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Validation of a model for predicting bracken production and control across the UK

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ABSTRACT

To develop a strategy for cost-effective bracken control we tested a computer simulation model designed to make long-term predictions. We produced predictions for 7 sites using nationally available climate data. At the site-scale, substantial improvements had to be implemented to the model to take account of slope and aspect. The model produced satisfactory results in two areas, (1) the broad scale ranking of untreated sites in terms of equilibrium biomass (except for one sheltered site), and (2) the approximate ranking of control treatment at 3 of the sites. The problem with the poor predictions for the sheltered site was probably the difficulty in deriving appropriate climate data. Equilibrium biomass was under-predicted at untreated sites with exceptionally high field values. The model was good enough for predictive purposes at the regional level but requires further development for site-specific predictions.

INTRODUCTION

The development of a control strategy for any pernicious weed requires knowledge of how the plant responds across the range of climatic and management practices for the country or region under consideration, and the ability to predict the impacts of changing environments or land-use change. One way to derive this information is to develop predictive models, which must be able to cope with local and or regional predictions. Bracken (*Pteridium aquilinum* (L.) Kuhn) is a problematic weed in many parts of the world, and is prevalent in marginal, upland land in the UK (Pakeman & Marrs, 1992). As bracken usually invades biotopes with a greater biodiversity interest (Pakeman & Marrs, 1992), its control is a priority within Agri-environment schemes. We have developed a series of computer-based simulation models (COBRA) for bracken productivity within the UK (Pakeman *et al.*, 1994), which make predictions at the site- or countrywide-scale. These models have been validated at a range of sites across the UK, but on those that are relatively flat, and where control treatments have been applied over a relatively short time period of three years (Paterson *et al.*, 1997). As many bracken-infested sites occur on steep slopes, further validation of these models is needed. This paper, therefore, charts our attempts to validate and further develop the COBRA models to produce reliable predictions under a greater range of conditions than hitherto.

METHODS

The model tested and refined was a site-specific, mechanistic physiological model (COBRA: Control of BRACKen; Pakeman *et al.*, 1994). The model computes daily changes in 3

compartments (frond biomass, rhizome biomass, rhizome carbohydrate concentration) over the yearly growth cycle (Williams & Foley, 1976). The model has been successfully tested against the raw data from which it was developed, and two independent studies. The model was refined (COBRA-E; Pakeman & Marrs, 1996) by the addition of equations to accommodate: (a) frond senescence, (b) a cost for all carbohydrate fluxes, (c) differential allocations between frond and rhizome during rhizome replenishment (Williams & Foley, 1976), and (d) a threshold was introduced for carbohydrate concentrations below which rhizomes could not persist. The environmental variables used to drive the model are: soil temperature, light, length of the growth season, and water status. All COBRA programs include options to assess the effects of different bracken control strategies. An extended version was then developed (COBRA-X) using MORECS climate data (Thompson *et al.*, 1981) to produce predictions for bracken equilibrium biomass at the countrywide scale, and to predict the outcome of climate change (Pakeman & Marrs, 1996). The extended individual site model (COBRA-E) was evaluated in a 3-year bracken control experiment at 6 sites across the UK (Paterson *et al.*, 1997) using meteorological data derived from the MORECS database (40 x 40 km scale). Individual site temperatures were re-calculated through a temperature correction via an altitudinal lapse rate; significant correlations were found between model predictions and experimental data (Paterson *et al.*, 1997).

Data for model validation

Measurements of rhizome biomass are essential to further validate COBRA-E. Recently, we collected new information on the effects of bracken control on rhizome performance from 7 experiments - Carneddau, Cannock (3 expts), North Peak, Sourhope (2 expts), 5-7 years after bracken control treatments were implemented on dense bracken (Table 1; Le Duc *et al.*, 2003).

Table 1. Descriptions of the 7 sites and the nearest Meteorological Station

Experiment	Sourhope		North Peak	Carneddau	Cannock Chase		
	1	2			1	2	3
Location							
Grid reference	NT 861202	NT846210	SK213870	SH690711	SJ976200	SJ987181	SJ987178
Physical							
Altitude (m)	325	285	290	350	145	165	175
Aspect (°)	280	130	275	190	140	125	175
Slope (°)	16	22	9	20	20	18	9
Meteorological Station							
Name (Grid reference)	Sourhope (NT845202)		Buxton (SK339873)	Aber (SH656731)		Penkridge (SJ920115)	
Approximate distance from site (km)	0.5	2	13	4		10	

The treatments were for six experiments: untreated control; cut once/year; cut twice/year; cut year 1 + sprayed with asulam year 2; sprayed once; sprayed year 1 + cut year 2. Restoration treatments were applied to sub-plots. Here, pooled means for the bracken control main-plot treatments were used for comparison with predictions. The seventh experiment (Cannock 3) had a different design and was used only to assess performance in the untreated plots.

Further validation: use of COBRA to predict bracken performance in six experiments

Initially COBRA-X model was used with MORECS climate data; the meteorological data were available at monthly intervals at 40 x 40 km resolution. Initial results were poor with equilibrium values lower than measured values. One of the reasons suggested for this poor fit was the limited availability of climate data at an adequate spatial-temporal resolution. Thus, we reverted to the original site-based COBRA-E model, and acquired daily climate data for each site from the British Atmospheric Data Centre database; data on soil temperature at 30 cm depth (°C), global irradiation on a horizontal surface (MJ m⁻²), potential and actual transpiration (mm) were derived. The number of days between the last spring frost and the first autumn frost was used to estimate the growing period length. However, results were still inadequate. As previous calibrations had been done on sites that were almost flat, and the sites in the present study were on slopes of varying aspects, we considered slope and aspect might be critical. Thus, we recalculated the global irradiation arriving at the site taking slope and aspect into account, and recalculated other environmental parameters needed for the model.

For global irradiation we assumed that the light integral does not change significantly within each MORECS square. The Junelight and Declight (June & December mean global irradiation) from the MORECS database was used to calculate daily global irradiation (I) using eqns 1-3.

$$I = c + \left(d \times \sin \left(\frac{(\text{DayNo} - 80) \times 2\pi}{365} \right) \right) \quad 1$$

$$c = \text{Junelight} - \left(d \times \sin \left(\frac{173\pi}{365} \right) \right) \quad 2$$

$$d = \frac{(\text{Junelight} - (0.0028 \times \text{Alt})) - (\text{Declight} - (0.0052 \times \text{Alt}))}{\left(\sin \left(\frac{173\pi}{365} \right) - \sin \left(\frac{540\pi}{365} \right) \right)} \quad 3$$

We included slope and aspect using the graph of the influence of slope and aspect on the yearly total direct radiation potentially received at Latitude 53°15'N (Pope & Lloyd, 1974) and recalculated the transformed Irradiation Received parameter (I_{transf}), eqn 4 and parameters derived from Pope & Lloyd's figures:

$$I_{\text{transf}} = I \times (1 - (0.017995 \times \cos(\text{aspect}) \times \text{slope})) \quad 4$$

The days of last and first frost were first estimated using the minimum temperatures from BADC, but then re-calculated, by plotting dates of both last spring and first autumn frost against the irradiance at the site (max light), eqns 5-6:

$$\text{Last frost} = -6.5601 (\text{max light}) + 236.7, r^2 = 0.303 \quad 5$$

$$\text{First frost} = 6.1273 (\text{max light}) + 182.16, r^2 = 0.232 \quad 6$$

Thus, the dates of the first and last frost were recalculated taking into account slope and aspect, the difference between the old max light and the new max light was multiplied by 6, and subtracted from the dates of the last frost and added to the dates of the first frost.

Daily actual evapotranspiration (AT) and the potential evapotranspiration (PT) were re-calculated using the daily irradiation and total yearly irradiation previously calculated and the 40 km square grid value of each site using eqns 7 and 8.

$$AT = 40kmvalue \times \frac{Dailylight}{Totalyearlylight} \times (1 + (0.002 \times Alt)) \quad 7$$

$$PT = 40kmvalue \times \frac{Dailylight}{Totalyearlylight} \times (1 + (0.002 \times Alt)) \quad 8$$

However, between May and September, rainfall can limit actual evapotranspiration, and during this period of the year AT' was calculated using eqn 9.

$$AT' = AT \times \frac{Monthlyrain}{meanmonthlyrain} \quad 9$$

Some soil temperature and minimum temperature data were missing for short periods; these missing values were estimated from regression equations (all $P < 0.001$, $r^2 > 0.84$).calculated between the variable in question and equivalent measures from the nearest station

Initially we estimated the equilibrium value for each bracken stand by running the model for 48 years using the seven years of climate data for each site (1994-2000) randomly permuted, with an initial rhizome starting value of 1500 g m⁻². Thereafter, the model was run with the 7 years climate data and the equilibrium value as a starting value and each of the applied bracken control treatments. The predicted values (1 January of the year immediately after the sampling) were then compared with measured rhizome data (Le Duc *et al.*, 2003). A sensitivity analysis at Sourhope 1, and North Peak identified daily evapotranspiration, light, growing season and accumulated soil temperature as most important.

RESULTS AND DISCUSSION

The equilibrium biomass derived from the model was reached relatively quickly (<21 years); at 6 sites biomass increased and reached an asymptote. Two problems, however, were noted: (1) an unusual result at Sourhope 1, where the equilibrium value reduced to near zero, and (2) an underestimation of equilibrium biomass, but especially when measured field values were high.

