants and, if not, why not?

S Petrovskii

Russian Academy of Science, Nakhimovsky Prospekt 36, Moscow 117218, Russia
Email: spetrovs@sio.rssi.ru

R P Blackshaw

University of Plymouth, Drake Circus, Plymouth, Devon PL4 8AA, UK

INTRODUCTION

Factors that can either speed up or slow down the spread of invasive species are of considerable current interest, and their identification and proper understanding are expected to lead to efficient management and control. In order to reveal the relevant factors, the main mechanisms of species transport must first be identified. In the case of plant species, it is widely accepted that local seed-spread is attributable to the impact of wind (Okubo & Levin, 1989). However, estimates based on this mechanism often appear to be at variance with field data. In particular, wind-driven seed dispersal would imply that invasive plants are strongly affected by wind pattern, which is not always the case. This apparent discrepancy is sometimes interpreted as the manifestation of certain specific features of wind mixing (Clark *et al.*, 1998), but it may also indicate that some additional or alternative mechanisms of seed transport have not been taken into account. In this paper, we assume that, at least in the case of some tree species, such a mechanism can be avian-based: birds eat seeds, but they do not assimilate all of them so that a part of the seed population is then egested at a different location. In order to study the impact of this mechanism, we apply a simple conceptual mathematical model, and show that avian-based dispersal can significantly modify the pattern of invasion by plant species.

MATHEMATICAL MODEL

We consider a plant species A, presumably an arbor, described by its population density $a(x,t)$. We assume that species A dwells in an unbounded, one-dimensional domain, with the following conditions at infinity: $a(\infty,t) = K$, $a(-\infty,t) = 0$. These conditions imply that the species range has a boundary situated at a certain x_0. Species invasion or colonization takes place if, with each new generation, x_0 moves to the left. Redistribution of species A in space takes place due to dispersal of its seeds which, having neglected the impact of other species, happens through wind mixing.

Next, we consider an avian species B, described by its population density $b(x,t)$. We assume that seeds of plant species A provide an essential source of food for avian species B. The relation between A and B is, therefore, one of the 'resource-consumer' or 'predator-prey' type. We then make use of the observation that a higher density of resource (or prey) often leads to a higher density of consumer (or predator), and assume that, within the range of species A, their densities are linearly related:

$$b(x,t) = \eta a(x,t). \qquad (1)$$

Apparently, dispersal of avian species is not affected by wind to the same extent as that of plant species, because of a bird's ability of self-motion. Moreover, some avian species are known to exhibit two-mode dispersal: a short-distance dispersal due to everyday round-about motion (which we assume to be random), and a long-distance dispersal due to small-scale migrations by individuals or groups of individuals (Hengeveld, 1989). The colonies created by groups of migrating birds are outside of the main range of species A, and thus relation (1) would not seem to be applicable. However, before migrating, the birds would have been feeding on seeds of A, and it is likely that some of these seeds would be transported to the place where a new colony is formed. Moreover, the more birds that have migrated the more seeds are transported; thus, we can assume that relation (1) also holds outside the species A range, albeit with a different coefficient.

The issue of species A invasion is, therefore, reduced to the issue of species B invasion. In order to describe the dynamics of population B, we use the model earlier developed by Petrovskii & Li (2003):

$$\frac{\partial b(x,t)}{\partial t} + \frac{\partial}{\partial x}\left[S(b)b\right] = D\frac{\partial^2 b}{\partial x^2} + F(b) \tag{2}$$

where D is the species diffusivity responsible for the short-term dispersal, $S(b)$ describes the intensity of small-scale density-dependent migrations responsible for the long-distance dispersal, and $F(b)$ describes local population growth of species B.

RESULTS

For a biologically reasonable parameterization of $S(b)$ and $F(b)$, equation (1) appears to be exactly solvable. Its solution describes the propagation of a population front, which apparently corresponds to the advance of the species range boundary. We calculate the speed of the front and show that, when seed dispersal is enhanced by an avian-based mechanism, the direction of the propagation front does not necessarily coincide with the direction of the wind. Moreover, the speed of the front appears to be several times greater, compared with that observed in the case of simple wind-driven dispersal. The latter result can, in some cases, account for the phenomenon of rapid plant invasion, both in ongoing ecological observations and in historical data.

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Investigations on the occurrence of grape phylloxera (*Viteus vitifoliae*) in Austrian viticulture

H Reisenzein

AGES, Spargelfeldstrasse 191, A-1226 Vienna, Austria

Email: helga.reisenzein@ages.at

INTRODUCTION

In Europe, grape phylloxera (*Viteus vitifoliae*) (Homoptera: Phylloxeridae), is an invasive insect on grapevines, and is listed as a quarantine pest (Council Directive 2000/29/EC, Annex II/AII). During the last half of the 19th century the pest was introduced into Europe and destroyed almost the entire European viticulture. In Austria, the first occurrence of phylloxera was documented in 1872 and, within a few years, the pest destroyed nearly all Austrian vine-growing. Phylloxera damage could be minimized by the mandatory use of phylloxera-tolerant rootstocks, one of the first integrated plant-protection strategies, the grafting of European and American *Vitis* species providing effective control. Because of the natural resistance of American *Vitis* species, the subterranean development of the pest is reduced and there is no formation of nodosities (galls on feeder roots) or tuberosities (galls on mature roots) as a result of phylloxera feeding. Resistant rootstocks have worked well for more than 100 years. While foliar phylloxera was sometimes observed on wild American *Vitis* species, and on rootstocks derived from them, a massive development of the root form of the insect did not appear in Austria. In the middle of the 1990s a new phylloxera outbreak was reported in European and American vine-growing areas. In Austria, the most recent outbreak of the pest was discovered in 2000 (Polesny & Reisenzein, 2000). The present study focused on the spread and population dynamics of the root-infesting form (= radicicolae) form of the pest in different Austrian vine-growing areas.

