

PCN grower guide



Foreword

UK potato growers produce exceptional high yielding crops that satisfy the demands of discerning and sophisticated consumers looking for quality and value. The grower base has witnessed considerable concentration over recent decades into highly specialised agri-businesses utilising the latest innovations in growing techniques. But for much of the UK land growing potatoes, quality is dependent on effective Potato Cyst Nematode (PCN) control. This Guide is a welcome update on the latest understanding and knowledge of PCN management and control. It brings together the latest scientific knowledge, as well as practical experience of sustainable integrated crop management techniques in tackling the problem. This Guide is an excellent aid to growers, agronomists and the potato agri-supply chain.



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The information in this guide was compiled by Anne Stone, AHDB Potatoes, based primarily on the project report Potato Cyst Nematodes (PCN) – Their Characteristics and Guide to Management, AHDB Potatoes (2017). Authors: Matthew Back, Vivian Blok, Kim Davie, Ivan Grove, John Jones and Jon Pickup. Co-ordinator/Editor: Sue Hockland.

AHDB Potatoes is grateful to all those who have commented and contributed to this production.

Introduction

Of all pests and diseases of potatoes in Great Britain, infestation by potato cyst nematodes (PCN), or eelworms, is the most damaging. Yield losses depend on pest populations, soil type and potato variety, with a range of 1–35% in trials, with infestations at 10–20 eggs per gram of soil.

At low levels of infestation, no symptoms are seen in the field. The first visible signs are usually patches of stunted crop with weak vigour. Typically, plants fail to meet in the row. The infected plants have a poor root structure and are more susceptible to drought. When the roots are examined carefully in situ, cysts can sometimes be seen with the naked eye or a hand lens. There are two species: *Globodera pallida* and *Globodera rostochiensis*.

The spread of *G. pallida* and the withdrawal of many plant protection products has necessitated changed approaches to management. Since the issue of the Ministry of Agriculture, Fisheries and Food (MAFF) guide to PCN management in 1999, knowledge of PCN sampling and its development have advanced. This guide presents a summary of current distribution and an up-to-date description of control methods. Used in isolation, none of these methods will prevent infestation or eliminate established infestations. Recommendations are given for 'best practice' measures that will offer a sustainable strategy for PCN management in the long term, as part of an integrated pest management (IPM) programme.

Relevant regulations have changed since the introduction of Directive 2007/33/EC, the PCN Control Directive, which can be found at data.europa.eu/eli/dir/2007/33/oj

UK crop hosts of PCN are potatoes, tomatoes and aubergines, though, in practice, only potatoes are field-grown and suffer from infestation.

Components of integrated control

- Sample soil to detect the presence of PCN and, if confirmed, determine the PCN species and population levels because these will influence the choice of management options
- Extend rotations to at least eight years to reduce PCN levels
- Use certified seed potatoes, produced on land tested for freedom from PCN
- Ensure hygienic practices that limit the movement of soil, including that from graders
- Control volunteer potatoes
- Use varieties that are resistant to the species of PCN present
- Use trap cropping and biofumigants in the rotation
- Use a granular nematicide

To determine the effects of any combination of the above recommendations, assess population levels at Pi (initial egg numbers at planting) and Pf (final egg numbers after harvest) by laboratory analysis.

Each of these control methods and methods of assessment is described in detail later in the guide.



Figure 1. Normal potato plant on the left and PCN-infested plant on the right

Background

The PCN species *G. pallida* and *G. rostochiensis* co-evolved with the wild potato *Solanum tuberosum* ssp. *andigena* in South America several hundred thousand years ago. The ancestors of populations in Europe probably came from very few introductions; current research suggests the most likely source as southern Peru in the 19th century when varieties with resistance to potato blight were being collected to provide new breeding material. Contaminated sacks used for guano, military and agricultural equipment are also believed to have carried PCN around the globe.

There is evidence of physiological differences between the two species at certain phases of their life cycles. In terms of hatching, development and persistence (including the use of fat reserves), *G. pallida* is the species better adapted to low temperatures, thus it gains an advantage over *G. rostochiensis* in cool soils.



Figure 2. *G. rostochiensis*, yellow PCN, developing females on potato roots



Figure 3. *G. pallida*, developing females on potato roots



Figure 4. Mature PCN cysts against a ruler with 1mm markings

Life cycle

The PCN cyst is the dead body of the female nematode, containing up to 600 viable eggs, each of which contains a dormant juvenile. It is the main means of survival and spread for the species. The cysts are usually about 0.5mm in diameter; just visible to the unaided eye.

G. rostochiensis cysts have an early yellow or gold phase, while those of *G. pallida* have a white/cream phase (Figures 2 and 3). These colours are only visible for a short time before the females die and both ultimately form hard, dark brown cysts. They are primarily root parasites, although they may also occur on underground stems, such as stolons and tubers.

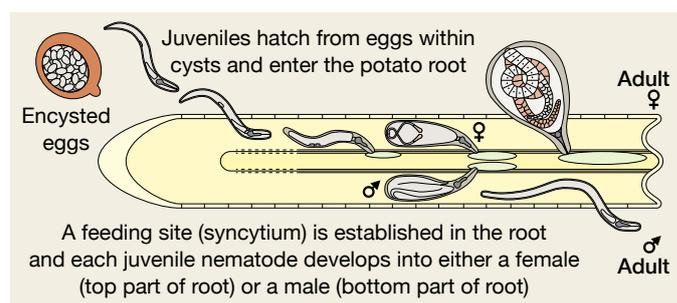


Figure 5. Life cycle of PCN

Hatching

The developing juveniles are protected from desiccation and parasites by their individual eggshells and the cyst, which enhances their ability to remain dormant for many years.

In the spring, up to a third of the eggs hatch spontaneously, even in the absence of a host plant, and the new juveniles can persist in the soil for several weeks. The remaining eggs that do not hatch in any one year are part of the PCN survival strategy. Viable juveniles can remain dormant within cysts in soil for as long as 40 years in exceptional cases. When potatoes are present, their roots produce substances called hatching factors, which gradually move into the surrounding soil and stimulate up to 90% of eggs to hatch. In a weak or poorly rooted crop, more eggs fail to hatch because root exudates do not reach all parts of the topsoil. *G. rostochiensis* hatches faster in response to these root exudates, while *G. pallida* has a slower initial hatch rate and is more responsive to plant chemicals released during later stages of growth. The hatching period usually lasts about 18 weeks in *G. pallida* and 12 weeks in *G. rostochiensis*, but there is great variation in hatching rates between different populations within both species.

Establishment in a host and reproduction

When juveniles reach a host plant they invade the roots, secreting enzymes that degrade the cell wall. They settle in the root tissue near to vessels that conduct food and water and form a 'syncytium', a complex food transfer cell. This causes much of the damage to the crop by stunting root growth and reducing uptake of nutrients and water from the soil.

The maturing female PCN feeds on the potato root and enlarges and, in doing so, ruptures the root and becomes exposed outside the plant root in the soil rhizosphere. She continues to feed during egg production. Pheromones from the female attract males that have left the roots. After fertilisation and death of the female, her body forms an egg-containing cyst. Multiplication of PCN is greater when the initial infestation is low. For example, in one series of experiments in Britain, populations of *G. pallida* multiplied between 46–100 fold from an initial population of fewer than 10 eggs per gram.

Some varieties do not support developing females, so reproduction is suppressed and fewer viable eggs are left in the soil for the future. These are known as 'resistant' varieties. There are no varieties that completely prevent PCN reproduction. Varieties with no adaptation to restrain formation of syncytia and reproduction are known as 'susceptible' varieties.

Some varieties are 'tolerant'; that is, they are able to withstand or recover from damage, so their yield is relatively little affected. Varieties that suffer most from feeding by juvenile nematodes are known as 'intolerant'; suffering marked yield reductions.

The implications of resistance and tolerance for control methods are described later in this guide, in the section on varietal resistance, where the outcomes of using resistance and tolerance are shown in Figure 19.

After the first generation there is no necessity for dormancy if the environmental conditions remain favourable. There is evidence that a second generation can occur within one season, when there is an extended period of warm, but not excessively hot soils.

Dormancy and decline

After the potato crop has been harvested, cysts remain and the eggs enter a dormant phase.

Decline of PCN in a field has two components. One is natural mortality, from causes such as parasitism. The second is from the spontaneous hatching of a proportion of the eggs each year; the juveniles from which will die if they fail to infest a host. The rate of decline of *G. pallida* in the UK is typically 20% – slower than that of *G. rostochiensis* at 30%. The rate of decline increases as soil temperature rises. Decline is fastest in sandy soils and slower in silt, clay or organic soils. In one study on a mineral soil with 100 eggs/g of *G. pallida*, it took 13 years for the population to decline to a level at which most varieties could be grown without yield loss.