The Sourhope 1 problem

Initial estimates of equilibrium biomass were near zero even when the corrections for slope and aspect were included (Fig.1). This was clearly incorrect as bracken exists on the site. Initially, we compared the predicted climate data for Sourhope 1 with that for Sourhope 2. This comparison showed huge discrepancies in length of growing season between the Sourhope sites. The respective average Julian days were 152 and 254 for the last spring frost and first autumn frost, giving a growing season of 102 days for Sourhope 1 compared to day 122, day 285 and 162 days for Sourhope 2. This suggests a reduction in growing season of about 2 months at Sourhope 1 compared to a site 1.5 km away.

Therefore, we re-ran the model for Sourhope 1 using the Sourhope 2 growing season length and an improved prediction was obtained (Figure 1), but still less than expected. We also (by chance) acquired some climate data from (a) an automated station on an exposed ridge near Sourhope 1 and (b) a local weather station adjacent to Sourhope 1. These stations were 700 m and 50 m from the Sourhope 1 experiment respectively. The respective average Julian days calculated using these data suggest further increases in predicted growing season length, with increasing proximity between the weather station and experiment (224 days and 231 days respectively), a further two-month increase. We re-ran the model using these values and there was a consistent improvement with increasing season length, although the predicted equilibrium biomass was still only 50% of predicted values (Figure 1). The extra 7 days growing season between the automated and local weather stations markedly improved predictions.

Clearly, at this site, there are problems in predicting site-specific microclimate data from national databases. Part of this problem may be that Sourhope 1 is very exposed on a northeasterly aspect, and is sheltered by a small conifer plantation. Predicting season length is difficult, and further more detailed on-site measurements are needed at this site.

The problem with equilibrium biomass prediction

There was a significant relationship between the equilibrium value (y) from COBRA and field measurements (x) ($y = 0.4181x + 1009.4$; $r^2 = 0.8879$), if we exclude Sourhope 1 (Figure 1).

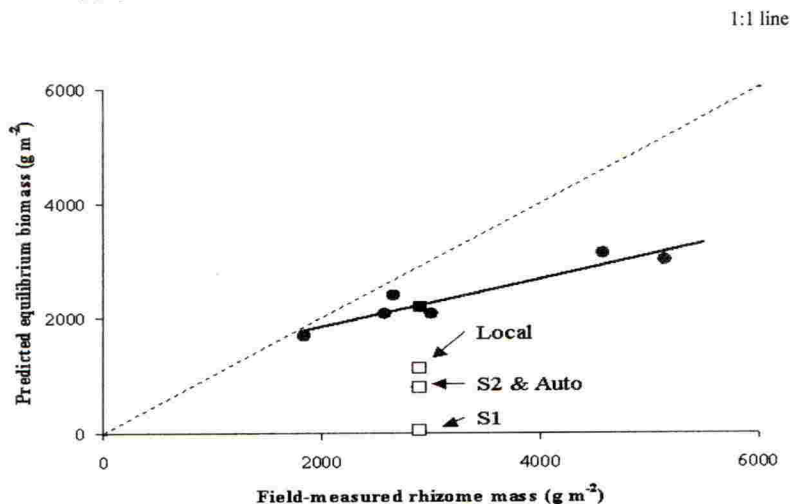


Fig. 1. Relationship between rhizome biomass of field and model predictions for untreated sites. Five predictions for Sourhope 1 are given: from regression line above (■) and with data from (□), S1 = Sourhope 1 weather data, S2 = Sourhope 2 weather data, Auto = Automated weather station, Local = local weather station; 2 & 3 are superimposed.

A notable result was that all equilibrium values were underestimated by the model, and the underestimation increased when the measured value was high (> 4 t/ha) at Cannock 1 and Carneddau. For 4 sites where the measured biomass values ranged between 1.5-3 t/ha, underestimation was small and sites lie close to the 1:1 line. However, although the North Peak

value is close to 1:1, this may still be an underestimate as the field samples were collected in 2000, a very wet year. Le Duc *et al.* (2003) speculated that these wet conditions were responsible for a reduced rhizome biomass.

Ideally, field measurements should always be less than the model predictions for equilibrium biomass. There are several reasons why this might not occur. (1) There is an issue over accurate field climate data, with difficulties illustrated above for Sourhope 1. Unfortunately there are no good long-term, on-site measurements of micro-meteorological data for these sites and such data are needed to further test and refine the models. (2) the model was derived from data collected from bracken stands with biomass values in the mid range of those reported here. Derivation of parameters from a wider range of sites may perhaps improve the validity of the models. (3) It is also possible that the model parameters change in very large rhizome systems in a non-linear way. (4) There may be a slight discrepancy as COBRA predictions are based on 1 January and field data were collected in late October-November. Late autumn measures would be expected to be slightly greater than January ones, because there should be some additional respiratory loss. (5) It is possible that the assumptions and simplification of the processes within the model require to be re-evaluated and improved.

Predicted effects of bracken control treatment

There was some agreement between predicted and measured values for *Pteridium* control treatments at three of the six sites (Carneddau, North Peak & Sourhope 2). These sites showed a similar order of effect and were close to the 1:1 line (Figure 2), indicating that the reductions in rhizome biomass predicted by the model were similar to those found under field conditions. For these sites the correlation coefficients (r) between actual and predicted values were 0.394, 0.792, 0.585 respectively; only the one for North Peak being significant at $p < 0.05$, the other two being significant at $p < 0.10$. Part of the reason for the low significance is the low sample size ($df = 3$) The correlations for the other Cannock 1 and 2, and Sourhope 1 had an $r < 0.15$ and were not significant.

The general rank order of predicted treatment effects were the same at all sites except Sourhope 1, from the worst treatment spray once < cut + spray < cut once < spray & cut < cut twice (the best). For most sites the field data did not show the same detailed rank order although spray once was usually the worst and cut twice usually the best.

CONCLUSIONS

In broad terms the model produced satisfactory results in two areas: (1) the ranking of untreated sites in terms of equilibrium biomass (except Sourhope 1), and (2) the approximate ranking of control treatments at 3 sites. The major problem was the very poor predictions for Sourhope 1, and this is almost certainly the result of the difficulty in deriving accurate climate data for this sheltered site. Thus, national climate data are not reliable enough to use for routine bracken performance predictions at the site level, although predictions for many sites are reasonable (6 out of 7). However, inspection of equilibrium biomass can provide an obvious means of identifying rogue sites, where climate data are inadequate. The models also under-predict biomass at sites with high measured values. Further development of COBRA is needed to improve predictions at such sites.

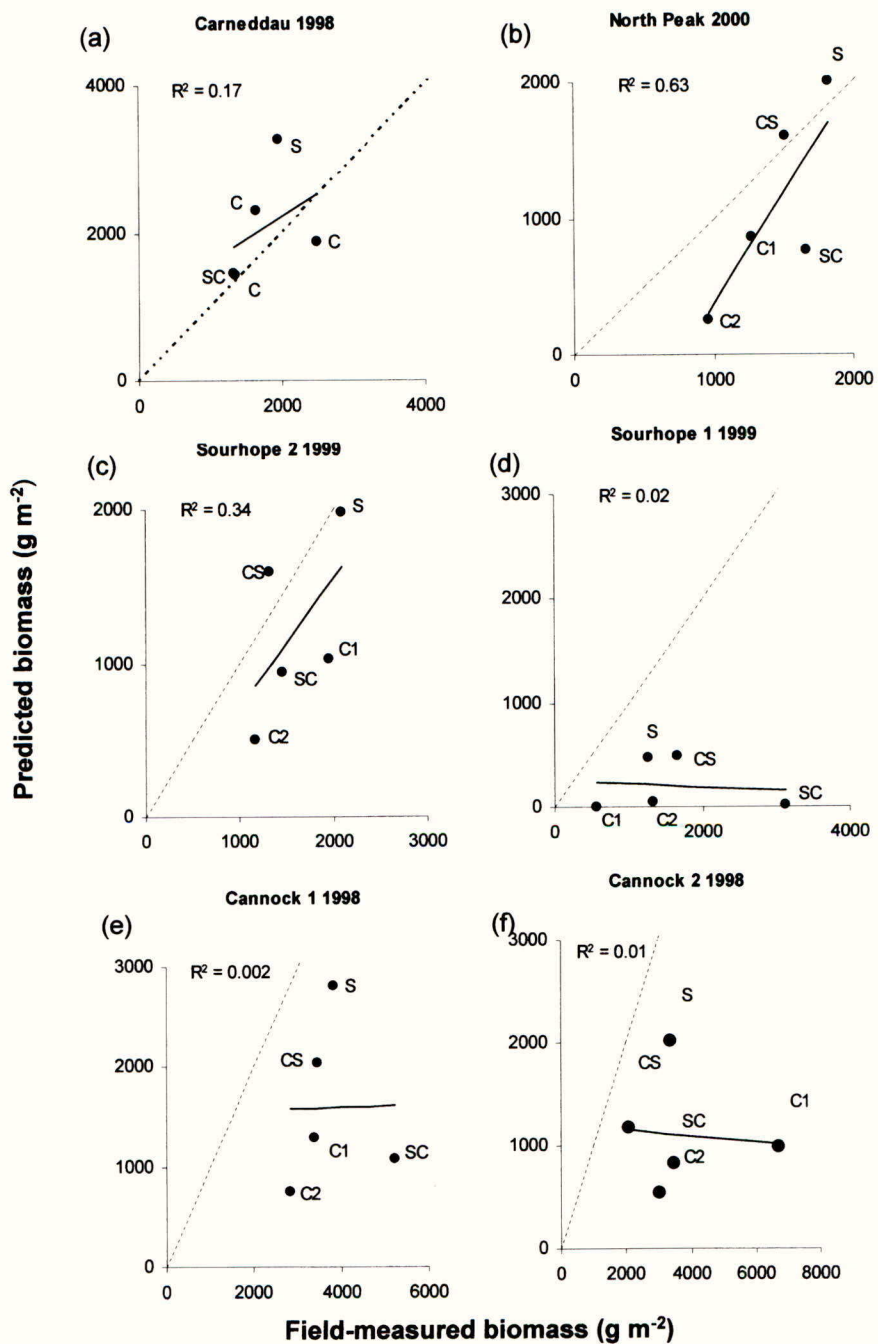


Figure 2. Relationship between rhizome biomass in the field and model predictions where bracken control treatments have been applied at 6 sites: (a-c) reasonable fits; (d-f) poor fits. Key: C1=cut once/yr; C2=cut twice/yr; S=Spray; SC=Spray+Cut; CS=Cut+Spray