MATERIALS AND METHODS

For the documentation of spreading of the radicicolae of phylloxera, 70 selected vineyards in various Austrian vine-growing areas were surveyed. Root samples were taken from the upper 30 cm of the soil horizon around the stems (sample size: 5 grapevines). Root infestation was estimated, using five categories (classes): class 0 = roots visually healthy; class 1 = few old nodosities on feeder roots; class 2 = low infestation, small new nodosities; class 3 = medium infestation, with nodosities 2–3 mm in length; class 4 = high infestation, with massive formation of nodosities > 3 mm in length, plus some tuberosities on mature roots. The assessment classes were used to calculate the frequency and intensity of infestations.

In order to investigate the population dynamics of the radicicolae, survey of eleven vineyards in different vine-growing areas of Lower Austria, Styria and Burgenland was conducted. One vineyard was planted with self-rooted *Vitis vinifera* (cv. Otello), the others were planted on Kober 5BB, 5C and SO4 rootstocks. Phylloxera populations were monitored by sampling randomly selected grapevines at each study site. Ten root samples were collected per vineyard, from April to September, six times over the 3-year study period (2000 to 2002). The samples were examined and infestation levels estimated, using the evaluation scheme.

RESULTS

Infestations with radicicolae were detected in 97% of the sites examined, and high infestation densities, and the formation of nodosities on feeder roots of phylloxera-tolerant rootstocks (Fercal, Kober 5BB, 5C, SO4), were observed in nearly all Austrian vine-growing areas. In 2001, the level of phylloxera infestation was 'low', with an average of c. 7%; infestation frequency varied from < 1% to > 20%. The next vegetation period (2002) showed an increase in populations at each site. The average infestation intensity was more than five-fold greater than in 2001. Infestation frequency ranged from 11 to 72%, with the majority of sites exhibiting rates of 35%. At some locations infestation rates in July and August reached 100%. In the third year of observation (2003), the level of infestation decreased, the average infestation intensity being c. 26%.

There were significant differences in phylloxera infestations at the investigated sites, and no reductions in the growth of grapevines, yield losses or evidence of grapevine decline due to phylloxera infestations were observed during these field studies. The root rot fungus *Roesleria hypogea* was frequently detected in infested root samples and, in some of these vineyards, slow grapevine decline was observed.

DISCUSSION

In the current study, the root form of phylloxera has been observed in Austrian vineyards since 2000. It can be assumed that the massive development of the pest started in that year, because previous studies on pedobiocoenosis in different Austrian vine-growing areas had not revealed any phylloxera infestation on roots. The widespread infestation of Austrian vineyards with the root-infesting form of phylloxera was demonstrated. The renewed occurrence of phylloxera could be explained by climatic changes over the past ten years. A pathogenic risk assessment of phylloxera revealed no direct damage to the grapevines in the present study; secondary pathogens seem to be more dangerous. The pathogenic fungus *Roesleria hypogea*, for example, which causes slow grapevine decline, was frequently detected in phylloxera-infested root samples. In future work, emphasis should be placed on various plant-parasitic nematodes, which can be implicated in phylloxera-related damage (Hoschitz & Reisenzein, 2004).

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Phenolic compounds in the leaves of alien and native *Impatiens* plants

B Šerá, N Vrchotová, J Tříska
Institute of Landscape Ecology, Na Sádkách 7, 370 05 České Budějovice, Czech Republic
Email: sera@jcu.cz

INTRODUCTION

The occurence of naphthoquinones was demonstrated in the aerial parts of various *Impatiens* species (*I. balsamina, I. capensis, I. glandulifera, I. noli-tangere* and *I. parviflora*), the amount present in *I. glandulifera* being several times greater than in the other species (Lobstein *et al.*, 2001). Naphthoquinones (e.g. 2-methoxy-1,4-naphthoquinone) are alleged to have allelochemical and pesticide characteristics (www.ars-rin.gov/duke), which is interesting mainly from the perspective of invasive behaviour of this alien species. *I. glandulifera* comes from the Himalayas and it is mainly in its significant height (1–3 m) and in the red-purple colour of blossoms that it differs from other species.

MATERIALS AND METHODS

Leaves were collected in only one type of biotope, so that the growth conditions were the same, either on 8 July 2003 (*I. noli-tangere* and *I. parviflora*) or on 6 August 2003 (*I. glandulifera*). Leaves (several per species) were put into bottles at the collecting site, and immediately covered with 80% methanol. These biomass samples were kept at laboratory temperature and in darkness for 2 weeks, then stored in the refrigerator. After 5 months the samples were centrifugated and the supernatants were stored at –20°C. Phenolic substances were separated using the HPLC method on a Phenomenex Luna C 18(2), 3μm, 2 × 150 mm colon. Separation was done at 25°C in a water-acetonitrile gradient, with the addition of 0.15% trifluoracetic acid. During chromatogram evaluation, emphasis was placed on flavonoids, phenolic acids and naphthoquinone derivates. As species samples were always mixed, the resulting measurements represent the whole population in a locality.