The PCN calculator

The PCN toolbox calculator predicts populations based on factors including soil type, initial PCN population, rotation length, variety and nematicide use. The calculator is available on the AHDB Potatoes website (potatoes.ahdb.org.uk/online-toolbox/pcn-calculator). The results are not always directly applicable to any particular situation because of the complexity of factors influencing rate of decline, but using the calculator is a helpful exercise because it draws attention to key factors influencing populations on a specific farm and can inform long term rotational decisions.

Some factors adding complexity, which the calculator cannot yet account for, involve soils and climate. Hatching factors from potato roots move easily in sandy soils, increasing the hatch in such soils compared with those containing more clay. Potatoes are best able to withstand high PCN infestations with little yield reduction in deep silts and organic soils. There is also evidence that soils with a lower pH and higher nitrogen content support higher levels of PCN. With a changing climate, two generations of PCN in one year may become more common in both species.

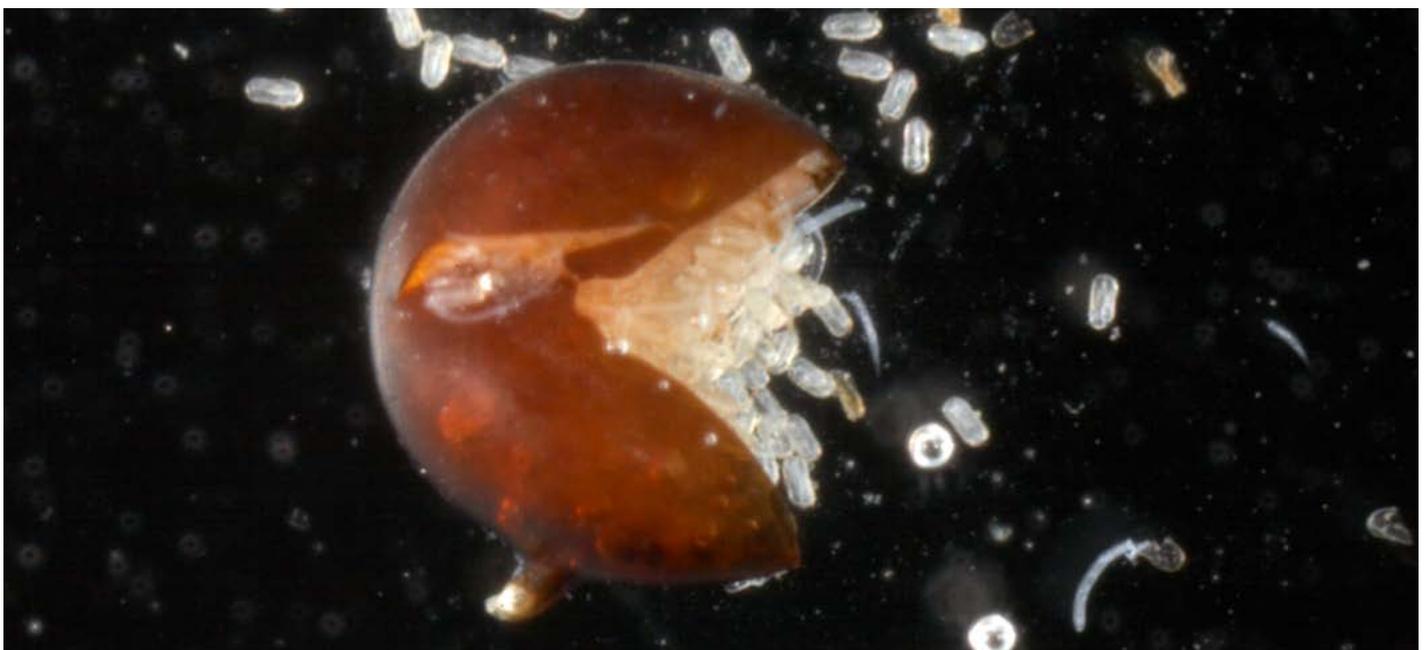


Figure 6. *Globodera* spp. cyst split to reveal eggs and juveniles

PCN prevalence surveys

Under the PCN Directive, testing for this quarantine pest is conducted each year by the statutory authorities: the Animal and Plant Health Authority (APHA) for England and Wales and Science and Advice for Scottish Agriculture (SASA) for Scotland. In some ways, the two surveys are the same; they each test 0.5% of ware production area each year in the season following potato production and fields are sampled at a rate of 400ml from 100 cores per hectare. All the soil is analysed and, if a field is found to contain PCN, that field is recorded as infested. Some aspects of methodology differ, so the results are not fully comparable and are presented separately. In both cases there are few sample numbers per county, so map illustrations are on a regional basis (see Figure 7a). The small areas tested in all PCN surveys, as a proportion of the total, mean the results must be treated with caution.

In Scotland, growers must register the fields in which they grow potatoes each year. SASA then applies a randomising algorithm to select fields for inspection, modified slightly to avoid returning to a farm more than once in six years. Within each field a unit of four hectares is selected, by agreement between the owner and inspector. Laboratory analysis uses a molecular method to detect the presence or absence of each species.

In Scotland, each hectare is sampled and analysed separately, though if any one part of the field contains PCN, the whole four-hectare unit is recorded as infested. It is common for one or more hectare sections to show absence of PCN, while one or more show presence. The map opposite presents results on a field basis rather than a sample basis. From the total of 563 samples, 14% contained *G. pallida* and 6% contained *G. rostochiensis*, with an overall PCN incidence of 18%, because some fields contained only one species while others contained both.

Each year in England and Wales, APHA receives a dataset of growers from AHDB and allocates them to regions. Within each region, there is random selection of growers. Selection of the fields to be tested is by agreement between the grower and inspector. When a field is selected, the whole of it is tested, in one-hectare sections. In the laboratory analysis at Fera, cysts are separated from all the one-hectare samples of one field as a single batch. All *Globodera* cysts are microscopically assessed for viability and counted; then, viable cysts are identified to species using molecular methods.

Figure 7b illustrates the average percentage of fields with PCN in the main potato-growing regions of England and Wales. PCN was detected in 50% of fields sampled. Of those fields where viable cysts were found, only 2% contained *G. rostochiensis*. There was very little overlap between the two species and 98% of fields with viable cysts contained only *G. pallida*.

Research survey of England and Wales

In addition to the statutory tests described above, a survey was completed in 2016¹; the first such survey since 2000. Between 2000 and 2016, the proportion of total samples found to be infested with PCN reduced from 64% to 48%. The most likely reason for this is the near-complete control of *G. rostochiensis* achieved at many sites following the use of several varieties with resistance to this species. In consequence, *G. pallida* has increased in dominance, as shown in Figure 8. It should be noted that although *G. rostochiensis* is seldom detected, small numbers may persist from previous infestations, with the potential to multiply in future if varieties lacking resistance to this species are grown.

The lack of strong *G. pallida* resistance in those pre-pack potato varieties currently grown causes a particular risk of increasing infestation in areas that specialise in the fresh market. Some areas, such as Wales and Southern England, are still relatively free of infestation, but this position is under threat.

¹K. Dybal unpublished (project supported by AHDB and Defra)