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Analysis of the risk of brown rot to the high-grade seed potato production in Scotland

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ABSTRACT

Potato brown rot is caused by the quarantine bacterium *Ralstonia solanacearum*. Outbreaks in Europe have been associated with the irrigation of potato crops with contaminated river water. Although *R. solanacearum* was detected in one Scottish river in the past, the bacterium has never been found in Scottish potatoes. After three years of eradication and monitoring, Scotland is now free of the bacterium. This paper assesses the risk of spread of *R. solanacearum* via surface water and of the occurrence of brown rot in seed crops under Scottish conditions. At least 50 Scottish river systems flowing through potato growing areas may be at risk from contamination with *R. solanacearum*. Long-term survival of the bacterium in Scottish waters is unlikely without the presence of the wild host *Solanum dulcamara*. This plant is not widespread in Scotland and has so far been found in only 6 out of 48 river systems surveyed. Ten of the 50 river systems potentially at risk from contamination have been used for irrigation in the past two years. Although the acreage of seed crops irrigated from these waterways is relatively small, prevention of infection is important to avoid the occurrence and spread of brown rot in Scotland, especially since *R. solanacearum* biovar 2, race 3 is able to infect Scottish potato varieties and cause disease under Scottish conditions. Testing of Scottish waterways for the presence of the brown rot pathogen continues to prevent the contamination of fields and ensure the good health of Scottish seed.

INTRODUCTION

Ralstonia solanacearum is a geographically widespread and genetically diverse Gram-negative bacterium that causes bacterial wilt in a broad range of host plants. The species is sub-divided into races and biovars on the basis of pathogenicity and carbohydrate metabolism respectively. *R. solanacearum* biovar 2, race 3 is the cause of brown rot, a destructive disease of potato. In Europe, infection with *R. solanacearum* usually results in compulsory destruction of the affected crop and associated stocks since this pathogen is classed as a quarantine organism within the EU. All English cases of brown rot and many cases elsewhere in Europe have been traced back to the use of surface water contaminated with *R. solanacearum* for irrigation. A number of waterways in several European countries, including England, the Netherlands and Belgium, are contaminated with the bacterium. In Scotland, *R. solanacearum* was previously found in parts of the Tay river system (Wood *et al.*, 2002) but, after removal of its wild host,

Solanum dulcamara, from the affected waterways, has not been detected for the past few years. The bacterium has never been found in Scottish potatoes. This paper offers an analysis of the risk of future river contamination and subsequent infection of crops in Scotland in order to aid prevention and safeguard the high quality standard of Scottish seed.

ENTRY OF INFECTED MATERIAL

For regions in Europe where *R. solanacearum* is not present, such as Scotland, the importation of infected plant material poses the greatest risk of entry. Outbreaks of brown rot as a result of imported contaminated potato seed have been reported in other countries but are less likely in Scotland as little seed is imported. The importation of contaminated ware potatoes from countries where *R. solanacearum* is widespread is the most likely source, but other plant material such as pelargonium may also pose a threat. Testing of stocks that are brought in and import restrictions aim to limit the risk of entry. However, the chance of finding and thus eliminating infections in potato stocks is low (<18% if <0.1% of tubers are affected) (Janse & Wenneker, 2002) and the amount of ware potatoes imported into the UK each year is considerable (357,000 t in 2002/2003) compared with the amount of testing conducted. There is therefore a continuous risk of entry of *R. solanacearum* into Scotland.

THE AQUATIC ENVIRONMENT

Contamination of waterways

Waste from plant material infected with *R. solanacearum* could reach the fresh water environment in a number of ways. Wastewater from households, plant nurseries or industry, contaminated through infected potato imports or other infected material, could be released into waterways directly. *R. solanacearum* may survive in non-treated wastewater from the potato processing industry for up to a week (Wenneker *et al.*, 1998) and industrial plants processing potatoes have been pinpointed as a likely source of contamination of surface water in several European countries (Olsson, 1976; Janse, 1996). In most of these cases, the potato waste discharged into the rivers was untreated. In Scotland, most wastewater is first treated in sewage plants or tanks and only then released into the aquatic environment. However, the efficacy of treatment may differ from plant to plant and in the case of the previous contamination of parts of the Tay river system with *R. solanacearum*, a primary wastewater treatment plant was suspected as the source of contamination (Wood *et al.*, 2002).

A study of waterways in Scotland has shown that at least 12 rivers flowing through potato growing areas are at risk from contamination via primary wastewater treatment plants. In addition, surface water in the same regions could be contaminated via accidental or illegal release of waste from septic tanks (39 waterways) or potato processing and packing plants (22 waterways). In total, at least 50 waterways in Scotland are potentially at risk from contamination with *R. solanacearum* although the degree of risk varies considerably between and within these river systems. Larger rivers such as the Tweed and Tay have the highest risk of contamination due to the large number of tributaries, and thus possible contamination sources, upstream.

Spread and survival in water

Not much is known about how long surface water remains contaminated with *R. solanacearum* without the presence of wild host plants such as *S. dulcamara* (see below). Assuming a single contamination event, the persistence of the bacterium will depend on a wide range of factors including the survival rate of *R. solanacearum* under particular environmental conditions, and the width, depth, length and flow rate of the river in question. Most rivers in Scotland have relatively high flow rates and non-persistent contaminations could be expected to be flushed out within a few days.

Survival of *R. solanacearum* in water is longest at temperatures ranging from 12 to 28°C, and in sterilised water, the bacterium can even multiply at these temperatures. An increase in population does not take place in natural non-sterile river water or sediment, but survival may be as long as four months (van Elsas *et al.*, 2001). Recent research has confirmed that the bacterium is able to survive in nutrient-poor aquatic environments but has also shown that survival in non-sterile river water is limited due to predation by protozoa. On the basis of this information, long-term survival of *R. solanacearum* in the aquatic environment in Scotland is thought to be limited without the presence of a natural host.

The role of wild host plants

Bittersweet or woody nightshade (*Solanum dulcamara*) has been identified as an important natural host of *R. solanacearum* biovar 2 race 3 in Europe. The presence of this plant plays a major role in the survival and multiplication of the bacterium in the aquatic environment and has been a factor in most if not all brown rot outbreaks caused by irrigation with contaminated water (Olsson, 1976; Janse, 1996). *S. dulcamara* is a deciduous woody vine that can grow with its roots immersed in water and, in this situation, infection by *R. solanacearum* may occur in contaminated rivers. The bacterium may spread systemically throughout the plant and overwinter in underground stolons. Large numbers of *R. solanacearum* cells can leach from infected submerged bittersweet roots, particularly in warmer weather, thus increasing the contamination level of the affected waterways (Elphinstone, 1996). The bacterium can also be spread downstream via infected detached plant parts such as root and stem pieces. This means that information on the presence of bittersweet in Scotland is very important for establishing the risk of infection of potato crops with *R. solanacearum*.

Although *S. dulcamara* can be found in places away from water, such as wasteground, hedges, dunes and woodland, it also typically inhabits marshes and banks of slow-flowing rivers. Slow flowing waterways are mainly found in lowlands and the abundance of the plant in this type of habitat concurs with its reported scarcity at higher altitudes (>200-250 m). The species also has a low tolerance to cold. In Scotland, *S. dulcamara* occurs much more sporadically than in England and most other European countries (Preston *et al.*, 2002). A survey of Scottish waterways at risk from contamination with *R. solanacearum* is underway and has shown that *S. dulcamara* is only abundant on the banks of streams with low flow rates (< 2 m³/s). Of the 48 river systems surveyed so far, only six harbour *S. dulcamara* on their banks. If the plant is present at a survey site, it is usually also found at all sites downstream, suggesting vegetative spread or dispersal of berries in the aquatic environment. Molecular genetic studies have indicated that populations of *S. dulcamara* in some Scottish rivers are clonal, but this is not always the case. The level of genetic diversity of *S. dulcamara* in a river system could affect the chance of survival by *R. solanacearum*, but further studies are needed in this area.

Black nightshade (*Solanum nigrum*), has also been suggested as a wild host for *R. solanacearum* (Olsson, 1976), but it is annual and not generally found on riverbanks (Preston *et al.*, 2002). Another reported weed host that is widespread in Europe is stinging nettle (*Urtica dioica*). However, natural systemic infection of this species by *R. solanacearum* is very rare (Wenneker *et al.*, 1999).