RESULTS AND DISCUSSION

In the extracts of *I. glandulifera* leaves, a dominant substance with a UV spectrum similar to quercetin (substance no. 1, Figure 1) was determined. This substance was also present in large amounts in the two other *Impatiens* species tested, but was not dominant. In *I. noli-tangere* and *I. parviflora*, peak no. 2 (Figure 1) was dominant; this compound has a UV spectrum similar to caffeic acid. In *I. glandulifera*, however, substance no. 2 was present only imperceptibly. We have not yet managed to determine these substances positively. Naphthoquinone was found in both *I. glandulifera* and *I. noli-tangere*. However, naphthoquinones were not dominant substances in the extracts. It is probable that naphthoquinones are accumulated mainly in other parts of the plants than leaves; for example, our results show that they are dominant substances in root extracts (unpublished data). The chromatographic profiles of leaf extracts of *I. noli-tangere* and *I. parviflora* were very similar to each other, and very different from the profile of *I. glandulifera*. These results correspond to the degree of kingship between the species of *Impatiens* studied (*I. noli-tangere* and *I. parviflora* being closer to one another than to *I. glandulifera*).
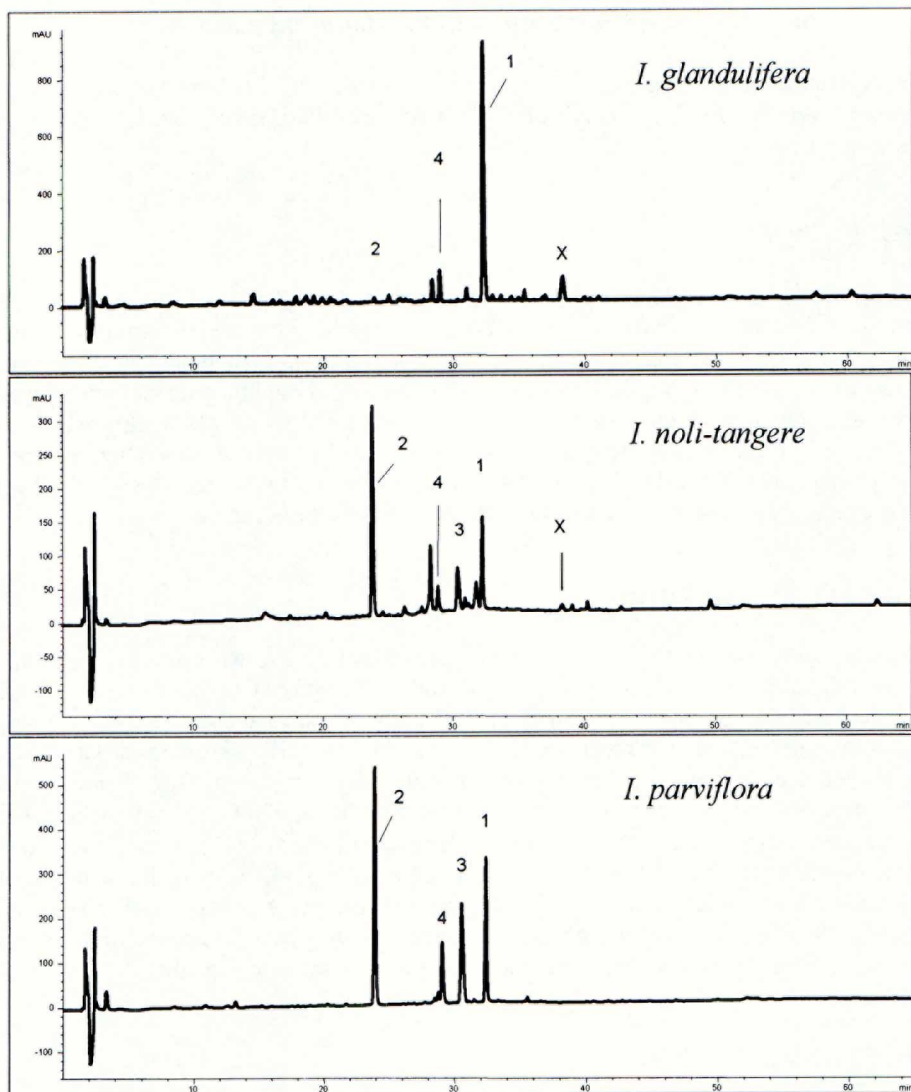
Figure 1. Chromatograms of phenolic substances extracted from the leaves of *Impatiens*. Peak labelling: 1 & 4 = substances with UV spectrum similar to quercetin spectrum; 2 & 3 = substances with UV spectrum similar to caffeic acid; X = naphthoquinone derivative.

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Impact of the invasive weed *Ageratum conyzoides* in the Shivalik Ranges of the north-western Himalayas, India

H P Singh, D R Batish, R K Kohli, V Arora, S Kaur
Department of Botany, Panjab University, 160014 Chandigarh, India
Email: hpsingh_01@yahoo.com

INTRODUCTION

In India, the north-western Himalayas are well-known for their rich floral diversity. However, during the last two decades a number of exotic weeds have invaded the area, thereby greatly threatening the natural vegetation (Kohli *et al.*, 2005), causing irreversible damage to the structure and dynamics of natural communities. The allelopathic interference of these invading weeds also plays an important role in their invasiveness (Callaway & Aschehoug, 2003). Billy goat weed (BGW) (*Ageratum conyzoides*) (Asteraceae) is an aromatic, invasive and exotic weed from tropical America that quickly encroaches upon any given area. BGW possesses a number of ecological strategies, such as fast growth rate, quick regenerative and reproductive potential, and greater tolerance/adaptability, helping it to form monocultural stands in the Shivalik Ranges of the north-western Himalayas. The weed has spread extensively in the Shivaliks, occupying various habitats (forests, plantations, agricultural fields, wastelands, grasslands) and now greatly affects the landscape (Kohli *et al.*, 2004). However, a quantitative assessment of the native vegetation following BGW invasion in different habitats have not been done. A study was therefore conducted to assess the status of vegetation in various habitats (such as grasslands, a plantation, riparian regions and wastelands invaded by BGW in the Shivaliks. The studies were further extended to explore the effect of BGW residues on the growth of native species.

MATERIALS AND METHODS

Various habitats invaded by BGW were surveyed for the number of species, plant density and vegetation biomass, using quadrats. Vegetation in nearby areas devoid of BGW was also assessed (as a control). Under natural conditions, large quantities of residues are formed in infested areas and, following decomposition, these become mixed with the soil. To simulate this, residues collected from the BGW-infested areas were mixed into soil (1 g/100 g soil) from BGW-free areas. Soil was also collected from invaded and uninvaded areas. The impact of all these soils was studied on the early seedling growth and biomass accumulation of *Cassia tora* – a sensitive and native species found in association with BGW. Residues were also analyzed for the presence of phenolic allelochemicals, as per Swain & Hillis (1959).

RESULTS AND DISCUSSION

Compared with controls, native vegetation in BGW-infested habitats was significantly reduced, including numbers of species, plant density and biomass (Table 1), especially in wastelands and grassland, indicating that invasion by BGW greatly affects the structure and composition of natural vegetation and also reduces plant diversity.