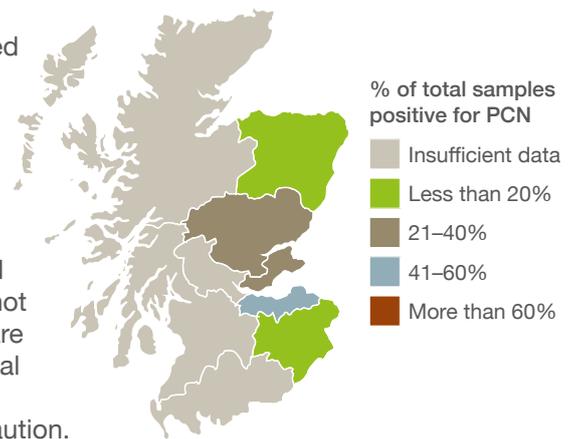


Figure 7a. PCN distribution on ware land in Scotland 2010–2016

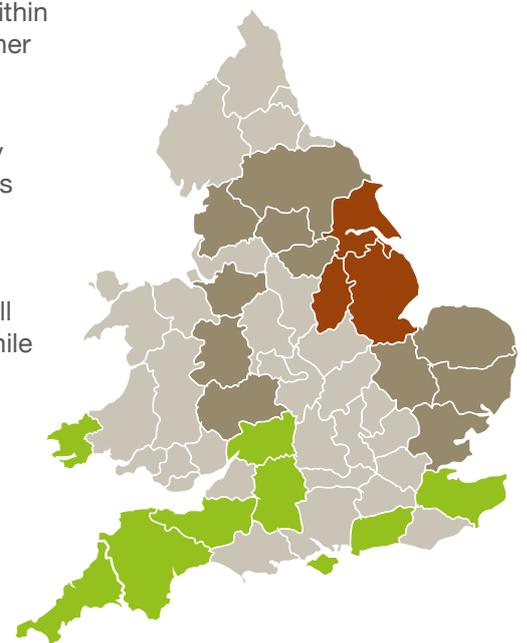


Figure 7b. PCN distribution on ware land in England and Wales 2012–2016

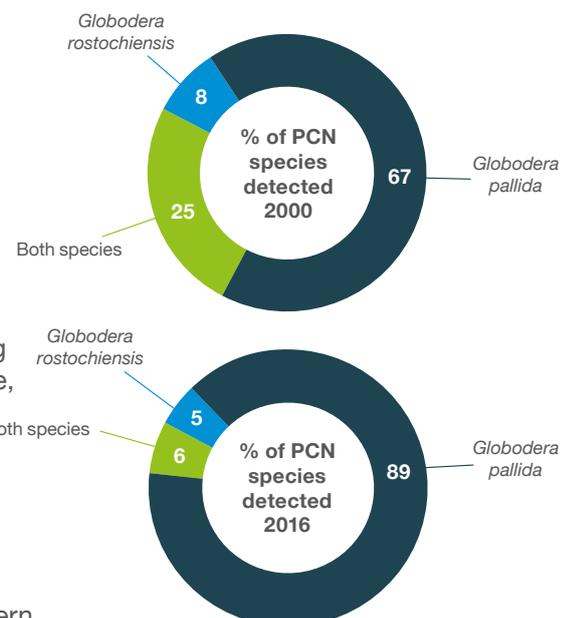


Figure 8. Changes in PCN species detected between 2000 and 2016

Distribution and soil sampling

Distribution of PCN across a field is neither uniform nor random. Following initial infestation at one or more points, the focus spreads in a patchy manner, giving rise to secondary foci. Figure 9 illustrates both the variation common within a field and the way it can change over a season. Foci, known as 'hot spots', are often elliptical in shape and spread in the direction of mechanical operations. Digital mapping using drones to record canopy development can indicate patches of a field with low vigour. Such areas often prove to be PCN foci.

Land for seed production must be free from PCN. Regulations relating to testing land for seed production are described opposite.

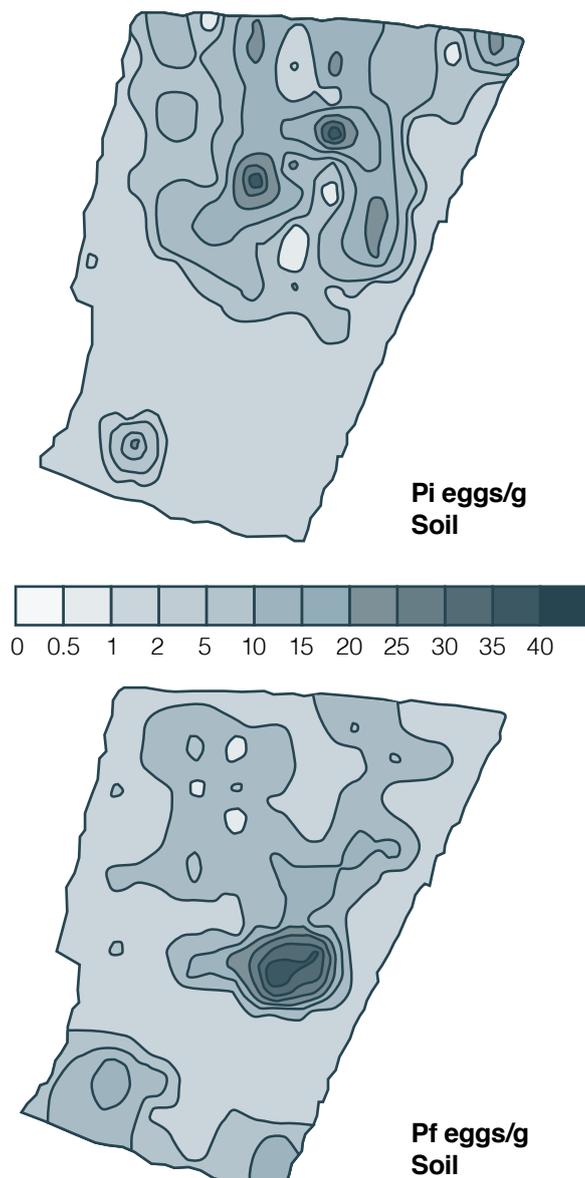


Figure 9. PCN egg numbers before and after a crop of potatoes in a field of 10.5ha

Sampling prior to seed crops

There are statutory regulations covering soil sampling for PCN in fields selected for potato seed production. These new arrangements came about in 2010 as a result of the implementation of Directive 2007/33/EC. It is a requirement of the Seed Potato Classification Scheme (SPCS) that all crops entered for inspection must be grown on land for which a PCN clearance certificate is in force at the time of planting. These certificates are issued by SASA in Scotland and APHA in England and Wales:

sasa.gov.uk/pcn-soil-testing-documents

gov.uk/guidance/the-seed-potato-classification-scheme

Sampling prior to ware crops

To manage PCN effectively, it is critically important to sample soil from fields prior to planting. This allows you to know whether PCN is present and, if it is, at what population density and what species, to guide management decisions such as nematicide use and variety. Research funded by AHDB has led to the development of standardised protocols for land destined for ware production.

In the field:

- Divide fields into blocks of one hectare or less for sampling
- Use of a corer with a 10–15mm diameter is recommended
- Take a minimum of 49 cores per hectare in a grid pattern
- Insert the corer to a depth of up to 25cm



Figure 10. PCN-infested field showing that hot spots are sometimes clearly delineated

A range of corer sizes is currently used. The diameter of the corer and the depth to which it is inserted into the soil determines the volume of soil collected. For example, a 13mm diameter corer inserted to a depth of 25cm and 49 cores taken per ha will generate about 1.626 litres of soil. All of the sample should be sent to the laboratory; no subsampling should occur in the field.



Figure 11. ATV used for soil sampling



Figure 12. Soil sampling machine attached to field vehicle

In the laboratory, there are different requirements depending on the purpose of the sampling. This is either:

- Detection of an early-stage infestation; or
- Estimation of the PCN population in later-stage infestations

Detection of early stage infestation

The larger the proportion of the field soil sample analysed in the laboratory, the greater the chance of detecting PCN.

Table 1 below shows the impact of subsample size on the probability of detecting early-stage infestations. It also shows how the likelihood of detecting infestations using 49 cores or 100 cores per hectare differs for a 400g sample. It can be seen that increasing the number of cores per hectare has less effect on detection than increasing the volume of soil analysed in the laboratory.

Estimation of the PCN population in later-stage infestations.

This applies when PCN is known to be present and the requirement is to know the number of eggs/g of soil so that management decisions (eg nematicide use, variety) can be made.

A key aspect of sampling for PCN is that eggs are not distributed uniformly, but are contained in cysts. The degree of concentration of eggs in cysts is itself not uniform and is affected by many factors. For example, there are more eggs per cyst when the previous potato crop had no resistance to the PCN species present, when the rotation was short and when there have been many volunteer potatoes.

Appendix 1 provides detail on how the clustering of eggs in cysts affects the uncertainty of test results.

Table 1. Comparison of the impact of subsampling in the laboratory on the probability of detecting 5 million cysts per hectare

Corer size diameter (mm)	Depth (cm)	Cores per ha	Approx volume (ml) of soil collected (per ha)	Approx dry weight soil (g)/ha**	Amount (g) of sample processed	Probability of detection (%)
13	25	49	1,626	2,600	2,600	67
13	25	49	1,626	2,600	1,000	48
13	25	49	1,626	2,600	600	38
13	25	49	1,626	2,600	400	31
13	25	49	1,626	2,600	200	20
13	25	100	3,319	5,310	400	39
Sampling specified under European Council PCN Directive (2007/33/EC) for seed potato land (provided by SASA and included for comparison):						
18	6	100	1,500	2,400	2,400	81
11	4	100	400	640	640	40

** Based on a bulk density of 1.6g/cm³

The AHDB project report's authors recommend that a representative subsample of at least 200g should be analysed in the lab. A subsample of 400g is recommended if the infestation is expected to be at a low population level, especially if the field has also recently been in potato cultivation, thus the eggs are likely to be highly aggregated within relatively few cysts.