THE FIELD ENVIRONMENT

Contamination of fields

Contamination of agricultural fields with *R. solanacearum* can take place via the use of infected plant material, in particular potato seed imports, which are limited in Scotland (see above). Other possible routes could include the dispersal of biosolids from sewage plants that have processed contaminated waste or manure and food debris from animals fed with infected plant material. However, the most likely route is via contaminated surface water, which can occur through the use of river water for the dilution of agrochemicals sprayed on crops or suspension of fertilisers applied to the field, although the amount of water used in these cases is generally relatively small. Flooding is another possible cause of contamination, although floods in Scotland often occur in winter when levels of *R. solanacearum* are low.

As indicated earlier, many cases of brown rot in Europe have been due to irrigation with contaminated water and this also seems to be the biggest threat to Scottish potato crops. Irrigation generally takes place in warm weather conditions when bacterial inoculum levels in contaminated water are at their peak. Of the 50 Scottish river systems potentially at risk from contamination with *R. solanacearum* (see above), ten were used for the irrigation of seed crops in the last two years. Four waterways used for extraction were downstream from a primary wastewater treatment plant. The acreage of seed irrigated with water from rivers at risk from contamination was only a small percentage (1.7%) of the total grown in Scotland (11,747 ha in 2004), but prevention of the infection of any Scottish seed is important to avoid the widespread occurrence of brown rot.

Survival in soil

Once a field has become contaminated with *R. solanacearum*, the chances of crops becoming infected depend on the survival of the pathogen in the soil and the soil conditions. Survival is decreased by continuous waterlogging and also by drought and low temperatures. It is shorter in sand than in loam soils (Hayward, 1991; van Elsas *et al.*, 2000). Survival in sand is known to be longer at 15°C than at higher or lower temperatures (van Bekkum *et al.*, 1997). Survival will also depend on the biotic aspects of the soil and, as in water, predation by protozoa probably plays an important role. The presence of potato groundkeepers or other susceptible host plants will extend survival of the bacterium in the field considerably.

It is hard to predict the survival of *R. solanacearum* in Scottish fields after a contamination event on the basis of current information. The bacterium is likely to persist long enough to infect a crop in the same season but long-term survival (in groundkeepers) until the next potato crop can be expected only in ware crops in view of the long rotation period for seed in Scotland (at least five years).

Infection and disease development

Very little clarity exists on the conditions necessary for successful infection of potato plants and subsequent development of brown rot by *R. solanacearum* biovar 2, race 3. Infection of solanaceous crops is known to be aided by root wounding, although this is not required when inoculum levels are high (Kelman & Sequeira, 1965). For this reason, the presence of plant pathogenic nematodes may also facilitate infection by *R. solanacearum*.

Ralstonia solanacearum biovar 2, race 3 is able to infect and cause disease under cool conditions, which is why it is a threat to the potato industry in Europe. Disease occurs regularly in cool climates such as areas of high elevation in South America where the mean temperature is around 13°C. Although the virulence of strains of *R. solanacearum* depends on temperature and symptoms take much longer to develop under cooler conditions, disease can be extensive at temperatures below 20°C (Ciampi & Sequeira, 1980). Experiments have confirmed that brown rot symptoms can develop in Scottish varieties at 18°C and research is now underway to determine the lower temperature threshold for infection and disease development in tubers. Trials in which the ten most widely grown seed cultivars were tested for their susceptibility to *R. solanacearum* have shown that none of these are resistant to the pathogen. Brown rot occurred in all ten varieties. There are reports that latent infections of daughter tubers may be common in the more resistant varieties (Ciampi & Sequeira, 1980) and at low temperatures (Nyangeri *et al.*, 1984). Seed tubers with latent infections would be much harder to detect than ones showing clear brown rot symptoms and latent infections could therefore play an important role in the spread of the bacterium.

The sum of the above factors indicates that *R. solanacearum* biovar 2, race 3 would be able to infect plants and spread to daughter tubers under Scottish conditions. The level to which latent tuber infections would be formed, epidemiologically a very important aspect, is still unclear and needs further investigation.

DISCUSSION

On the basis of data from the literature and our study of Scottish waterways and experiments on infection and disease development, it is clear that *R. solanacearum* biovar 2, race 3 poses a threat to the Scottish seed potato industry. Many river systems in Scotland, which are at risk from contamination with the pathogen, flow through seed producing areas and some of these waterways are used for irrigation. However, analysis of the situation in other countries has shown that the presence of the wild host *S. dulcamara* is a key factor in the outbreak of brown rot and our survey has shown that this plant is not widespread in Scotland, significantly reducing the risk in many seed potato-growing areas. The prevention of contamination of fields used for potatoes is crucial as indications are that infection and disease development by biovar 2, race 3 strains of *R. solanacearum* are possible under Scottish conditions. An extensive scheme of testing of Scottish waterways for the presence of *R. solanacearum* is undertaken annually. A more sensitive detection method than previously applied is under development and this should hopefully ensure that any river contamination is found quickly and the use of contaminated water for irrigation prevented. Regular testing of Scottish waterways will continue in the future to safeguard the health and reputation of Scottish seed potatoes.

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Eradication of the first UK outbreak of chrysanthemum stem necrosis virus (CSNV)

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ABSTRACT

In November 2002 an outbreak of chrysanthemum stem necrosis virus (CSNV) was detected for the first time in the UK in an all year round (AYR) chrysanthemum crop. Stem lesions rendered 50% of stems in one variety un-marketable. Eradication and containment action was taken; the main priority was to prevent the spread of the virus at the outbreak site and to other sites growing chrysanthemums or tomatoes. A comprehensive insecticide regime which targeted the Western flower thrips (WFT) vector was used. Sticky traps were installed to monitor the WFT population and to obtain individual thrips for testing for CSNV by TaqMan . It was decided that the virus could be declared eradicated following one complete life cycle of the WFT with no findings of the virus in either plants or thrips. No CSNV was detected in plants and no infected thrips were trapped after March 2003 and the CNSV outbreak was declared eradicated in August 2003.

INTRODUCTION

Chrysanthemum stem necrosis virus (CSNV) was detected in a crop of all year round (AYR) chrysanthemums in South West England in November 2002. As CSNV had never been found in the UK and the disease was considered to pose a significant threat to chrysanthemums, tomatoes and potentially other crops, emergency statutory action was implemented by Defra to eradicate the disease via management of both the infected hosts and the thrips vector.

CSNV was first recorded on *Chrysanthemum morifolium* and *Dendranthema morifolium* in Atibaia, São Paulo, Brazil (Duarte *et al.*, 1995). Necrotic lesions surrounded by yellow areas, first on the leaves and then on the stems were observed on chrysanthemum plants. More recently, in Minas Gerais State, Brazil, CSNV has been found to infect tomatoes (*Lycopersicon esculentum*) (Nagata *et al.*, 1998). During 1994/95, CSNV was found on four nurseries in the Netherlands on chrysanthemums imported as cuttings from Brazil (Verhoeven *et al.*, 1996), however there have been no findings of CSNV in the Netherlands since 1996 (Verhoeven & Roenhorst, 1998).

As with all Tospoviruses, CSNV is transmitted by thrips. The Western flower thrips (WFT) (*Frankliniella occidentalis*), which is widely distributed throughout the UK and Europe in protected cultivation, is known to be an effective vector of CSNV (Nagata *et al.*, 2004), with the virus being acquired by the first and second larval instars as a result of feeding on infected plants (Nagata *et al.*, 2001). Thrips remain infected throughout their development, allowing the

adult thrips to infect new crops, however infection is not transmitted from the adult to the egg. *Frankliniella schultzei*, which is not present in the UK but regularly intercepted on cut flowers, has been shown to be an important vector of CSNV in Brazil (Nagata *et al.*, 2004).

CIRCUMSTANCES OF THE UK OUTBREAK

The UK outbreak was confirmed when a sample of the crop, which was exhibiting dark stem lesions and some leaf necrosis, was sent to CSL for testing for verticillium wilt. However, given the symptoms and the absence of fungal pathogen, the plants were tested by ELISA for the presence of three tospoviruses: CSNV, Tomato spotted wilt virus and Impatiens necrotic spot virus. CSNV was detected. Electron-microscopic examination of the sap was carried out and spherical tospovirus-like particles were found. Mechanical inoculation of a range of indicator plants with sap from the affected stem material resulted in typical symptoms and tested positive for CSNV when tested by ELISA (Mumford *et al.*, 2003).

The infected plants were found in one of the six glasshouses on the site, all of which were under continuous chrysanthemums. The affected glasshouse contained three blocks of chrysanthemums of different cultivars. The infection was confirmed in cvs. Fiji and Calabria which had been imported as cuttings from Brazil in August 2002; these were mature and nearly ready for harvesting when CSNV was confirmed. The plants sent to CSL for testing not only showed symptoms of CSNV but also some evidence of feeding damage by thrips. Examination of existing sticky traps revealed that the vector (WFT) was present at a low level in the glasshouse. Most of the infected plants were found in four beds containing cv. Fiji, rendering up to 50% of plants unmarketable. A single plant of cv. Calabria was found to be exhibiting symptoms. These observations suggest that the source of infection was the planting material of cv. Fiji. The third cultivar in the glasshouse showed no sign of CSNV.

A set of recommendations for pest management/eradication were prepared by CSL and formed the basis of statutory action, carried out under the supervision of the Plant Health and Seed Inspectorate (PHSI). The aim of this action was to eradicate the virus by targeting both the infected hosts and the viruliferous vectors still present in the glasshouse.