Table 1. Status of vegetation in different habitats invaded by *Ageratum*.

Habitat	Species types	Density (plants/m^2)	Biomass (g/m^2)
Uninvaded control	45	390.4	978.5
Invaded grasslands	7	137.8	363.9
Invaded plantation of *Dalbergia sissoo* (a native tree)	11	105.7	483.5
Invaded riparian zone	9	115.2	465.6
Invaded wastelands	7	128.7	888.4

All values significantly different from control at $P < 0.01$.

Table 2. Effect of *Ageratum*-infested and amended soil on the growth of *Cassia tora*.

Treatment	Seedling length (cm)	Dry weight (mg/seedling)
Ageratum-free soil	10.94	24.1
Ageratum-infested soil	9.41*	21.9*
Ageratum residue-amended soils	6.68*	19.7*

* Means significantly different from uninfested control at $P < 0.01$

Following invasion by BGW, large quantities of plant residues accumulate, which eventually decompose and become mixed with the soil. It is possible that such residues could affect the growth of sensitive native plants (such as *C. tora*); data presented here (Table 2), indicate that this is so, whether plants are grown in soil from BGW-infested areas or grown in soil amended with BGW residues (including rates as low as 1 g/100 g soil). This suggests that the BGW residues might be contributing biomolecules to the soil that interfere with plant growth. These could possibly be phenolics, which are well-known growth inhibitors (Mizutani, 1999). Upon release, phenolics accumulate in soil and have a growth-retarding effect. Here, the BGW residues were found to contain 470 mg phenolics (per 100 g). It is concluded that BGW invasion drastically affects native vegetation and that residues from the weed, following the release of phenolics, play an important role in this process.

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Spread of horse chestnut scale (*Pulvinaria regalis*) in Germany

P Trierweiler, H Balder

University of Applied Sciences Berlin, Luxemburger Strasse 10, D-13353 Berlin, Germany
Email: balder@tfh-berlin.de

INTRODUCTION

In Europe, the horse chestnut scale (*Pulvinaria regalis*) (Homoptera: Coccidae) is an invasive species, believed to be of Asian origin. The insect was first found in Europe (in Paris, France) in 1968, and has since continued to spread. The first record for Germany was in 1989, since when the pest has spread to most parts of Germany and central Europe. At present, 65 plant species from 24 families are known to be hosts, with particularly important infestations in urban areas occurring on *Acer*, *Aesculus* and *Tilia*. The main means of the spread lies in the transportation of prunings, following tree care, or via the transportation of infested nursery trees. Furthermore, the young, non-sedentary nymphs are able to spread from tree to tree with the help of the wind. It is also possible that eggs and nymphs may be spread by birds. Primarily, horse chestnut scale damages host plants through the extraction of plant juices, and also by contaminating leaves with honeydew (upon which sooty moulds develop), resulting in a decrease in the rate of photosynthesis. The ornamental value of heavily infested plants is decreased considerably, and the immediate surroundings are persistently contaminated. Currently, there are no suitable plant-protection measures (Arnold, 2003; Schröder *et al.*, 2003).

METHODS AND RESULTS

In order to determine the present situation concerning the spread, infestation levels and management of infestations of horse chestnut scale in Germany, a nationwide Plant Protection Service survey was done in 2004, within some cities as well as in nurseries (Trierweiler, 2004). A questionnaire was issued, of which from 60 to 70 per cent were returned. Information was obtained from all parts of Germany, apart from two eastern German regions, on tree infestations in nurseries, private gardens and parks, and on roadside trees. None of the responses indicated the presence of resistant species of *Acer*, *Aesculus* or *Tilia*. However, *Tilia pallida*, *Tilia euchlora* and *Sophora japonica* were confirmed as newly recorded hosts. A relationship between infestation and species of host plant could not be established. However, there was a relationship between the degree of the infestation and the local factors, such as climate, ground cover (whether 'open' or 'sealed') and vegetation. The supposition, that the pest increasingly appears in 'sealed' (e.g. paved) locations, could not be confirmed, and the degree of infestation seems to depend on many different factors.

Specific control measures (such as the use of chemical pesticides, biological agents or the mechanical cleansing of infested trees) are presently the exception. In urban locations, for example, along streets and in pedestrian zones, mechanical measures result primarily in visual improvements, but they have little effect on the intensity of attacks. In nurseries, the insecticides dimethoate and imidacloprid are used to good effect. Also, initial experiences of the Baden-Württemberg Plant Protection Service, in the use of the parasitoid *Coccophagus*

lycimnia (Hymenoptera: Apelinidae) for biocontrol have been encouraging; however, ideally, weather conditions at the time of using biocontrol agents, need to be ideal.

DISCUSSION AND OUTLOOK

At present, knowledge of horse chestnut scale in Germany is limited. In recent years, the pest has increased its geographically range and infestations have also become more severe. The pest is no longer restricted to the major cities, but has also now spread to smaller ones. Control measures in public places are the exception, rather than the rule. The rapid nationwide spread during recent years suggests that infestations are often introduced on plants originating from tree nurseries, and this is confirmed by the authors' own observations. The national significance of host plants in many German cities further exacerbates the problem, as does their planting in monocultures.

Currently, the pest occurs mainly in urban locations, rather than in nurseries, where it mainly an aesthetic problem in green areas, such as city parks. In cities, the public complain about the unsightly appearance of infested trees, and also about contamination of the surroundings. Nevertheless, infestations in nurseries appear to be increasing, so commercial problems are to be expected.

Pest control on parkland and roadside trees is rarely practiced, even following infestations of horse chestnut scale. Usually, control measures (and then, typically, mechanical methods) are taken only on high-quality (e.g. specimen) trees. In general, firm recommendations and advice for the adoption of chemical and biological control measures are wanting. Possibly, horse chestnut scale will prove no more than an ephemeral problem, since (after five to seven years) infestations will begin to decrease, following the adaptation of native beneficial organisms (parasitoids and predators) to prey upon them (Arnold, 2003). Populations also decline in unfavourable weather conditions (e.g. low temperatures).