A flow diagram summarising the standardised protocols is provided below (Figure 13).

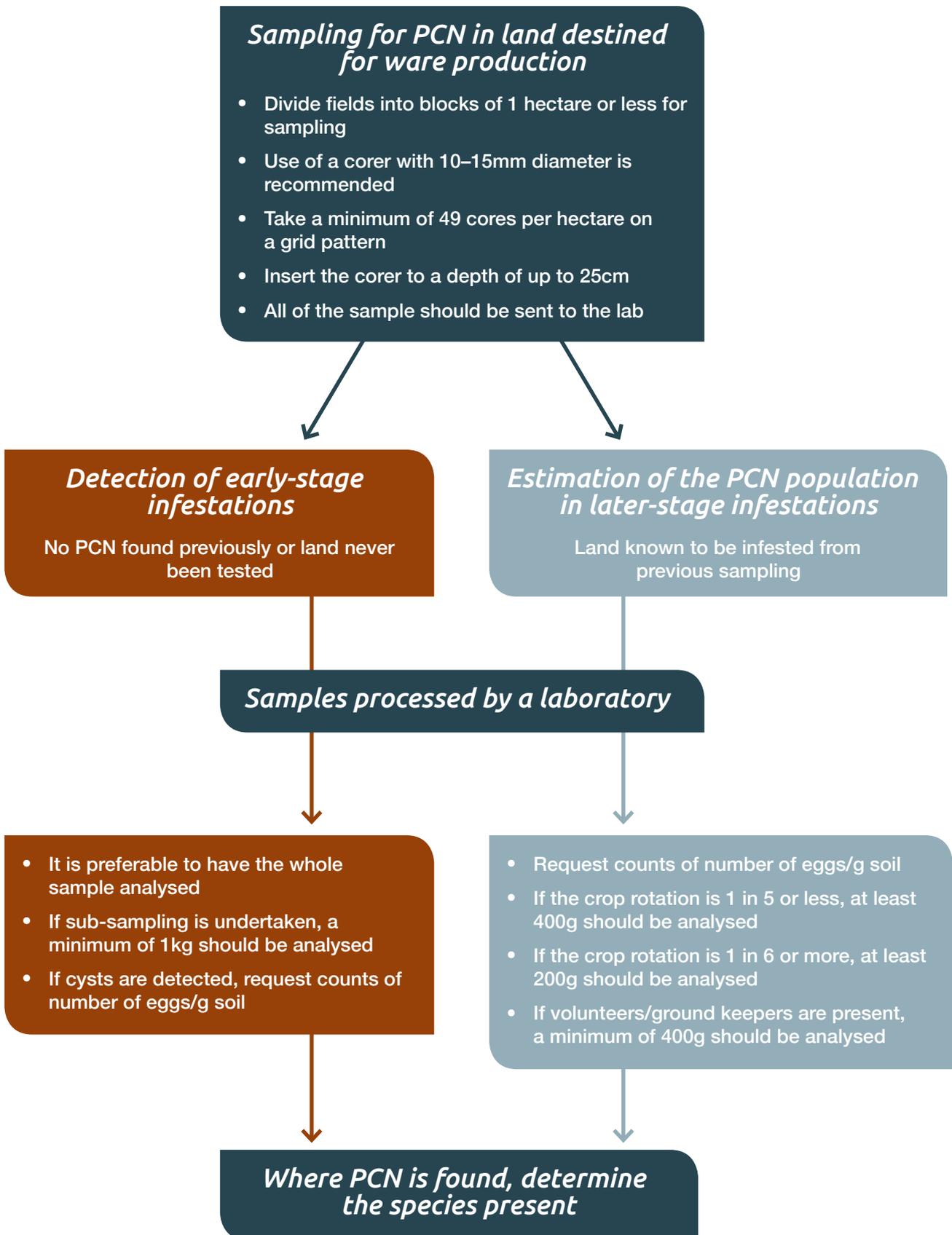


Figure 13. Sampling for ware in land destined for ware production



Figure 14. Fenwick can, in use to extract cyst nematodes from wet soil.

Whatever the method of testing used, clean the equipment between one sample and the next to avoid contamination.

The results from soil tests should be discussed with your agronomist/advisor and will affect the development of an IPM strategy. It is important to know the species of PCN present (*G. pallida*, *G. rostochiensis* or both). Potato varieties differ in their resistance to the two species. More information is available in the AHDB Potato Variety Database (varieties.ahdb.org.uk). Varieties also vary in tolerance; that is, how well their yield withstands PCN attack.



Figure 15. Wye washer, an elutriator for extraction of cyst nematodes from wet soil.

GPS-marked testing

If the locations of sampling are recorded using GPS, they can be returned to when sampling next takes place, or if an unexpected result is found that prompts re-sampling. This advantage of repeatability will improve accuracy in PCN population monitoring, with any changes less likely to be caused by random variation. If patches of damage in the crop indicate a hot spot that was missed by sampling, GPS marking allows for the sampling grid to be moved to include the affected zone.

There are different approaches to GPS-marked testing. For example, a trace around a hectare block may be recorded, without the location of every subsample point being recorded with GPS. Clarify with the sampling company the method that is to be used.

Timing

Historically, sampling has been done in autumn/winter, immediately before potato cropping. However, to enable rotations to be planned well in advance of potato planting, sampling can be done at other times within the rotation. For the greatest chance of detecting an early infestation, sampling should take place shortly after a potato crop, before natural decline reduces populations below the detectable level. Testing post-harvest also permits the efficacy of control measures to be assessed, although at this stage, clumping of cysts into hot spots is more likely. Distribution will be more uniform and so counts will be more reliable if samples are taken once several cultivation operations are completed after potato harvest. This ensures mixing of cysts within the soil profile.

Even when all of the above precautions are taken, errors are still inherent in sampling, so it is long-term data on trends and yields that provide growers with the best picture for cropping decisions in any particular field.

Thresholds for action

In the past, different forms of control were recommended depending on threshold infestation levels. Given the evidence that PCN can multiply from population levels that are not detected to highly damaging levels in one susceptible crop, even the slightest infestation should be taken seriously.



Figure 16. Delegates participating in the Potato Cyst Nematode identification workshop, SASA, 2016

Control methods

Rotation length

Although PCN can survive for many years, a proportion of the eggs die each year. The natural rate of PCN decline is greatest in the first 10 years. Research in the UK has shown that decline rates are typically 20% per year for *G. pallida* and 30% for *G. rostochiensis*. Figure 17 shows how these declines apply to two population densities of PCN commonly found after a susceptible crop.

In practice, the rate of decline can be reduced by the presence of volunteer potatoes. Also in practice, a wide range of decline rates have been observed experimentally, from 10–50% per year for *G. pallida*. This means it is more appropriate for decline rates in individual fields to be monitored rather than calculated.

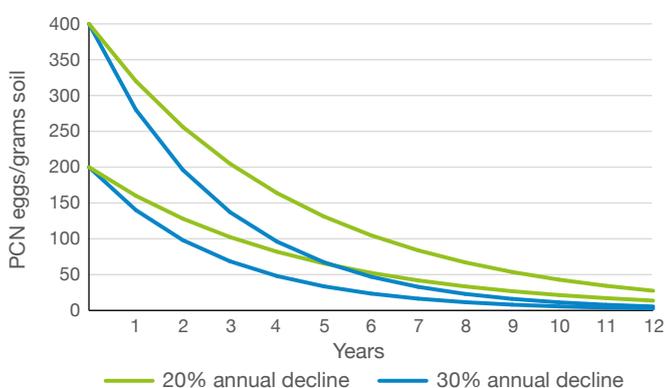


Figure 17. PCN decline curves

Seed potatoes

PCN cysts can be carried in soil, adhering to seed potatoes or in the eyes. As a major potential route to infestation, many aspects of testing and enforcement in relation to seed were altered following the adoption of the PCN Control Directive. Implementation varies with jurisdiction, as shown in Table 2 below.

There is a considerable risk of introducing PCN to a previously clean field if farm-saved seed is used from a field that was not rigorously tested for PCN prior to planting. The crop may not have shown any symptoms of infestation because the PCN population was relatively low. Soil adhering to seed tubers carries a particular risk. Infective juvenile nematodes move a maximum of about 1 metre in the soil and, if planted with the seed, they can readily attack the growing roots.

Hygiene

Cysts are moved from field to field by wind, flood water, farm machinery, implements and footwear, as well as on soil adhering to plants, other than potato, for transplanting. Cleaning machinery and constructing natural windbreaks inhibits new infestations.