RECOMMENDED STATUTORY ACTION

An initial set of recommendations was given in November 2002. Immediate action required the removal of visually infected plants, which were destroyed by burning. Symptomless flowers from the infected glasshouse were allowed to be harvested but were required to be sent direct to retail or wholesale and their movement onto any other nursery growing chrysanthemum or tomatoes was prohibited. The removal of any weeds in or immediately outside the glasshouse was also required, as these could act as an alternative host for the WFT. The weeds were removed and destroyed by burning or deep burial. At this time it was too late in the life of the crop (flowers were ready for cutting) to apply insecticides to control the WFT. Blue sticky traps were installed in the glasshouse to monitor the population of WFT and its spread. From these traps individual WFT were obtained for testing by a newly established PCR technique ('TaqMan') (Boonham *et al.*, 2002) to establish whether CSNV was being carried by the thrips in the glasshouse. Trapping, identification and testing of the thrips continued throughout the

eradication campaign. A steam treatment of the soil was required between the end of one crop and the beginning of the next to target potentially infected pupae within the soil.

Inspections of the neighbouring glasshouses for symptomatic plants were carried out by the PHSI. All other chrysanthemum cuttings of the same stock, which had been imported in the same week as the infected stocks, were traced and these were also inspected by local PHSI.

To control the WFT in the following crop (second crop), planted in early December 2002, four different chemical treatments, to simultaneously target different stages of the WFT life cycle were recommended. All insecticides recommended were to be applied as per UK label instructions. Product 'Calypso' (thiacloprid), which is a systemic and translaminar compound, was recommended to target the feeding larval and adult stages of the WFT. 'Decis' (deltamethrin), which acts by contact and ingestion toxicity, was recommended for application soon after the thiacloprid. A nicotine space treatment was recommended to target the flying adults, and 'Nemolt' (teflubenzuron), an insect growth regulator, was also included to target the moulting stages of the pest.

CSNV was detected in the second crop at a much reduced level, so statutory control measures continued in the third crop, which was planted in March 2003, following a second steam treatment of the soil in the area where the heaviest infection was observed. A revised schedule of insecticide treatments was then recommended. 'Temik' (aldicarb) was to be applied to the soil, as the systemic nature of this product would provide activity against the leaf-feeding stages of the thrips. 'Malathion 60' (malathion), a contact insecticide, was used together with 'Dynamec' (abamectin), which has a contact and ingestion action against WFT. A space treatment of dichlorvos was used to target the flying adults, which would otherwise have escaped foliar treatments. Table 1 shows the timetable for spray applications.

Table 1. Timetable of spray applications for the third crop

Weeks no. after planting	Treatment
1	aldicarb
2	thiacloprid
3	malathion, dichlorvos*
4	abamectin
5	thiacloprid, dichlorvos*
6	
7	thiacloprid, dichlorvos*

*dichlorvos was applied as 3 applications in 9 days

Another new spray programme was devised for treatment of the fourth crop, which was planted in early June 2003, as there was still a small possibility of virus transmission at this stage, even though thrips numbers were lower (Figure 1). The control programme (Table 2) utilised products with different modes of action, as there were concerns that the WFT may be developing resistance to the older products, which had been used on the nursery for some time. Therefore two products with a contact action, 'Nico-soap' (nicotine) and 'Conserve' (spinosad), were added to the programme. It has recently been established that combinations of spinosad with other insecticides do not affect the efficacy of spinosad in controlling WFT (Warnock & Cloyd, 2005).

Table 2. Timetable of spray applications for the fourth crop

Weeks no. after planting	Treatment
1	nicotine, spinosad
2	spinosad, dichlorvos*
3	
4	nicotine, dichlorvos*
5	
6	spinosad
7	nicotine, dichlorvos*

*dichlorvos was applied as 3 applications in 9 days

It was decided that the outbreak of CSNV on the nursery could be considered eradicated when there had been no findings of CSNV in either plant or WFT for more than 135 days. This period of time was determined with reference to the life cycle of the WFT. For example, at 15°C it takes approximately 45 days for a WFT to develop from an egg to an adult, during which time it could obtain the virus from symptomless plants. The adults can live up to 90 days depending on the temperature (EPPO, 1997). The temperature in the glasshouse was kept at a constant 20°C which would increase the rate of development of the thrips compared to 15°C, but it was considered that 135 days would allow for a life cycle of the thrips to be completed, plus a safety margin.

RESULTS OF THE ERADICATION CAMPAIGN

The destruction of infected plants was carried out, and the sale of symptomless plants was permitted under official restrictions described previously. Inspection of the other glasshouses on the outbreak site and other sites which had received the same stock of plants in the same week as the infected nursery, revealed no further infection. Some removal of weeds from the glasshouse and surrounding area was carried out, but during later visits to the site, weeds, which could potentially harbour WFT, were observed.

The spray programme recommended for the second crop was modified by the local PHSI and the grower, to utilise insecticides that were already available to the grower. The revised programme included treatments with dichlorvos, malathion, deltamethrin and abamectin, as described above, and a low level of infected plants and WFT were found in this crop. Infected thrips were found on traps removed from the glasshouse on 26 November 2002, 18 December 2002, 8 January 2003 and 18 January 2003. The infection in the plants became visible at the end of February 2003 as the second crop reached maturity. These infected plants were confined to one small area of the glasshouse where the heaviest infection of virus had been located in the first crop. This suggested that the steam treatment applied after harvesting the first crop was not fully effective, resulting in the survival of CSNV-infected pupae in the soil. Alternatively, thrips may have survived on some nearby chickweed in the glasshouse, which had not been removed. The same action regarding the destruction of infected plants and the sale of symptomless plants was applied to this crop as was taken against the first infected crop.

Figure 1 shows changes in WFT numbers from February to July 2003, in relation to the timing of chemical controls implemented when the third crop was planted and continued after the

fourth crop was planted. This programme led to a general decrease of the WFT population, as revealed by trap catches over time, with a few exceptions which can be explained by the life cycle of the thrips. At points 'a' and 'b' in Figure 1, there was a marked increase in the numbers of WFT trapped; this was thought to be due to the fact that the insecticides applied prior to these peaks, were foliar treatments and therefore only targeted those stages present on the plants; i.e. they would have had no effect on pupae in the soil, allowing subsequent 'flushes' of adult thrips to emerge. It is also a possibility that the foliar treatments disturbed the population of thrips and stimulated adult flight, increasing the likelihood of their being captured on the traps. Regular testing of the trapped thrips for CSNV continued throughout this period but no infected thrips were found. No symptoms of CSNV were visible in either of these two crops.

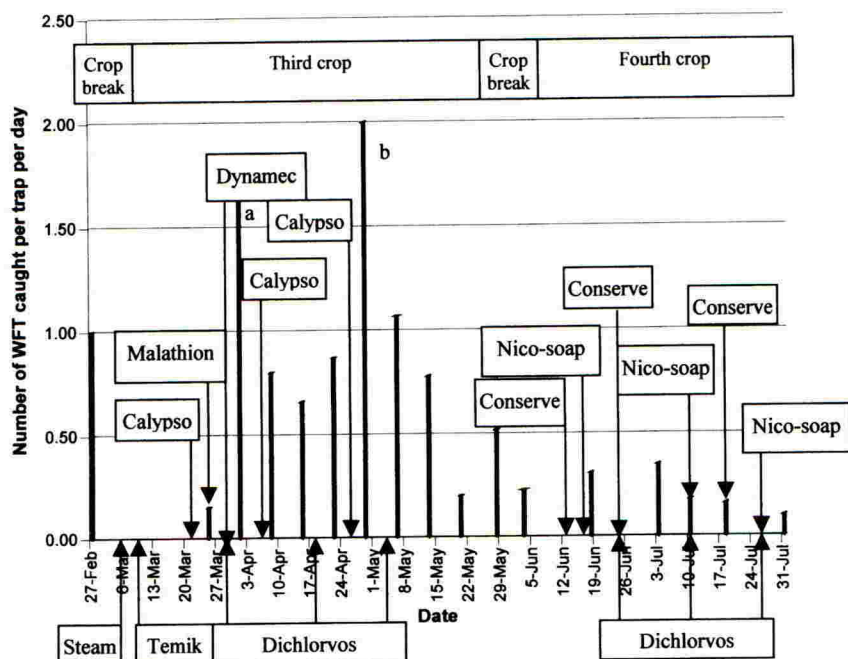


Figure 1. Number of WFT caught per trap per day and the spray programme implemented during the third and fourth chrysanthemum crops

The outbreak was officially declared eradicated on 14 August 2003. During the period when the third and fourth crops were growing, over 135 days (the maximum life span of the WFT) had passed since the last symptoms had been seen in the plants or since the last infected thrips had been trapped. The WFT had also been very nearly eradicated from the glasshouse. There have been no further outbreaks of CSNV at this site or elsewhere in the UK since this outbreak.

CONCLUSIONS

Although it is not possible to identify a single critical element in this eradication campaign, a few interesting conclusions can be drawn, in particular that action against the thrips vector

needs to be immediate and rigorous, and the grower needs to fully appreciate the importance of rapid and effective control of WFT. This type of eradication action is expensive and needs to take into account the grower's business needs, as far as possible. Although destruction of the whole crop and complete fumigation treatments of the glasshouse was considered initially, it proved possible to achieve eradication of the disease by effective management of the vector, thus avoiding the grower hardship, which would have been caused by complete crop destruction. A vital factor in this campaign was the development and utilisation of 'TaqMan' in determining if CSNV infection was still present in the vector. Without this procedure, it would not have been possible to monitor the progress of the outbreak, and to know, with confidence, that all the virus-carrying thrips had been eliminated.