Post-infestation observations of pest population densities should still be made in urban areas, in order to confirm that beneficial organisms do eventually adapt and treat horse chestnut scale as a new food source. Future spread within the industry can be prevented only by strict monitoring of traded plants. In nurseries, therefore, frequent plant inspection by experts is of the highest priority, in order to maintain total freedom of the pest in such sites, and so guarantee the absence of problems being spread by commercial trade in nursery plants.

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Physiology of Benghal dayflower (*Commelina benghalensis*), a new invasive weed of the south-eastern USA

W K Vencill, P J Steptoe

3111 Miller Plant Sciences, Athens, 30602-7272 GA, USA

Email: wvencill@uga.edu

INTRODUCTION

In the USA, Benghal dayflower (*Commelina benghalensis*) (also known as tropical spiderwort) is listed as a noxious weed in 35 states. In Georgia, in a span of just 3–5 years, Benghal dayflower has gone from a relatively unknown plant to the most troublesome weed in cotton. In most cropping systems, chemical, cultural and biological control for this invasive weed is limited and, in glyphosate-resistant cotton, glyphosate often fails to provide >70% control. The plant's ability to germinate and thrive under a crop canopy limits many traditional cultural weed controls strategies. In addition, the ability of Benghal dayflower to root from the nodal areas severely limits tillage as a control tactic. There are no known biological controls.

The objectives were to examine specific parameters of the leaf surface of Benghal dayflower in relation to herbicide uptake (including leaf composition, thickness and morphology) under three soil moisture regimes (75, 50 and 25% of maximum relative soil capacity) and to compare herbicide uptake in moisture-stressed and non-moisture-stressed plants.

MATERIALS AND METHODS

Greenhouse-grown Benghal dayflower was moisture stressed, using procedures similar to those described by Earl (2003), stress being determined by keeping pots at desired maximum relative soil water contents of 75, 50 and 25%.

Leaf characterization was done at the University of Georgia Center for Ultrastructural Research, with a JEOL 100 CX TEM. From these images, cuticle thickness and the number of trichomes/μm^2 were calaculated (see Table 1).

The dose response of Benghal dayflower to several herbicides was examined at the three soil moisture regimes described above. Plants were sprayed with four non-zero rates of 2,4-D, diclosulam, flumioxazin, glufosinate, glyphosate, metolachlor and glyphosate + metolachlor, to derive an ED_{50} (effective dose to kill 50% of the test population, expressed in g/ha) value at each moisture level, based on a visual injury scale of 0–100% (0 = no control; 100 = complete control). Plants (all then c. 15–20 cm tall) were sprayed following exposure to moisture stress for 2 wk, and the stress continued for 7 d after spraying. ^{14}C-herbicide uptake was examined in excised leaves following treatment with diclosulam, flumioxazin, imazapic, sulfentrazone, atrazine, glyphosate, metolachlor, ^{14}C-glyphosate + metolachlor and ^{14}C-metolachlor + glyphosate, a field-use rate being mixed with the ^{14}C-herbicide counterpart. Ten μlitre drops of herbicide solution (containing a total of 20,000–200,000 dpm) were applied uniformly to the upper surface of each mature leaf. Unabsorbed ^{14}C-herbicide was

then removed by washing the treated leaf in 2 ml of 70:30 methanol:water v/v in a scintillation vial. The percentage of absorbed herbicide was then calculated by determining the amount of ^{14}C-herbicide in the leaf wash. Samples were quantified by liquid scintillation counting. ^{14}C uptake by the plant was expressed as the percentage of that applied.

RESULTS

Table 1. Cuticular thickness and trichome density of Benghal dayflower exposed to three soil moisture regimes.

	Moisture level (% of field capacity)		
	25	50	100
Cuticle thickness (μm):	1.1 ± 0.26	0.55 ± 0.07	0.30 ± 0.07
Trichomes/μm^2:	0.045	0.030	0.020

Leaf cuticle and trichomes increased with soil moisture stress (Table 1). Benghal dayflower was more susceptible to flumioxazin, glufosinate and metolachlor at higher moisture levels. The response to flumioxazin (ED_{50} = 2.9 to 0.1 g/ha; from 25 to 100% soil moisture), glufosinate (ED_{50} = 2.6 to 0.9 g/ha) and metolachlor (ED_{50} = 3.0 to 1.1 g/ha) increased significantly with soil moisture. Soil moisture did not affect the response of Benghal dayflower to 2,4-D, diclosulam or glyphosate. For 2,4-D (ED_{50} = 2.3 to 6.7 g/ha), this was due to the excellent response at all soil moisture levels. For diclosulam (ED_{50} = 38 to 62 g/ha) and glyphosate (ED_{50} >1,000 g/ha at all moisture levels), it was due to the poor response at all moisture levels. Whole plant responses to atrazine imazapic and sulfentrazone were not examined. Foliar uptake of flumioxazin, glyphosate, and metolachlor increased with soil moisture. For flumioxazin (37–82% uptake from 25 to 100% soil moisture), glyphosate (18–58% uptake) and metolachlor (31–78% uptake) uptake increased with soil moisture from 25 to 100% of field capacity. The addition of metolachlor to ^{14}C-glyphosate improved uptake over that of glyphosate alone, at all moisture levels (65–96%), and the addition of glyphosate to ^{14}C-metolachlor improved uptake over that of metolachlor alone, again at all moisture levels (79–92%). Foliar uptakes of atrazine, diclosulam, imazapic and sulfentrazone were not affected by soil moisture; 2,4-D and glufosinate uptakes were not examined. These data indicate that soil moisture can affect the response of several herbicides that are currently available to control Benghal dayflower in various cropping systems.