All waste, soil and byproducts from potato grading must be disposed of in line with the Code of Good Agricultural Practice. Under the PCN Control Directive, potato processors must have officially approved waste disposal procedures for soil residues before they can be used on agricultural land as a fertiliser or filling material. At the very least, growers grading on farm should remove the waste soil to land outside of a potato

Table 2. Summary of seed potato rules related to PCN control

Topic	Scotland	England and Wales
Website	www.sasa.gov.uk/pcn-soil-testing-documents	www.gov.uk/government/publications/potato-cyst-nematode-application-for-soil-sampling
Sampling	Standard rate 1,500ml/ha. Reduced rate 400ml/ha. Where land has been recorded as infested, all de-recording tests are at the standard rate. Other testing is at the reduced rate if: <ul style="list-style-type: none"> - No potatoes have been grown for six years prior to planting - No PCN found in the previous two tests, or - No PCN or dead cysts found in a visual PCN test Further reductions are made in quantity of soil analysed per ha when land is tested in larger areas (see links above for details).	
Inspection	If PCN is found, the growing of seed potatoes for sale is prohibited. The use of potatoes as farm-saved seed is also prohibited.	If PCN is found, the growing of seed potatoes for sale is prohibited.
Restrictions on ware	Ware may be grown on land recorded as infested if a control programme is approved and set down in a statutory notice before planting. There are sanctions for landlord and/or tenant if such land is planted in potatoes without such a programme.	Ware may be grown on land where PCN has been detected if the potato varieties to be grown are resistant to PCN, or where APHA authorises the growing of a crop to be harvested before the cysts mature, or where a soil treatment approved by APHA for control of PCN is used.
Restriction on home saved seed	The direct progeny from 1 year's worth of classified seed potatoes may be planted as farm-saved seed (FSS), but require PCN testing of the land where the FSS is produced, unless it comes from the same holding (tightly defined).	Tubers from land recorded as PCN-infested may not be used for seed.

rotation. Potatoes harvested from heavily infested land may carry cysts in the eyes even after washing.

Study of soil borne diseases shows the value of hygienic control of every item that could move the organism into a clean field. Such meticulous hygiene has not been the practice with potatoes. One reason is the long time period between introducing cysts and seeing crop damage, in contrast to a disease such as club root which shows its effects more quickly.

In Germany, where the Biowaste Ordinance prescribes sanitation of organic waste before it can be used on arable land, composting for 7 days where all the material reaches 50°C has been shown to kill cysts of *G. rostochiensis*.

Control of volunteers

Volunteer or ground-keeper potato plants at any density can maintain or increase PCN numbers where the rest of the field has low infestation. Potatoes are difficult to control in some vegetable crops because the only effective herbicides are often toxic to the crop. Between-row cultivations and shielded broad spectrum herbicide applications are sometimes used to control volunteers.



Figure 18. Volunteer plants emerging with a planted potato crop

Varietal resistance to PCN

Resistance refers to the ability of a variety to limit the increase in population of PCN and, in some instances, to reduce numbers of this pest. It is measured by the ratio of Pi (initial egg numbers at planting) to Pf (final egg numbers after harvest).

The most effective control measure for suppressing PCN multiplication is the use of highly resistant varieties, shown in Table 3. When many varieties with high levels of resistance to *G. rostochiensis*, such as Maris Piper, became available and widely planted, the incidence of and damage caused by this species greatly reduced.

The term 'pathotype' is used to classify variant populations of PCN according to their ability to overcome resistance in the host plant. Resistance to *G. rostochiensis* pathotypes Ro1 and Ro4 in current varieties is controlled by a single major gene. The nematodes introduced to Europe are limited in their genetic diversity and this gene has continued to give stable high-level resistance over several decades. However, if a wider range of *G. rostochiensis* pathotypes were ever to be introduced, the damage to the potato industry caused by this species could increase markedly.

The introduction of genetic resistance to *G. pallida* has been slower and more difficult, partly because of the greater genetic diversity of this pest. Although most UK populations fall into the pathotypes Pa2 and Pa3, there is no clear boundary to differentiate populations. A further pathotype of *G. pallida*, Pa1, is found in Scotland and Northern Ireland and a recent report suggests this pathotype may be more widespread than originally thought. Resistance to *G. pallida* requires a combination of genes from several chromosomes, because no major gene conferring resistance to this species in the potato host is available.

Two sources of *G. pallida* resistance are the wild *Solanum* species, *S. vernei* and *S. andigena* and the most resistant varieties have genetic material from both sources. Most of the commercial varieties available in the UK that show resistance to *G. pallida* are slightly more resistant to Pa1 populations than to those classified as Pa2 or Pa3.

The data in Table 3 is from independent variety trials (IVTs), except for those marked *, which are provided by the breeder. In some cases the breeder has tested *G. pallida* resistance against the Pa2 and Pa3 pathotypes separately.

Although many varieties now have resistance to *G. pallida*, their adoption has been slow, possibly because of market demands for particular marketable characteristics. Table 4 shows that there is still scant resistance to *G. pallida* in the most popular varieties, while resistance to *G. rostochiensis* is plentiful.

There are various commercial reasons for choice of variety; nevertheless, the susceptible varieties most commonly grown exacerbate the *G. pallida* problem and are unsustainable in the long term. More effort can be put into breeding for resistance when a major market is available. This may partly explain why processing, where a few large customers determine the varieties grown, is better supplied with resistant varieties than the fresh market.

Table 3. Varieties with resistance to PCN, Great Britain 2016

Potato variety	Area grown (to nearest 50ha)	<i>G. pallida</i> Pa2/3, 1 (A)	<i>G. rostochiensis</i> Ro1 (A)	Market use
Taurus	2,800	3	8	Crisps
Innovator	2,450	Pa2 8*, Pa3 9*	Not resistant	Fries/chips
Royal	2,400	3	9	Fries/chips
Harmony	2,050	4	4	Fresh
Arsenal	1,300	8/9*	6	Crisps
Ramos	1,200	4	8	Fries/chips
Lanorma	800	5	9	Fresh
Sapphire	700	3	4	Fresh
Eurostar	400	9*	9*	Fries/chips
Diva	<400	5	3	Fresh, fries
Panther	<400	8	2	Prepack
Leonardo	<100	Pa2 7* Pa3 3*	9	Fries
Crisps4all	<50	6	9	Crisps
Alcander	<50	Pa2 9*, Pa3 8*	8/9*	Crisps
Performer	<50	9*	5*	Chips, crisps
Heraclea	<50	Pa2 6*, Pa3 1*	–	Crisps
Camel	<50	8*	9*	Fresh
Swift	<50	4	–	Fresh

Notes: (A) Rating on a 1–9 scale; 1 = least resistant, 9 = most resistant

* Information from breeder

Table 4. Resistance status of the top 12 ware potato varieties by area in Great Britain, 2017

Potato variety	GB planted area, Ha	Resistance to <i>G. pallida</i> Pa2/3,1 (rating)	Resistance to <i>G. rostochiensis</i> Ro1 (rating)
Maris Piper	16,310	2	9
Markies	6,030	2	9
Maris Peer	5,000	2	2
Melody	4,300	2	9
Lady Rosetta	3,460	2	9
Estima	2,990	2	2
Taurus	2,770	3	8
Pentland Dell	2750	2	2
Marfona	2,400	2	2
Innovator	2470	8/9	Not resistant
Sagitta	2440	Not resistant	Resistant
Royal	2390	3	9

Source: IVT. Data is available on the AHDB Potato Variety Database

■ Resistant
 ■ Semi-resistant
 ■ Susceptible

Tolerance

An important concept related to resistance is that of tolerance. A variety is considered tolerant to PCN if the yield is not reduced in the presence of PCN compared to the yield in the absence of PCN in the same field situation.

Tolerance is sometimes measured by the difference in yield between plots treated or untreated with nematicide. This method is imperfect because nematicide treatment provides incomplete and variable levels of nematode control, depending on a variety of environmental factors.

Soil type, particularly the ability of the soil to hold water, has a major influence, so irrigation can improve the tolerance of a crop. The potential combinations of resistance and tolerance, as illustrated in Figure 19, are crucial to understanding how best to use resistance in control of PCN.