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Interceptions and outbreaks of *Bemisia tabaci* in the UK

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ABSTRACT

The highly polyphagous whitefly, *Bemisia tabaci* was first found in the UK in 1987. There were 98 outbreaks that year, 95 of them at glasshouses growing poinsettias. The 'poinsettia strain', or B-biotype, vectors over 120 highly damaging plant viruses and is primarily a threat to protected cultivation (especially edible crops) in the UK. The UK maintains a protected zone for *B. tabaci* and is one of a small number of EU countries free of the pest. Although sporadic outbreaks do occur at protected ornamental propagators, they have always been eradicated, and outbreaks are usually short-lived. Poinsettias are imported as cuttings from a small number of EC countries. This remains the most prevalent pathway for the introduction of *B. tabaci* to ornamental nurseries in the UK. However, *B. tabaci* is also the most commonly intercepted quarantine pest on imported cut flowers and herbs from a large number of non-EC countries. The number of countries supplying ornamental plants to the UK is increasing, as flower-producing and other companies source a wider range of exotic material and also relocate propagation facilities to Asia, Africa and South America. Thus, the potential for introducing non-European and/or insecticide-resistant strains of *B. tabaci* – as well as new plant viruses – is likely to increase.

INTRODUCTION

The biology and ecology of the vector and viruses

Bemisia tabaci (Gennadius) (Hemiptera: Aleyrodidae), the tobacco whitefly, is a highly polyphagous plant pest of a very wide range of protected ornamental and vegetable crops (Smith *et al.*, 1997) and a vector of over 120 plant viruses, almost none of which are present in the UK (Jones, 2003). *B. tabaci* is a cosmopolitan pest, found in Africa, Asia, Australasia and Central America; also in several South American countries and in the southern parts of North America. In the UK, *B. tabaci* can only survive/establish in the long-term on protected crops.

In vegetable growing areas of the EuroMed region, infestations of *B. tabaci* have led to the introduction of a number of new viruses, with devastating results. The severity of symptoms is variable, depending on host plant, cultivar and level of vector control, but *B. tabaci* appears to be the principal agent responsible for the expansion in the geographic ranges of these viruses (Jones, 2003). For example, it is estimated that as many as 64 viral pathogens transmitted by *B. tabaci* naturally infect tomato (D. Jones, *pers. comm.*). Tomato yellow leaf curl virus (TYLCV) spread throughout tomato crops in the south and southeast of Spain in the early 1990s and appeared for the first time in Portugal (Algarve) in 1995 (Bedford, 2000). Cucumber and pepper crops are also at risk from *B. tabaci*-vectored viruses: including Curcubit yellow stunting disorder virus (CYSDV) and Cucumber vein yellowing virus

(CVYV). There are also reports from Spain of a new *B. tabaci*-transmitted yellowing disorder in French bean (*Phaseolus vulgaris*) (Segundo *et al.*, 2004). Thus, maintaining the *B. tabaci*-free status of protected salad crops, especially cucumber and tomato, in the UK is a major policy of Defra Plant Health Division.

Origins and spread

Bemisia tabaci was originally known as a pest of cotton and other tropical or sub-tropical crops such as cassava, sweet potatoes, tobacco and tomatoes. However reports of a newly evolved strain appeared in the mid-1980s and, by 1988, what became known as the B-biotype had spread to almost every poinsettia (*Euphorbia pulcherrima*)-growing region in the US (Brown *et al.*, 1995). *B. tabaci* is now known to be a species complex – undergoing evolutionary change – with many different biotypes. Populations of the B-biotype have high levels of similarity, suggesting that it has been spread throughout the world by contaminated propagation material (Guirao *et al.*, 1997). First reports of the pest in northern European countries – including the UK, The Netherlands, France and Germany – were in 1987 on imported ornamentals, mainly poinsettias. By the early 1990's, the B-biotype had become well established in Italy, France and Spain, all of which exported poinsettia cuttings to the UK. The pest also became established in the Netherlands, where it occurs on a wide range of ornamental and salad crops, including tomatoes, cucumbers and sweet peppers (Fransen, 1994). In 2003, *B. tabaci* occurred on nearly all the pepper nurseries in Westland (NL), as well as on a few tomato nurseries (Anon., 2003). There has only been one known outbreak of *B. tabaci* on a salad crop in the UK: low levels were detected on a cucumber crop in the Lea Valley in 1998. The outbreak was eradicated.

The quarantine status of *Bemisia tabaci*

Bemisia tabaci is now established in most countries in Europe. There are however, a handful of EU countries – Finland, Ireland, certain regions of Portugal, Sweden and the UK – that remain free from establishment and maintain a protected zone under Annex IB of the Plant Health Directive (2000/29/EC). In these so-called Protected Zone (PZ) countries, there is a policy of eradication whenever *B. tabaci* is found. As a quarantine-listed organism, *B. tabaci* presents a number of challenges: including the existence of several biotypes with different degrees of host specificity, and insecticide resistance. A particular challenge faced by the UK Plant Health and Seeds Inspectorate (PHSI), when faced with inspecting tens of thousands of imported plant cuttings, is detecting *B. tabaci*; particularly eggs. If *B. tabaci* remains undetected on an initial inspection of cuttings, the pest may rapidly spread to other hosts, and if the cuttings have already been 'stuck', the costs of destroying the imported material and the chances of an outbreak, increase greatly.

INTERCEPTIONS AND OUTBREAKS IN THE UK

The first outbreak of *B. tabaci* in the UK occurred in 1987; there were 98 outbreaks, all on poinsettias, predominantly from The Netherlands (Bartlett, 1992). The following year (1988), there were 87 outbreaks, again predominantly on poinsettias from The Netherlands. However, in 1989 the number of outbreaks declined markedly, to just 15 (Cheek & MacDonald, 1993). Numbers of outbreaks then increased in the period, 1990-91, before declining again, to 36 in 1993 (Cheek & MacDonald, 1994). The first outbreak on *Begonia* occurred in 1990. The

number of outbreaks of *B. tabaci* in recent years is shown in Figure 1; not all of the outbreaks were associated with an identified interception. Some of the outbreaks were protracted, and difficult to eradicate, particularly those in permanent indoor tropical landscapes where there is no crop break. However, all were – or are – in the process of being eradicated.

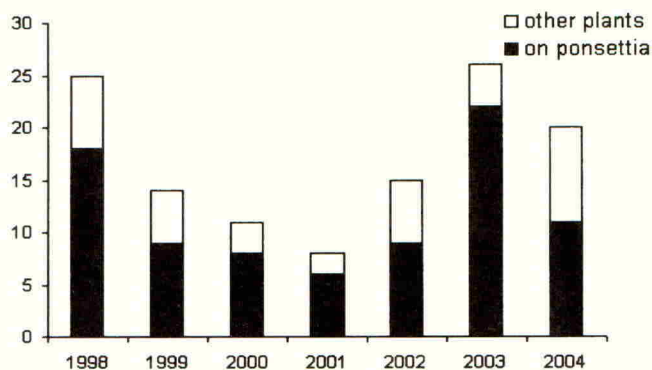


Figure 1. The number of outbreaks of *Bemisia tabaci* in England and Wales (1998-2004) and the proportion attributed to the import of poinsettia plants

In recent years (2000-04), the majority of interceptions of *B. tabaci* on poinsettia plants in the UK have originated from a relatively small number of countries: mainly The Netherlands, UK (internal movements), Portugal, Germany, Denmark, Kenya, Italy, Sweden, Spain and the USA (Table 1). There were more interceptions on plants from the Netherlands than any other country in 2000-2004. Other identified foreign sources of outbreaks (2000-04) were *Ajuga*, *Lamium*, *Fuchsia* and *Lavandula*, all from Israel; *Hibiscus* from the Netherlands; *Dipladenia* from Italy; *Antigonon leptopus* from Australia; and *Psidium guajava* from the USA.

Table 1. Interceptions of *B. tabaci* on poinsettia (*Euphorbia pulcherrima*) in England and Wales from 1996-2004

YEAR	EU countries									Non-EU			TOTAL
	NL	UK	PT	DE	DK	KE	IT	SE	Others	SP [‡]	US	Others	
1996	8	10	0	1	0	0	0	0	0	0	0	1	20
1997	12	10	1	9	1	0	0	0	0	0	3	0	36
1998	23	12	13	6	3	0	0	0	1	0	0	1	59
1999	16	17	0	19	8	0	0	0	0	1	0	0	61
2000	29	32	22	5	0	0	2	0	0	0	3	2	95
2001	20	3	2	2	0	0	6	0	0	0	0	0	33
2002	27	4	0	7	0	18	0	0	0	0	0	0	56
2003	47	29	14	8	3	0	0	8	0	0	0	1	110
2004	22	13	0	15	3	0	3	9	1	5	0	3	74
Total	204	130	52	72	18	18	11	17	2	6	6	8	544

[‡] = Canary Islands which are non-EU for plant health

It is important to note however, that the numbers of interceptions on imported poinsettias is significantly out-weighted by the total number of interceptions of *B. tabaci* on all other hosts. *B. tabaci* is intercepted on a very wide range of plants from many different countries: for example, in 2004, on 51 different host plants from 24 countries. During the period, 1996-2004, fifteen plant trades accounted for 59% of all interceptions. Top of the list were poinsettia and *Solidago*, which together accounted for 46% of import interceptions in this period. Other significant hosts, include *Ajuga* and *Hibiscus*. Israel and The Netherlands were the countries of origin of the highest number of *B. tabaci* infested plants, accounting for 67% of all interceptions. However, many of these interceptions were on 'lower risk' produce – e.g. cut flowers, which are rapidly 'consumed'. Plants imported from Israel (*Solidago*, *Ajuga*, *Trachelium*, *Hypericum* and *Gypsophila*) accounted for 34% of interceptions in 2001, but less than 10% in 2003 & 2004. In contrast there was an increase in the number of other Third (i.e. non-EC) country interceptions.