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Biologically active compounds as a possible cause of invasibility of knotweeds (*Reynoutria* spp.) from eastern Asia

N Vrchotová, B Šerá, J Tříska
Institute of Landscape Ecology, Branisovska 31, 370 05 Ceske Budejovice, Czech Republic
Email: nada@uek.cas.cz

E Dadáková, S Kužel
University of South Bohemia, Studentska 13, 370 05 Ceske Budejovice, Czech Republic

INTRODUCTION

Two species knotweed (*Reynoutria* spp.) spread from East Asia to Europe at the beginning of the nineteenth century. Now, Japanese knotweed (*Reynoutria japonica*), giant knotweed (*Reynoutria sachalinensis*) and, in addition, their hybrid Bohemia knotweed (*Reynoutria ×bohemica*) are widespread in central Europe. The hybrid is a very persistent, invasive plant which suppresses the original species and in this way changes biological diversity wherever it penetrates. Therefore, in our regions, Bohemia knotweed is undesirable.

The investigation is now concentrated on a study of resveratrol, piceid and other stilbenes in the roots of Japanese knotweed (Vastano *et al.*, 2000; Vrchotová *et al.*, in press). Several biologically active substances with the estrogenic activity have been found in the roots of Japanese and giant knotweeds, e.g. emodin and derivatives of 9,10-anthrachinon (Inoue *et al.*, 1992; Yang *et al.*, 2001), and extracts from the latter species contain substances with fungicide effects (Konstantinidou-Doltsinis & Schmitt, 1998). The pattern of phenolic compounds in the aerial parts of knotweeds have not yet been described in detail. However, catechins, chlorogenic acid, caftaric acid and quercetin derivatives have been found in *Reynoutria* by Vrchotová *et al.* (2004).

MATERIALS AND METHODS

The leaves and stems of knotweeds were collected in May, flowers in September and rhizomes in October 2002, in the Český Krumlov district (Czech Republic), from five specimens of each species. The leaves, stems and flowers were dried at laboratory temperature; the material was then pulverized and extracted with diluted methanol. Rhizomes were extracted fresh. The samples were analyzed using HPLC with a DAD detector on C18 column (2 × 150 mm, 3 μm), in water – acetonitrile gradient with the addition of trifluoroacetic acid.

For determination of total quercetin, the extracts were hydrolyzed in hydrochloric acid and then purified on a SPE column (C_{18}). After purification, the extracts were analyzed using CE with a UV detector, and borate buffer (pH 9.2) with the addition of SDS and methanol.

RESULTS AND DISCUSSION

Catechins (sum of catechin and epicatechin) were found in all parts of *Reynoutria,* but the largest amounts were found in the flowers of Japanese knotweed (2,035 mg/kg d.w.). The amount of catechins reached 2,400 mg/kg f.w. in the roots of giant knotweed, about twice that in Japanese knotweed (see Vrchotová *et al.*, in press). Large quantities of quercetin derivatives were found in all aerial parts, e.g. the total amount of quercetin in the flowers of Japanese, giant and Bohemia knotweed was 11,100, 16,600 and 19,500 mg/kg d.w., respectively. Resveratrol and its derivatives occurred mainly in the subterranean parts. The amount of resveratrol in stems was 11-13 mg/kg d.w., but none was found in the flowers or leaves. The largest amount of resveratrol was found in the rhizomes of Japanese knotweed (1,800 mg/kg f.w.); only 24 mg/kg f.w. was found in the rhizomes of giant knotweed by Vrchotová *et al.* (in press).

Stilbenes and catechins are substances with antioxidative, antimicrobial, anticancerogenic and fungicidal effects. The viability and resistance of knotweeds against parasitic fungi and pollutants are connected most probably with the high content of the above-mentioned phenolic compounds. The invasive plants have only a few natural enemies. Japanese knotweed is more invasive than giant knotweed, and this could be due to the high content of stilbenes in the roots. This may also be the reason why dead masses of the knotweed decay slowly and incompletely.

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Host-range studies of the pathogen *Phytophthora ramorum* on ornamental plants

S Wagner, K Kaminski, E Idczak, S Werres
BBA, Messeweg 11/12, 38104 Braunschweig, Germany
Email: s.werres@bba.de

INTRODUCTION

The pathogen *Phytophthora ramorum* is a heterothallic fungus-like organism which causes diseases on numerous ornamentals, shrubs and trees. It was first described in Europe, causing twig blight on *Rhododendron* and dieback on *Viburnum* (Werres *et al.*, 2001), before being confirmed as the same pathogen responsible for the newly observed (and widespread) death of oak trees (*Quercus* spp.) and other plants in California, USA, where the disease is known as 'sudden oak death' (SOD). In Europe, the pathogen has been found mainly on *Rhododendron* and *Viburnum* plants in ornamental nurseries, garden centres and a few public gardens (http://www.eppo.org/QUARANTINE/Alert_List/fungi/oak_death.htm), in 14 different countries. The pathogen has also been detected on other ornamental plant species, including *Camellia*, *Kalmia* and *Pieris*.

To date, just a single infection of a nursery plant of *Calluna vulgaris* has been reported (G Szkuta, personal communication) (in Poland); Tooley *et al.* (2004) have also tested some ericaceous ornamental host species. The aim of the presented studies was to test whether *P. ramorum* can infect ornamental cultivars of *Calluna vulgaris*, *Erica gracilis* and *Rhododendron simsii*.

MATERIALS AND METHODS

The following plant material was used: detached twigs of *Calluna vulgaris* (cvs Amethyst and Long White), *Erica gracilis* (cvs Weißes Schloß and Glasers Rote), and leaves of *Rhododendron simsii* (cvs Desta, Gloria, Kolibri and Palomai). Four isolates of *P. ramorum* were tested (three European and one American): isolate 1376 (from *Viburnum tinus*, cv. Eve Price, UK); isolate 1577 (from *Rhododendron catawbiense*, Germany); isolate 1578 (from *Rhododendron grandiflora*, UK) and isolate 1403 (from *Vaccinium ovatum*, USA). Test plants were inoculated with c. 10^5 zoospores/ml. The apices of seven wounded and seven unwounded twigs or leaves were dipped into the zoospore suspension for 5 s. Sterile distilled water was used as a control. After incubation for 6 d in a wet chamber at 20°C, with 16 h light, the length of discoloration was measured and the disease rate was calculated. Infection was verified trough PCR amplification, using the specific ITS Primer for *P. ramorum* (http://www.bba.de/inst/g/pramorumneu/pramorumstart.pdf).