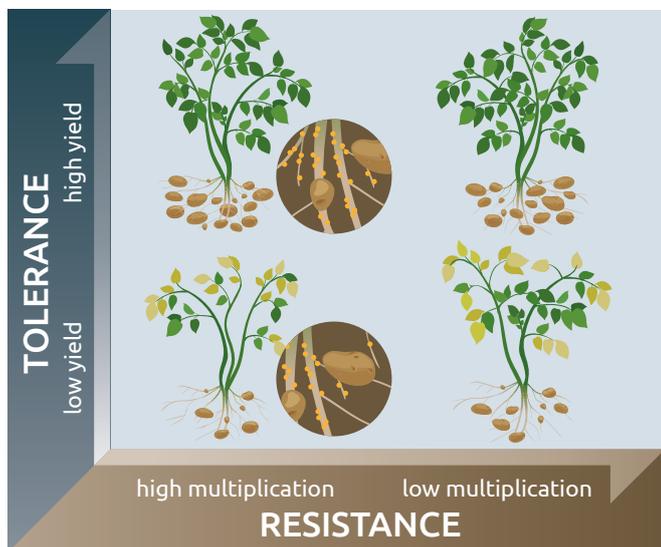


Figure 19. Consequences of varieties with different combinations of resistance and tolerance

Some resistant varieties are tolerant because they suffer less from the feeding of developing female nematodes. In general, the more vigorous and late-maturing varieties with strong top growth are the most tolerant because they are able to compensate for loss in leaf area caused by nematode damage. Variety databases do not provide assessment of tolerance or vigour, so information must be sought anecdotally from breeders and other growers, or from the limited number of trials that have taken place (Table 5).

An experiment by Vegetable Consultancy Services in 2016 compared the yields of some resistant and non-resistant potato varieties, with and without fosthiasate nematicide. The difference between the yields indicated tolerance, since a tolerant variety maintains its yield when challenged by PCN. The site was on sandy soil with an average Pi of 11 eggs/g and there were four replicates.

Table 5. Field tolerance testing at Elveden Estates

Variety	% Yield decrease without nematicide	Tolerance rating (subjective)
Panther	41	Intolerant
Innovator	22	Intolerant
Maris Peer	13	Moderately intolerant
Maris Piper	13	Moderately intolerant
Eurostar	13	Moderately intolerant
Arsenal	13	Moderately intolerant
Forza	6	Moderately tolerant
Lanorma	6	Moderately tolerant
Cara	3	Tolerant
Performer	-5	Tolerant
Royal	-6.5	Tolerant

It is tempting to plant a tolerant variety in a field known to have PCN, since yields are likely to be higher than when an intolerant variety is planted. This can be true in the short term. A susceptible, tolerant variety, however, is the least sustainable in the longer term because it permits the greatest multiplication of PCN.



Figure 20. Ariel view of the field tolerance trial at Elveden Estate

Trap cropping

Trap cropping involves growing a host plant that produces a high level of hatching factors, but which is either an unsuitable host for nematodes, or is destroyed before nematode reproduction is completed. In the first case, the PCN juveniles ultimately die, so are unable to multiply, reducing the PCN population in the field.

Alternatively, a potato crop is harvested before the nematodes multiply, but this method is risky. Research into the life cycles of PCN in the field suggests this method may be hazardous because of the unpredictability of the weather (either unusually warm, or too wet to

harvest) and other environmental factors in determining the correct time to destroy the crop. Some members of the genus *Solanum*, other than potatoes, are better suited as trap crops because they stimulate cysts to hatch but do not permit PCN multiplication.

Trap cropping on commercial farms currently uses *Solanum sisymbriifolium*, sticky nightshade, marketed by Greenvale AP as DeCyst and by Branston Ltd as Foil-Sis. To stimulate nematode hatching, trap crops must develop a substantial root system and accumulate at least 700g/m² dry matter. A model using field data from 2002 showed this requirement was most likely to be achieved in the south rather than the north of the UK.

Sticky nightshade is most effective when sown in May to June, with soil temperatures not dropping below 12°C and consistent moisture for germination. A fine seedbed is required, with seed covered by 0.5–1cm soil, rolled to retain moisture. The seed rate is 3kg/ha when using a precision drill, but sowing with a cereal drill is also practiced, after bulking the seed by mixture with lentils. Irrigation is sometimes necessary to achieve a complete stand, especially in sandy or peaty soils. Weed control is essential in the early stages of crop growth and details of suitable herbicides can be obtained from the seed suppliers.

Cutting back the crop to 45cm in September can stimulate new growth and increase efficacy. At the end of the season, the trap crop may be chopped and ploughed in at any time. Reduction in egg numbers of up to 80% have been reported from a well-grown crop of sticky nightshade.

AHDB research indicates that some other *Solanum* species have potential for use as PCN trap crops, as shown in Table 6.

The economics of trap cropping can improve as part of an integrated management strategy, when a field is heavily infested with both blackgrass and PCN, at a time

of low cereal prices. During spring, the field can be cultivated a few times to obtain multiple flushes of blackgrass, destroyed prior to sowing a PCN trap crop in May or June.

Biofumigation

This term refers primarily to the suppression of soil-borne pests, pathogens and weeds by isothiocyanates. These are gases released from Brassicas and related plants when thorough mechanical damage to tissues (maceration) leads to widespread rupturing of cell walls. Maceration ensures that glucosinolates are liberated from the cell vacuoles, allowing them to be broken down by the action of the enzyme myrosinase, which is released from separate myrosin cells. Of the products released, isothiocyanates have the greatest activity against PCN. Typically the effect is achieved by growing the appropriate crop for 10–14 weeks in late summer and early autumn, followed by maceration, incorporation into the soil and ground-sealing.

Biofumigation has been trialled both by researchers and farmers, but few growers are using it on a regular basis. It is more complex and demanding than the use of nematicides and the results are highly variable. Reductions of 35–70% in viable PCN population densities have been found in field experiments in England.

Currently, 132 glucosinolates have been identified, with those most active against PCN being sinigrin, gluconasturtiin and glucotropaeolin.

Three factors interact to determine the efficacy of biofumigation:

- Concentration of active glucosinolates
- High biomass of plants with high glucosinolate content
- Method of maceration, incorporation and sealing

Table 6. Results of a 3-year experiment on potential trap crops – (R468 final report)

Species	Preferred sowing date	Ease of establishment	Pre-emergence herbicide	Control of PCN eggs (%)
Black nightshade <i>Solanum nigrum</i>	Late May	Medium	Tolerant to clomazone and metribuzin. Sensitive to pendimethalin, flufenacet and prosulfocarb	45–76
Garden huckleberry <i>Solanum melanocerasum</i>	May–June	Medium, vigorous once established	Tolerant to clomazone and low rates of pendimethalin, flufenacet and prosulfocarb. Very sensitive to metribuzin	45–65
Sticky nightshade <i>Solanum sisymbriifolium</i>	May–June, through August; sowing may succeed	Relatively difficult	Tolerant to clomazone and pendimethalin (from evidence outside of this trial)	26–56

Note: This experiment was conducted in large containers rather than in the field. Results were influenced by year, soil type and origin of seeds.

Biochemical analysis in pot trials has shown the species and varieties with the most genetic potential for producing the most active glucosinolates. Many such varieties are commercially available, with examples shown in Table 7. Variety selection depends both on genetic potential and whether its characteristics fit the farming system. Agronomic conditions should maximise biomass of the species selected.

Maximising biomass depends on time of planting, agronomy and soils. About 100kg of nitrogen and 40kg of sulphur is usually required to achieve the best biomass and glucosinolate content. Seed suppliers can give advice on seed and fertiliser rates, including sulphur which can vary from 25–60kg/ha depending on species and variety.

Biofumigants produce more biomass when the photoperiod is long and temperatures are in the range of 15–25°C. Similarly, Brassica species grown in longer days with temperatures of 15–25°C, high light intensity and dry conditions have the highest total glucosinolate content. When sown following crops such as peas, winter barley or oilseed rape, such conditions can usually be achieved in England and Wales, though the window of opportunity is shorter in Scotland and the risk of crop failure or reduced biofumigant activity is greater.

Even when a large production of active material has been achieved, the benefits will only be realised if the isothiocyanate gases are released and trapped in the soil for sufficient time. A flail cutter leaves much plant material undamaged, so machinery that not only flails but also crushes the material will release isothiocyanate gases more quickly. This sudden flush of gas is more effective than a period of slow release during which some gas is lost to the atmosphere.

Fumigant gases are rapidly released after flailing and crushing the crop, so incorporation by spading or

rotavating should take place as soon as possible. Ensure that the three machines used for flailing, incorporation and rolling operate tightly together for maximum efficacy. The seal is better in moist soil.

Three factors determine the best time to incorporate: crop maturity, soil temperature and soil moisture. If the soil is dry then irrigation could greatly improve the soil seal and thus the efficacy of the biofumigation. As an enzymatically mediated reaction, the production of isothiocyanates is faster in warm temperatures and is favoured by incorporation when soils are at 12–13°C or warmer, typically in mid-October. If soils are still warm and the crop is large and leafy with some flowers appearing, it is desirable to incorporate.