ERADICATION MEASURES FOR *BEMISIA TABACI* IN THE UK

Chemical control and insecticide resistance

The elimination of outbreaks of *B. tabaci* on poinsettias, or mixed ornamentals, relies on chemical control programmes, since pest eradication in these circumstances is generally incompatible with the use of biological control agents (Cheek & Cannon, 2003); although there are opportunities for biological control in managing alien pests (Cheek & Cannon, 2002). The approach to eradicating *B. tabaci* in ornamental crops in the UK is to utilise a combination of 'space' and soil treatments, together with foliar sprays, aimed at targeting all of the different life-stages of the pest. Imidacloprid is one of the preferred treatments, and can be applied as a drench and a granule; the systemic insecticide is taken up by the plant and controls larvae and adults. Foliar applications of imidacloprid – such as 'Confidor' or 'Provado' – which are widely used in vegetable cultivation in Spain, are not registered in the UK. Once the plants have enough roots to be self-sufficient, foliar sprays may be used: preferred products are 'Applaud' (buprofezin), 'Calypso' (thiacloprid) and 'Oberon' (spiromesifen). Other options include 'Chess' (pymetrozine), 'Nicosoap' (nicotine) and 'Nemolt' (teflubenzuron), 'Eradicoat' (a combination of starch and plant oils), 'Agri-50e' (hydrated propylene glycol alginate) and 'Mycotal' (*Lecanicillium muscarium*). Other insecticides with reportedly good efficacy against *B. tabaci* – but which are not registered in the UK – include pyridaben and pyriproxifen; the benzoylphenyl ureas, diafenthiuron and novaluron; and acetamiprid – a second-generation chloronicotinyl insecticide.

Intensive use of insecticides has resulted in reduced susceptibility of *B. tabaci* to several chemical classes, which can affect control programmes in the UK if the pest originates from regions where insecticide use is intensive. The extent of the resistance however, is often highly variable between populations, over time and from site to site (see pesticide-resistant arthropod database: <http://www.pesticideresistance.org>). Worldwide, there are many reported incidences of imidacloprid resistance in *B. tabaci*, including greenhouses in Italy, Spain, Greece and Germany (e.g. Gorman *et al.*, 2003). In general however, neonicotinoid-based products have proved relatively resilient and genuine cases of resistance are rare and relatively localised (Denholm *et al.*, 2002).

DISCUSSION

Despite regular interceptions and outbreaks of *B. tabaci* in the UK, it has not become established, and the protected zone (PZ) for this pest remains in place. The maintenance of the PZ involves striking a balance between avoiding the potential costs to UK salad growers, and mitigating the costs of eradication incurred by ornamental growers. Nevertheless, the benefits of continuing with the present policy considerably outweigh the costs of implementing it (Morgan & MacLeod, 1996). Furthermore, the *Bemisia tabaci*-free status of UK tomato, cucumber and pepper crops enables the industry to utilise IPM systems with judicious use of chemicals. Efforts are underway to reduce the costs borne by the flower and herb propagation sector in relation to interceptions and outbreaks of *B. tabaci*, e.g. by carrying out research aimed at developing more effective and robust disinfestation treatments for poinsettia cuttings.

The damage caused by this pest has already been seen in Mediterranean Europe. Sporadic outbreaks of TYLCV have occurred in France since 1999, with an increasing number of new outbreaks occurring in 2003. *Tomato chlorosis virus* (ToCV) is also spread by *B. tabaci*, and tomato crops in Spain (Malaga and Almeria) were first affected in 1997 (Navas-Castillo & Moriones, 2000). The economic threat to tomato production in heated glasshouses in the UK was estimated by Morgan & MacLeod (1996) to be approximately £11.5m, should the pest become established. *Cucumber vein yellowing virus* has been widespread in Europe and the Middle East for many years, but only emerged as a serious disease in cucurbit crops in southern Spain in 2000, and on melons and other crops in Portugal (Algarve) in 2002 (Louro *et al.*, 2004). Detection of viruliferous *B. tabaci* using 'TaqMan' assays, has confirmed the presence of TYLCV on pupae intercepted on cut flower produce and whole plants from outside the EU (R. Mumford, CSL, *pers. comm.*), demonstrating the presence of a real threat to tomato production in UK glasshouses.

The potential difficulties that salad growers would face from an outbreak of *B. tabaci* will increase as more growers use artificial lighting and heating to facilitate all year round (AYR) production. There is also a general trend in the UK towards larger glasshouse complexes for ornamental and salad production, and potentially AYR cucumber production (Shaddick, 2003), which will make it more difficult to control any outbreaks. The importation of poinsettia has remained the most common source of *B. tabaci* outbreaks (see Figure 1) and the host plants on which the pest was most commonly intercepted from 1996-2004 were poinsettia and *Solidago*. From 1996-2004, 42% of all interceptions were on plants and produce from Israel and 26% were on plants and produce from the Netherlands. However, when only interceptions at growing sites are considered, there were more on plants from the Netherlands than any other country every year from 2000-2004; whereas interceptions on plants from Israel appear to be declining.

Another recent trend is an increase in interceptions at growing sites on plants that have been imported from Third countries. Such interceptions have a high plant health risk, because of the potential of introducing non-European strains, or non-indigenous viral diseases, to the EU. One of the reasons for this change is the increasing numbers of flower producers based in the Netherlands and other European countries that are relocating production facilities outside of the EU, especially to Asia, South America and Africa. The Dutch flower auctions serve as the hub for about half of the world production of flowers and plants. Thus, there is no indication from overall trends in interceptions and outbreaks that the threat to UK plant health from *B. tabaci* is

declining and so a continuation of the present inspection and eradication measures will be necessary to keep the UK free of the pest.

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The analysis of detections in consignments to identify and target pests' entry pathways

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ABSTRACT

An analysis of detections of quarantine pests in consignments of imported commodities of plant produce was used to identify pathways that present a phytosanitary risk. Of 263 consignments contaminated with quarantine pests in England and Wales during 2001, almost 70% were cut flower consignments, particularly *Solidago* and *Gypsophila*. The most frequently detected quarantine pest was *Bemisia tabaci*. Such analysis provided evidence for changes in the perception of the risks posed by specific commodities to European crop production and led to amendments of European Community Plant Health legislation. From April 2003 *Solidago* and *Gypsophila* cut flower imports have required a phytosanitary certificate for entry into the EU. A comparison of contaminated consignments of *Solidago* and *Gypsophila* before and after the legislative changes shows a significant drop in the incidence of quarantine pests, from 7% of consignments contaminated to 3%, indicating that the measures introduced are having the desired effect.

INTRODUCTION

The international trade in plants and plant product commodities acts as the primary conduit for the unintentional introduction of non-indigenous pests (Levine & D'Antonio, 2003). For example, 82.6% of non-indigenous pests that established in the USA between 1980 and 1993 were judged to have entered the USA unintentionally through international trade (Jenkins, 1999). Pimentel (2002) estimated that globally the damage caused by non-indigenous pests, together with control costs, may exceed \$314 billion per year. The World Trade Organization (WTO) allows member countries to protect the life and health of their people, plants and animals from the risks of trade, such as pest introductions, arising from the importation of commodities by applying protective measures to trade pathways. The WTO Agreement on Sanitary and Phytosanitary Measures requires that protective measures are, amongst other things, scientifically based and are the least trade-restrictive (Anderson *et al.*, 2001). One simple phytosanitary measure is to inspect imported products and look for pests. However, due to the huge volumes shipped, it is impractical and unjustifiable to inspect everything. The USA quarantine agency has sufficient resources to inspect 2% of incoming shipments (National Research Council, 2002) whilst in New Zealand no more than 18% of shipping containers can be inspected (Everett, 2000). Yet these are countries perceived to guard themselves with a high level of phytosanitary protection and where the concept of taking action to prevent damaging species introductions is very apparent. Because not all imports can be inspected, there is a need

to target inspection effort. With the volume and frequency of plants and plant commodities that are imported worldwide increasing annually, the available resources for phytosanitary inspection must be utilised efficiently to achieve the objectives of National Plant Protection Organisations (NPPOs). Identifying pathways with a history of carrying pests is therefore a strategy that benefits resource managers and enables them to plan import inspections.