RESULTS

With the dipping inoculation method (using a zoospore suspension), all detached plant parts could be infected with all tested *P. ramorum* isolates. For the *C. vulgaris* and *E. gracilis* cultivars (Table 1) the disease rates were 100%, except for one trail with 85% (*E. gracilis* cv. Glasers Rote and isolate 1578); the length of discoloration varied between 2 and 5 cm. The infection trails with wounded plant material produced predominantly more extensive (longer) discoloration than with non-wounded plant material, and the length of necrosis in *E. gracilis* cultivars was longer compared with those in *C. vulgaris* cultivars. The US isolate (1403)

seemed to be more aggressive than the three European ones. For the tested cultivars of *R. simsii* (Table 1), disease rates following wound inoculation were 100% with all isolates. When unwounded leaves were inoculated, disease rates varied from 39 to 100%. The infected leaves of *R. simsii* showed only slight discoloration (spots), to a maximum length of 1 mm.

Table 1. Disease rate (%) and length of discoloration (cm) after zoospore inoculation for all tested plant cultivars and isolates (n = 27, mean ± standard error).

	Calluna vulgaris / Erica gracilis			*Rhododendron simsii*	
Cultivar	Wounded	Unwounded	Cultivar	Wounded	Unwounded
Amethyst	3.62 ± 0.20	3.35 ± 0.38	Paloma	0.29 0.04	0.20 0.05 [2]
Long white	3.89 ± 0.36	2.20 ± 0.34	Desta	0.67 0.10	0.52 0.11 [3]
Weißes Schloß	4.06 ± 0.30	2.87 ± 0.29	Gloria	0.42 0.09	0.24 0.04 [4]
Glasers Rote	4.52 ± 0.15	4.19 ± 0.14 [1]	Kolibri	0.32 0.04	0.20 0.03 [5]
Isolate					
1376	3.79 ± 0.15	2.28 ± 0.25		0.43 0.11	0.13 0.05 [6]
1577	3.24 ± 0.47	2.79 ± 0.18		0.41 0.08	0.41 0.08
1578	3.53 ± 0.10	3.65 ± 0.47 [1]		0.39 0.05	0.10 0.04 [7]
1403	4.71 ± 0.30	4.80 ± 0.33		0.47 0.07	0.47 0.08

Disease rates 100%, except: [1] = 85%; [2] = 68%; [3] = 71%; [4] = 82%; [5] = 79%; [6] = 57%; [7] = 39%.

DISCUSSION

The presented studies confirmed the results of Hansen *et al.* (2005) that the detached-leaf-dip inoculation method provides a rapid assay and permitted a reasonable assessment of susceptibility to leaf blight. The results indicate that *P. ramorum* isolates could infect *C. vulgaris*, *E. gracilis* and *R. simsii* and the pathogen does not need to enter the plant tissue via wounds. Further studies must verify these results under natural conditions. To estimate the potential risk of *P. ramorum* for ericaceous ornamental plant species, additional tests are necessary to verify the differences between cultivars and isolates.

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Evidence for the eastward spread of the aggressive oilseed rape pathogen *Leptosphaeria maculans* in Europe

J S West, B D L Fitt

Rothamsted Research, Harpenden, Hertfordshire, AL5 2JQ, UK

Email: jon.west@bbsrc.ac.uk

M Jędryczka

Instytut Genetyki Roslin, P.A.S., Strzeszynska 34, 60-479 Poznan, Poland

Z Karolewski

August Cieszkowski Agricultural University, Dabrowskiego 159, 60-594 Poznan, Poland

INTRODUCTION

Leptosphaeria maculans is a serious pathogen of oilseed rape (canola, rapeseed, *Brassica napus*), causing seedling death and lodging or early senescence of adult plants (phoma stem canker or blackleg) in Europe, Australia and North America (West *et al.*, 2001). In Europe, *L. maculans* has been distributed mainly in western countries that grow oilseed rape, such as Germany, France and England. A closely related species, *L. biglobosa*, occurs throughout Europe, North America and parts of China, but is less damaging. The pathogens can be spread to new areas by seed movement, although this is rare (Wang *et al.*, 2003). Additionally, they produce wind-dispersed ascospores from fruiting bodies on crop debris. Most ascospores are deposited within a few kilometres of the source, but potentially a small number may blow considerable distances. Until the mid-1970s, phoma stem canker disease in Canada was caused only by *L. biglobosa* and was not considered to be important. However, *L. maculans* spread throughout Canada by the late 1980s (reviewed by Gugel & Petrie, 1992), having been first detected in Saskatchewan during the mid-1970s. Recently, *L. maculans* has been reported for the first time in Mexico (Moreno-Rico *et al.*, 2001) and Brazil.

EASTWARD SPREAD IN EUROPE

For many years, the phoma stem canker pathogen population in western European countries has been predominantly *L. maculans*, although there are small seasonal and regional differences in the ratio of *L. maculans* to *L. biglobosa* colonies isolated from diseased plants. Until the mid 1990s, phoma stem canker in Poland was almost exclusively caused by *L. biglobosa* (Jędryczka *et al.* 1994). A survey of the pathogen population structure, reported by Karolewski *et al.* (2002) showed that in some western regions of Poland rapeseed plants were highly infected with *L. maculans*, whereas in eastern parts of Poland infections caused only by *L. biglobosa* were found. Changes in ratio between the two species were also observed in the Czech Republic (Jędryczka *et al.*, 2002a), Hungary (Szlavik *et al.*, 2003) and Austria (Maria Eckert – personal communication). Until now, Russia remains the only country in east Europe where only *L. biglobosa* has been reported (Jędryczka *et al.*, 2002b; Gasich *et al.*, 2003).