Brassica species host club root (*Plasmodiophora brassicae*) and cabbage root fly (*Delia radicum*), so carry a high risk to a rotation with oilseed rape or vegetable Brassicas. Some oilseed radishes are not club root hosts and so are safer rotational crops.

Partial biofumigation

Biofumigants release glucosinolates in root exudates. Myrosinase-producing microorganisms break down the glucosinolates to access glucose, which leads to release of isothiocyanates. Such exudates from brown mustard, rocket and oilseed radish have been shown to reduce numbers of PCN in field experiments; a process known as partial biofumigation.

Growers sometimes plant overwintered cover crops before potatoes, for benefits including improvements to soil structure and reduction in erosion. They hope the use of crops with high glucosinolate content such as oilseed radish will have the additional benefit of killing PCN. Oilseed radish has restricted potential as a conventional biofumigant because its glucosinolates are stored mainly in the swollen roots, which are difficult to

Table 7. Species that produce high quantities of glucosinolates

Species	Common name	Examples of commercial varieties	Winter hardy	Applicable for PCN management	Content and mode of action
<i>Brassica juncea</i>	Indian mustard Caliente mustard Brown/yellow mustard	Caliente 99 or 199 Scala Vitasso Spudguard	No	Yes	Rich in sinigrin
<i>Raphanus sativus</i>	Oil/oilseed radish	Bento Doublet	Yes	Yes	Suitable for partial biofumigation from root exudates Contains glucoraphanin
<i>Eruca sativa</i>	Rocket Arugula	Nemat Trio	Yes	Limited data	—
<i>Brassica carinata</i>	Ethiopian mustard Abyssinian mustard	Carbon Cappuccino	Yes	Limited data	—
<i>Sinapis alba</i>	White mustard	Smash Architect Vitaro	No	Limited data, often used in combination with other species	Rich in myrosinase Low quantity of glucosinolates
<i>Brassica rapa</i>	Canola Turnip rape		Yes	Limited data	—

macerate, especially in stony soils. It may be more suitable as a partial biofumigant, where its frost hardiness is an advantage. However, partial biofumigation effects have not yet been scientifically demonstrated for any overwintered crop.

Chemical nematicides

Treatment with a nematicide should not be considered a panacea because it offers only partial control. The effect of these chemicals is first to stun the eelworms in what is called nematostatic action. Some of these will then die, while others may recover later in the season, invade roots and reproduce. In consequence, nematicides tend to protect the yield of the current crop, but often do little to reduce the number of PCN eggs left in the soil for the future. For this reason, nematicides should not be the only method of control used, at any level of infestation.

Chemical control affects the most vulnerable stage of PCN, the juveniles, migrating from the protective cyst to potato roots. During this phase, the outer cuticle of the nematode is in direct contact with soil solutions and absorbs chemicals within those solutions, which is why nematicides are applied before planting potatoes.

Nematicides formulated as granules are classified as non-fumigants because they have low volatility and do not require soil sealing. The current granular nematicides available are the carbamate Vydate 10G® (10% oxamyl) from DuPont® and the organophosphates Nemathorin® (10% fosthiazate) from Syngenta® and Mocap 15G (15% ethoprophos) from Certis. When Mocap 15G replaced Mocap 10G the active ingredient applied was reduced from 10kg/ha to 6kg/ha of ethoprophos.

Fosthiazate and ethoprophos are organophosphates. They work only by contact and are rapidly broken down in plants. Oxamyl, a carbamate, has both contact and systemic action. It can work inside the plant, leading to reduced feeding and moulting, but this gives a higher risk of chemical residues in the plant. Following the product labels, particularly regarding harvest interval, is important to prevent exceedance.

Organophosphates tend to be adsorbed onto organic matter, making such nematicides less effective in organic soils. On organic soils, the carbamate oxamyl is more likely to be effective.

Carbamate nematicides such as oxamyl inhibit the enzyme acetylcholinesterase in nerve tissue. At field concentrations, nematodes may be paralysed, which is described as a nematostatic effect. When the concentration declines, these juvenile nematodes may regain activity. The persistence of oxamyl increases with acidity, from 10–14 days at pH 7.0 to 16 weeks at pH 4.8. While this can make oxamyl more effective in acid soils, it may also lead to a higher risk of residues in produce, requiring extra residue tests at harvest.

The persistence of a chemical nematicide is described in terms of its half-life. The half-life of fosthiazate has been measured as varying from 2–6 weeks. When emergence and growth of the crop is delayed by cold weather after planting, the nematicide will be less

concentrated by the time PCN juveniles travel to potato roots. Heavy rain or irrigation can wash nematicides away before they can exert their effect.

The half-life of oxamyl is extremely variable, dependant on soil pH, moisture content and temperature. One other factor appears to be whether the soils have previously been treated with oxamyl. A few isolated cases have been reported showing enhanced degradation from bacteria in the soil, when soils have been tested in the laboratory. However, scientific evidence is lacking for such microbial degradation across a variety of crops in rotation. Enhanced degradation does not appear to affect fosthiazate.

The collected evidence suggests that the use of nematicides, while not guaranteeing yield equivalent to that in a PCN-free field, can significantly reduce yield loss compared to untreated, particularly when used in conjunction with other tools in an IPM approach to PCN management. Their effect on multiplication, however, is less reliable. Research shows reduction in some cases and not in others. Multiplication is best suppressed when the initial PCN population (P_i measured in eggs/g soil) is low. The result depends on the efficiency of incorporation and individual nematicide properties, the hatching period of the PCN species and the characteristics of the varieties grown; also on soil temperature from planting, moisture and soil physical and chemical characteristics.

With so many factors involved the result in a particular case is uncertain so growers are advised to monitor the P_i and P_f of the populations in their own fields to assess the effectiveness of chemical control as a management tool. Certainly, if chemicals are used, they should be applied as soon as PCN is detected in a field.

As nematicides have short persistence, they give poorer control of *G. pallida* than *G. rostochiensis* because of the different hatch patterns of these two species. The peak of egg hatch of *G. pallida* is 6–7 weeks after planting, with an extended hatch of up to 12 weeks, compared to a peak at 3–4 weeks with *G. rostochiensis* and a hatch period of about 6 weeks.

Correct incorporation of nematicides has a major influence on efficacy because, at the approved rate, a little chemical has to treat a considerable soil volume, as illustrated in Figure 21. For example, with oxamyl, where the rate of Vydate 10G is 55kg/ha, if incorporated to 15cm, each 5g treats 150L of soil. If incorporated to 30cm, each 5g must treat 300L of soil.

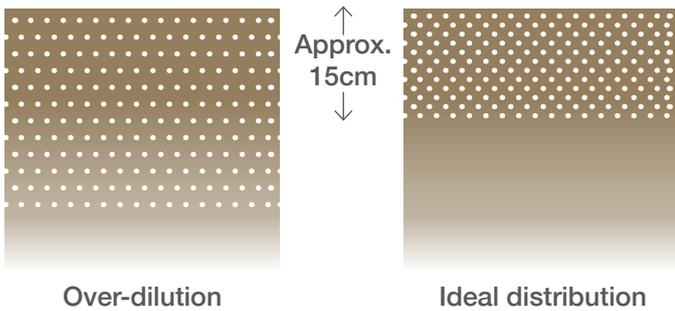


Figure 21. Representation of over-dilution of the chemical by incorporating too deeply

Nematicides are sometimes incorporated during bed tilling. Bed tillers are often set to work more deeply than is recommended for incorporation, diluting the chemical and reducing efficacy. Nematicides are sometimes incorporated with de-stoners. This has important drawbacks and is not recommended because of excessively deep incorporation, loss of some chemical into the wheelings and banding of granules, which is most common with star separators.

When used for control of free-living nematodes such as *Pratylenchus* or *Trichodorus*, oxamyl is usually applied in-furrow by an attachment on the planter. The grower may hope that the same application will also control any PCN present. PCN juveniles can move 20cm through the soil from a protective cyst to a potato root, so application in-furrow is unlikely to give good control.

The best incorporation, with minimal waste, is achieved by applying nematicide to the front of the planter.

Incorporate with a shallow rotavator on the front linkage, working at a low rotor speed, to a depth of about 15cm, ensuring granules are in the zone around the seed tubers.

Selective treatment of hot spots of infestation, based on GPS mapping, does not appear to have improved control. In fact, it is a false economy, because many foci of infestation are unlikely to be detected unless an intensive programme of sampling is undertaken.