The EU Plant Health Directive 2000/29/EC (Anon., 2000), effectively divides growing plants and plant produce such as fruit, vegetables and ornamental cut flowers into three categories. First, there are those that present a very low risk of facilitating the spread of pest species into the EC and which can therefore enter the EC without any phytosanitary controls. Secondly, there are those that can present some risk, but the risk is mitigated if criteria set out in the Plant Health Directive are met, such as being examined before export to ensure the commodity is free from quarantine pests. If the criteria are met, then a phytosanitary certificate can be issued by the NPPOs of exporting countries indicating that the commodity meets the plant health requirements of the importing nation (FAO, 2001). Thirdly, there are those commodities that present such a high risk of carrying pests that the only satisfactory phytosanitary measure is to prohibit the import of the commodity entirely. Commodities in the second category requiring a phytosanitary certificate are routinely inspected by the England & Wales Plant Health & Seeds Inspectorate (PHSI) and quarantine pests are found occasionally (Malumphy, 1996). To ensure that low risk material, which is allowed free access to the EU, is in fact free from harmful pests, PHSI also inspect a proportion of such commodities at airports and docks and submit suspect pests that are found to the Central Science Laboratory (CSL) where organisms are identified. Data pertaining to each inspection are held in a PHSI database. Those data can be compiled to identify pathways carrying pests. A pathway provides the means of pest entry (FAO, 2002) and can be described using various descriptors, commonly the country of origin, country of import, and the commodity (MacLeod & Baker, 2003). By targeting the pathways with the highest numbers of quarantine pests and then taking appropriate action, the NPPO can focus limited resources against recognised pests. This paper describes the use of records of detections in consignments in England & Wales to identify those pathways, previously perceived to be of low risk, that transport quarantine pests. The effectiveness of introducing phytosanitary measures designed to inhibit the entry of quarantine pests on these pathways is also investigated.

MATERIALS AND METHODS

Identifying pathways on which quarantine pests are found

PHSI inspection data for 2001 and for April 2002 to March 2004 were extracted from the PHSI import inspection database. The data were imported into an Excel spreadsheet and checked, e.g. to remove blank rows. The data consisted of seven fields, i) a unique reference code, ii) date of inspection, iii) genus of plant / plant product inspected, iv) species of plant / plant product inspected, v) country of origin, vi) taxa of organisms detected in consignments either to family, genus or species level, and vii) plant pest status of organisms detected. Organisms that are not plant pests or are already widely distributed in the UK were excluded from further analysis. Once the raw data had been checked and filtered, the pathways on which they were detected were determined using the 'Pivot table' function in Excel.

Assessing the effectiveness of phytosanitary measures acting on pathways

Inspection records and details of quarantine pests detected in imported consignments of *Gypsophila* and *Solidago* cut flowers during the period April 2000 to June 2005 were extracted from the PHSI database. The data were grouped into monthly data and the proportion of consignments contaminated with quarantine pests was determined. The mean proportion of contaminated consignments during the 36 months, April 2000 to March 2003, were compared to the mean proportion of contaminated consignments between the 27 months, April 2003 and June 2005. Statistical comparison of the mean proportion contaminated was based on 'Agresti-Coul' confidence intervals (Brown *et al.*, 2001).

RESULTS

Identifying pathways on which quarantine pests are found

In 2001, pests were found on 721 consignments of 85 plant genera from 44 non-European countries. Pests were diagnosed on 161 pathways; the majority of these had only one contaminated consignment. Over 50% of contaminated consignments were from 20 (12.4%) of the 161 pathways. In 2002-4 there were 889 contaminated consignments of 103 plant genera from 53 countries, representing 152 pathways. Again the majority of contaminated consignments occurred on a minority of the pathways. Of the quarantine pest species identified, *Bemisia tabaci* was found most frequently. Over 85% of detections of quarantine pests occurred in consignments of cut flowers. *Solidago* and *Gypsophila* cut flowers were responsible for over half of all contaminated cut flower consignments (Table 1).

Table 1. Genera of imported cut flowers found during import inspections to harbour quarantine pests, January to December, 2001, and financial years 2002/3 and 2003/4

Cut flower Genus	Country of origin	Quarantine pests detected	No. consignments contaminated		
			2001	2002/03	2003/04
<i>Solidago</i>	IL, IC	<i>Bemisia tabaci</i> , <i>Liriomyza huidobrensis</i>	66	48	37
<i>Gypsophila</i>	IL	<i>Bemisia tabaci</i> , <i>L. huidobrensis</i>	40	23	9
<i>Lisianthus</i>	IL, KE, CO	<i>B. tabaci</i> , <i>Helicoverpa armigera</i> , <i>L. huidobrensis</i> , <i>Spodoptera littoralis</i>	7	1	6
<i>Hypericum</i>	IL, ZW	<i>B. tabaci</i>	6	7	1
<i>Aster</i>	IL, IC	<i>B. tabaci</i> , <i>L. trifolii</i>	5	0	2
<i>Bupleurum</i>	IL, ZW	<i>L. huidobrensis</i>	5	2	0
<i>Carthamus</i>	IL, KE	<i>L. huidobrensis</i>	5	0	2
24 others	19 others	<i>B. tabaci</i> , <i>H. armigera</i> , <i>L. huidobrensis</i> , <i>L. sativae</i> , <i>L. trifolii</i>	50	21	32
Total cut flower consignments contaminated with quarantine pests			184	102	89

Key: CO Colombia, IC Canary Isles, IL Israel, KE Kenya, ZW Zimbabwe.

Similar analysis by MacLeod (unpublished data) was used by the EC Standing Committee on Plant Health (SCPH) as evidence to support Commission Directive 2002/36/EC of 29th April 2002 that added *Gypsophila* and *Solidago* to Annex IVA1 and VB of EU 2000/29/EC. Plants and plant produce listed in the Annexes of EU 2000/29/EC require a phytosanitary certificate to enter the EU. The April 2002 Directive came into force on 1st April 2003.

Assessing the effectiveness of phytosanitary measures acting on pathways

By comparing the detections of quarantine pests found on *Solidago* and *Gypsophila* before and after they became regulated commodities, the effectiveness of the phytosanitary measure could be determined (Figure 1).

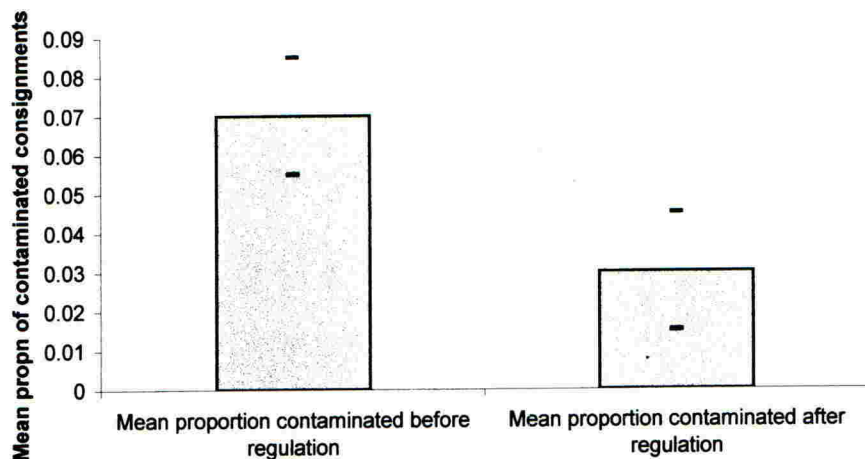


Figure 1. The mean proportion of inspected consignments of *Gypsophila* and *Solidago* cut flowers, contaminated with quarantine pests, before and after these cut flowers became regulated

Figure 1 shows that regulation has had a significant impact in reducing the proportion of contaminated *Gypsophila* and *Solidago* cut flower consignments imported into England & Wales. Following regulation, the mean level of contamination has fallen from approximately 7% to approximately 3%. Based on 'Agresti-Coul' confidence intervals (Brown *et al.*, 2001), the mean proportion of contaminated consignments after regulation is significantly lower than the proportion of contaminated consignments before regulation ($p < 0.002$).

DISCUSSION

This work has shown that examining historical data can identify those pathways carrying quarantine pests, enabling risk managers to allocate scarce resources to target pathways appropriately. The work also quantifies the impact that a specific phytosanitary measure has had in reducing detections of quarantine pests on particular pathways.

There are a number of caveats to be borne in mind when considering the data and its analysis in this report. Specifically, there are large differences in the volume of each commodity imported. Only trades where pests have been detected are included. Pathways on which no pests were found are not shown. Except for the analysis of the *Solidago* and *Gypsophila* pathways, the total number of inspections on each pathway was not available, consequently there is no indication of the proportion of pest-free consignments.

Historically, the perceived risk from importing pests on produce such as cut flowers has not been considered to be great since it was assumed that most produce was purchased relatively quickly from retailers, by the public and then effectively destroyed, through use, by consumers. This was thought to severely limit the probability of pests becoming established. Hence no action was usually taken on produce found to carry pests. However, the public now demand year round supply of some commodities and growers/ producers are importing produce to supplement or replace domestic production. Importing produce which carries pests into plant nurseries provides a clear pathway for new pests to enter and potentially establish. It is becoming increasingly clear that cut flowers are being taken to places of plant production where they can be held in close proximity to growing crops before being combined with domestic production to fill customers orders. In 1998, there were at least 70 plant nurseries where both imported cut flowers and pot plants were handled. However, providing firm evidence that an outbreak of a quarantine species was caused specifically as a consequence of importing contaminated material is very difficult due to the time lags between import and detection of outbreaks. Nevertheless, there have been some outbreaks in the UK where imported cut flowers were strongly suspected of being the source of the pests. For example, a 1991 outbreak of *Spodoptera littoralis* in southern England, was believed to have originated from Israeli imports; a long running outbreak of *Liriomyza huidobrensis*, in south-east England, in 1995/96 was attributed to imported cut flowers.

There have been other examples of the effect of legislation on interceptions, for example, a separate analysis of EU wide detections of *Thrips palmi* (Thysanoptera: Thripidae) in consignments of cut flowers between 1995 and 1997 led to tightening of import regulations for Thai orchids in 1998 (MacLeod & Baker, 1998), since when there have been fewer interceptions of *T. palmi*. Such analyses are a key component of plant health strategy and should be carried out routinely for all pathways.

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