DISCUSSION

Recent evidence suggests that the aggressive pathogen *L. maculans* is becoming more prevalent in central and eastern Europe. The spread of *L. maculans* in North America, particularly Canada in the 1970s and 1980s, was due to the introduction of *L. maculans* to that continent, where it was previously absent. In Europe, however, it is not clear as to why, until recently, the pathogen's distribution remained confined to the west. Reasons for the eastward expansion of the pathogen's range could include transport of contaminated seed from the west into central and eastern Europe, following changes in political regimes, and/or global warming, which may have reduced disease escape owing to shedding of frost-damaged leaves in more eastern parts of Europe. This suggests that the occurrence of *L. maculans* in central and eastern Europe should be monitored more closely and information provided to growers to improve its control.

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Interactions between defoliation and herbicide application on the growth and recovery of Japanese knotweed (*Fallopia japonica*) in pot and field experiments

A Wilcox, T A Germany

Harper Adams University College, Newport, Shropshire, TF10 8NB, UK

Email: awilcox@harper-adams.ac.uk

INTRODUCTION

Japanese knotweed (*Fallopia japonica*) is one of the most invasive plant species in the UK and is a serious weed of many non-agricultural habitats. Chemical and mechanical methods are the most common forms of control, but they are expensive and can be labour intensive. Classical biological control has been proposed as potential cost-effective option and studies are currently underway to isolate and develop suitable agents (Shaw *et al.*, 2001). This study aims to simulate the effects of a herbivorous (foliar-feeding) invertebrate on plant growth and survival, when used in conjunction with the commonly applied herbicide glyphosate.

MATERIALS AND METHODS

For the pot experiment, 96 individual plants were regenerated from similar sized root fragments in large pots in May 1997. The following treatments were applied: (a) *herbicide application* – plants were either untreated (= control) or treated with glyphosate at a rate of 1,800 g/ha; (b) *herbicide timing* – during plant development, herbicide was applied either 'early' or 'late'; (c) *defoliation* – to simulate the action of a foliar-feeding agent, plants had either none (0%), half (50%) or all (100%) of their individual leaves removed; (d) *defoliation timing* – plants were defoliated either 'early' or 'late' in their development. Treatments were applied in a factorial design, with four replicates of each treatment combination (i.e. $2 \times 2 \times 3 \times 2$). Above-ground vegetation from each plant was harvested six months after initial planting, oven dried and weighed. In the field experiment, 40 individual established plants were selected within an extensive stand in Granville Country Park, Telford, UK (Grid Reference SJ721121). Two treatments (a) *herbicide application* – plants were either untreated (control) or treated with glyphosate at the rate of 1,800 g/ha in July 1997 and again in July 1998; (b) *defoliation* – to more realistically simulate a foliar-feeding agent, plants had either none (0%) or half (50%) of each individual leaf present on the plant removed by cutting across the leaf blade in July 1997 and July 1998. Treatments were applied in a factorial design with 10 replicates of each treatment combination (i.e. 2×2). Each plant was harvested, oven dried and weighed following natural stem death in November 1997 and November 1998.

RESULTS AND DISCUSSION

For the pot experiment, dry weights from individual plants were analyzed by ANOVA, using the initial fresh weight of the root fragment as a covariate. Herbicide application ($P = 0.02$) and defoliation timing ($P = 0.05$) were the main effects and there were no significant interactions (Figure 1). Dry weights of individual plants from the field experiment were analysed using ANOVA for each individual harvest year (1997 or 1998). In 1997, there were

no statistically significant main effects or interactions. In 1998, herbicide application reduced biomass, although this was not significant ($P < 0.09$). There were no significant interactions. A repeated measures analysis showed that overall dry weight was lower in 1998 when compared with that in 1997 ($P < 0.001$) (Figure 2).

In the experiments described, there was limited evidence of interactions between defoliation and herbicide application on Japanese knotweed biomass. However, the field experiment did show that a combination of herbicide, defoliation and cutting could substantially reduce regenerating plant biomass. More-elaborate field experiments, performed over a longer period, are required before any subtle synergistic effects of a foliar-feeding biological control agent and the use of herbicide on Japanese knotweed control can be fully discounted.
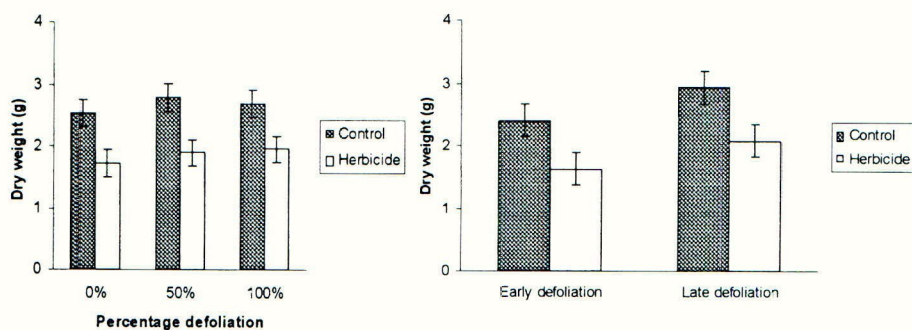

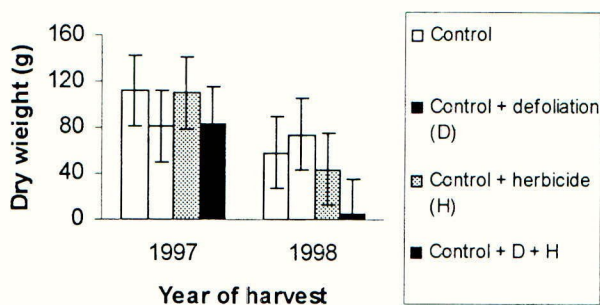Figure 1. Mean dry weights of Japanese knotweed from pot experiment +/- LSD (at $P = 0.05$).

Figure 2. Mean dry weights of Japanese knotweed from field experiment +/- LSD (at $P = 0.05$).

REFERENCES

Shaw R H; Child, L E; Evans H C; Bailey J P (2001). The Biological Control Programme for Japanese Knotweed (*Fallopia japonica* in the UK and USA - Phase 1). *Welsh Development Agency & United States Department of Agriculture Forest Service.*