Chemical fumigation

In the past, chemical fumigants were used that killed nematodes while still inside the cyst. Only the granular fumigant Basamid (Certis) is still approved. Basamid contains 97% w/w dazomet, which releases methylisothiocyanate (MITC). The cost of this product means it is normally reserved for higher value crops.

Efficacy of Basamid fumigation depends on correct soil moisture and temperature, typically achieved in September. A good seal is essential, using a smear roller with close attention to the joints.

Selective treatment of infestation hot spots using GPS mapping is a contentious approach. It can easily fail because hot spots are either inaccurately identified or missed completely. With thorough testing and mapping it is possible that Basamid could hit the hot spots, followed by use of granular nematicide and/or resistant varieties on the whole field. Care is required to prevent recontamination of the rest of the field with PCN from hot spot soil carried on tractor tyres.



Figure 22. Nematicide applicator and rotavator on the front of a tractor pulling a planter

Nematicide Stewardship Programme

The Nematicide Stewardship Programme (NSP), initiated in 2015, identifies and promotes best practice. The full programme, which is evolving, and a list of BASIS-approved advisers specialising in PCN management and nematicide stewardship, is available at nspstewardship.co.uk

NSP requirements

- Operators must be qualified to apply nematicides (NPTC PA4 or PA4G certification)
- Those applying nematicides must have completed the Industry Stewardship Training Module
- Applicators must be fitted with a device in cab that allows the operator to shut off nematicide granule flow at least 3 metres from the end of each row

Biological control

In the UK and elsewhere, trials have taken place with parasites and predators of PCN, but none of these are available commercially.

In laboratory tests, arbuscular mycorrhizal fungi have been shown to inhibit root invasion by PCN. A fungal parasite of nematode eggs, *Pochonia chlamydosporia*, has also been investigated. It has performed well in some trials, but has not been multiplied to commercial quantities and could be vulnerable to fungicides used in the field. Other fungi that may have potential as predators or competitors to PCN are *Trichoderma harzianum*, *Plectosphaerella cucumerina* and *Penicillium oxalicum*.

Another biological approach is to use hatching factors or hatching inhibitors. A variety of composted waste products have also been found to reduce eggs, juveniles and cysts, with the suppressive effect on nematodes increasing in relation to ammonia content and compost rate. Specific botanical products with nematicidal properties include garlic extracts, sweet wormwood (*Artemisia annual*), jasmonate and neem; but none have yet been shown to be effective in the UK.

Integrated control

All of the control methods described above can be used within a rotation. On a single crop it is often appropriate to use a resistant variety, nematicide and cultural control methods. No resistant variety is fully tolerant, so nematicide can be used to minimise root damage during establishment, protecting its yield while the plant resistance acts to reduce PCN multiplication.

The best practice for stabilising or reducing PCN soil populations is an IPM strategy employing longer rotations, control of volunteers and use of resistant varieties where possible, with nematicides used only as part of such a strategy.

Key facts

- Potato is the only field grown crop host for PCN in the UK
- In the absence of a host, PCN eggs hatch over an extended period, so the numbers left viable decline each year
- When potatoes are grown, specific chemicals exuded by the roots of both susceptible and resistant cultivars stimulate most (up to 90%) of the eggs to hatch
- Juvenile nematodes are attracted to potato root tips, invade them and feed within them. This causes much of the damage to the crop by stunting root growth and reducing uptake of nutrients and water from the soil. This feeding can cause considerable yield losses without necessarily producing large numbers of cysts, especially in resistant, intolerant varieties
- Juvenile nematodes develop within the root. On susceptible cultivars they develop and small (0.5mm), round females eventually protrude through the surface of the root to form a cyst. In July and August they are visible to the naked eye on the outside of the root
- Tolerant varieties can withstand or recover from damage and produce a yield
- Resistant varieties can prevent or restrict nematode reproduction, although yield may be severely impaired

Recommendations

Soil testing

- Examine field history to decide if infestation is expected. If it is not expected or is unknown, follow the procedures given in this document for detection of early-stage infestations. In such cases, analyse the whole soil sample to give the best chance of detecting small pockets of PCN
- When an infestation is found, determine the species
- In established infestations use Figure 13 to determine the quantity of soil to analyse in the lab. Samples of 200g and in short rotations, samples of 400g, are recommended to give accurate assessments of infestation
- To best detect low levels of infestation, or to identify the efficacy of controls used, test selected fields after harvest

Seed

- Buy certified seed rather than using farm-saved seed. In Scotland, land for growing farm-saved seed must be tested and be free from PCN
- If planning to grow seed for use in the same business, test soil to the same standard as if growing seed for sale

Hygiene

- Clean and wash machinery before and after use where PCN transfer is a risk
- Dispose of waste grading soil to minimise risk of PCN transfer

Volunteers

- Take vigorous measures to control volunteer potatoes quickly in the rest of rotation. Such volunteers reduce the efficacy of other forms of PCN control

Varietal resistance

- To improve sustainability, the industry should move away from non-resistant and tolerant varieties to resistant varieties, where possible
- Determine the PCN species and numbers present prior to making the decision to grow a particular variety
- Grow susceptible varieties only on land that has been tested and found to be free of PCN

Nematicides

- Use nematicides only in conjunction with other methods for reducing PCN
- Incorporate nematicide granules to the depth of seed planting
- Use product stewardship guides produced by the chemical company suppliers for advice on application, harvest interval for products and sampling protocols for tuber residue testing

Integrated Control

- Use a variety of integrated control measures to reduce the PCN population at the time of planting. These include hygiene, clean seed, control of volunteers, trap crops, biofumigants and nematicides. In general, the resistance of the particular variety being grown will have the greatest effect on the PCN population following harvest



Figure 23. PCN cyst

Appendix

A modelling approach was used to determine the most important factors influencing the precision of PCN population estimates. The fact that eggs are packaged into cysts is relevant and has a consequence for subsampling in the laboratory. At higher numbers of eggs/cyst (e.g. 400 eggs/cyst) the greater the margin of error associated with the population estimate.

For example, if the average PCN population in a field is 5 eggs/g of soil and there are high numbers (400) of eggs in each cyst (as would occur soon after growing a susceptible potato variety), then processing a small subsample (eg 100g) of the soil collected from the field would result in a population estimate with a margin of error ranging between 0 and 13.8 eggs/g soil (ie, see bottom left corner of Table A below).

Table 8. Confidence limits (CL) for identification of PCN

Sub-sample	100g		200g		400g		1,000g	
	Lower CL	Upper CL						
A Target population = 5 eggs/g								
10	2.9	7.1	3.1	6.9	3.3	6.8	3.4	6.7
50	1.5	8.5	2.3	7.7	2.8	7.2	3.2	6.8
100	0.4	9.6	1.6	8.4	2.4	7.6	3.0	7.0
150	0.0	10.6	1.0	9.0	2.0	8.0	2.8	7.2
200	0.0	11.4	0.4	9.6	1.7	8.4	2.7	7.3
400	0.0	13.8	0.0	11.3	0.6	9.5	2.1	7.9
B Target population = 10 eggs/g								
10	6.7	13.4	7.0	13.0	7.1	12.9	7.2	12.8
50	4.9	15.1	5.9	14.1	6.6	13.4	7.0	13.0
100	3.3	16.7	4.9	15.1	6.0	14.0	6.8	13.2
150	2.0	18.0	4.1	15.9	5.5	14.5	6.6	13.4
200	0.9	19.1	3.3	16.7	5.0	15.0	6.4	13.6
400	0.0	22.6	1.0	19.0	3.5	16.5	5.6	14.4
C Target population = 20 eggs/g								
10	14.7	25.3	15.1	24.9	15.3	24.7	15.4	24.6
50	12.4	27.6	13.8	26.2	14.6	25.4	15.2	24.8
100	10.3	29.7	12.4	27.6	13.8	26.2	14.9	25.1
150	8.5	31.5	11.4	28.6	13.2	26.8	14.6	25.4
200	7.0	33.0	10.3	29.7	12.6	27.5	14.3	25.7
400	2.15	37.85	7.05	33.0	10.5	29.5	13.4	26.6

Lower CL = lower confidence limit; Upper CL = upper confidence limit. These terms are used interchangeably with the term margin of error.

NOTE: A colour coding system has been used in the tables:

- If the difference between the upper and lower confidence limits is less than the target population
- If the difference is greater than the target population by a factor of less than 1.5
- If the difference is greater than 1.5 times the target population.

If subsampling is carried out in the lab, the most appropriate subsample size is that which results in a green coded value. However, given that the number of eggs/cyst and the "target" (ie, the PCN population estimate) are not known at the outset, it is difficult to select an appropriate laboratory subsampling strategy in advance.

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