INDUSTRY BIOSECURITY PLAN

FOR THE GRAINS INDUSTRY

Threat Specific Contingency Plan

Cereal cyst nematodes

Specific examples detailed in this plan:

Heterodera latipons

H. filipjevi

***H. avenae* (exotic strains)**

Prepared by Plant Health Australia

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Plant Health AUSTRALIA

GRDC Grains Research & Development Corporation

CRC PLANT biosecurity

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1 Purpose and background of this contingency plan

This contingency plan provides background information on the pest biology and available control measures to assist with preparedness for an incursion into Australia of exotic Cereal cyst nematode threats to the Grains Industry. This contingency plan focuses on the three nematode species that are considered to be of greatest economic impact and risk to the Grains Industry, namely *Heterodera latipons*, *H. filipjevi* and exotic strains of *H. avenae*.

It provides guidelines and options for steps to be undertaken and considered when developing a Response Plan for incursion of cyst nematodes that affect cereals. The control and management will specifically be for *Heterodera latipons*, *H. filipjevi* and *H. avenae*; however, the general principles may be applicable to incursions of other Cereal cyst nematodes. Any Response Plan developed using information in whole or in part from this Contingency Plan must follow procedures as set out in PLANTPLAN and be endorsed by the National Management Group prior to implementation.

The information for this plan has been primarily obtained from documents as cited in the reference section. For each of the three high priority Cereal cyst nematode threats, information on background, life cycle, host range, distribution and symptoms is given, with the emphasis of this document on the management options in the event of an incursion in Australia.

2 Australian grains industry

The Australian Grains Industry is primarily situated in a narrow crescent running through the mainland states, known as the grain belt. This area stretches in a curve from central Queensland, through New South Wales, Victoria and southern South Australia. In Western Australia, the grain belt covers the south-west corner of the state.

The grains industry is the largest plant industry in Australia and grain crops are grown in all states and territories. The gross value of grains and oilseeds in 2006/07 was $5.3 billion, down on the 2002/03-2006/07 average of $7.4 billion per annum largely as a result of the severe drought conditions experienced by eastern Australia (ABS data). The grains industry consists of 25 leviable crops; however, Cereal cyst nematode is predominantly a threat to wheat, barley and oat crops. Of these crops, wheat is the most important in terms of area cropped and economic value, with an average of nearly 19 million tonnes per year grown over 12.4 million hectares (ABS data for five year average to 2008). The average annual production of barley is approximately 7.1 million tonnes and the average annual area sown to barley is 4.3 million hectares (ABS data for five year average to 2008). The average area sown to oats is approximately 1.0 million ha, producing around 1.3 million tonnes of grain per year (ABS data for five year average to 2008), although a significant proportion of oats grown are used for forage or hay production.

Due to Australia's relatively small population and domestic demand, export markets are essential for the viability of Australian grain farms. Australia currently exports around 60% of its grain with wheat and barley accounting for 62% and 19%, respectively, of total grain exports. With this reliance on exports, maintaining our current plant health status through appropriate biosecurity measures is of utmost importance in retaining access to these markets.

3 Eradication or containment decision matrix

The decision to eradicate should be based on the potential economic impact of host damage resulting from Cereal cyst nematode, the cost of eradication and on technical feasibility. Eradication costs must factor in long term surveys to prove the success of the eradication program. Given the ability of cysts to remain viable for long periods, a minimum of 5 years with no detections of Cereal cyst nematode will be necessary before pest free status can be declared, however for each incursion response, specific advice may be required from a Scientific Advisory Panel to confirm this time period.

No specific eradication matrix has been determined for Cereal cyst nematode, however the general decision process as outlined in

Figure 1 should be followed in determining if an incursion of this pest will be eradicated or managed/contained. The final decision on eradication or management will be made by the National Management Group.

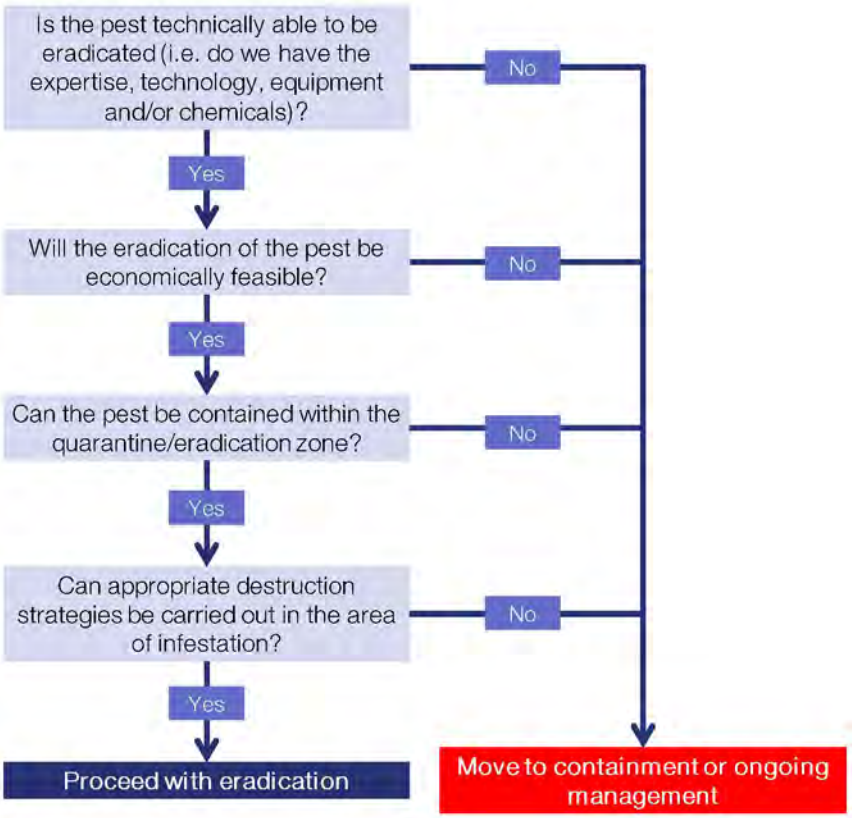

Figure 1. *Decision outline for the response to an exotic pest incursion*

4 Pest information/status

4.1 Pest details

Taxonomic position: Kingdom: Metazoa; Phylum: Nematoda; Family: Heteroderidae

4.1.1 Background

Ten nematode species are currently believed to make up the Cereal Cyst Nematode complex: *Heterodera avenae*, *H. latipons*, *H. filipjevi*, *H. cordecalis*, *H. zeae*, *H. mani*, *H. bifenestra*, *H. pakistanensis*, *H. arenaria* and *H. pratensis*. The most economically important are *H. avenae*, *H. latipons* and *H. filipjevi* (Nicol *et al*., 2003). *H. filipjevi* was previously referred to as the Gotland strain of *H. avenae* (Ferris *et al.* 1999).

H. avenae is the most damaging species on temperate cereals with yield losses of up to 80%. In Australia, a single race of *H. avenae* (Ha13) has been recorded which is widespread in South Australia, Victoria, southern New South Wales and in regions of Western Australia. Considerable effort has been made in screening and breeding for both resistance and tolerance to manage this species.

In 2002, the Australian (Ha13) pathotype was proposed to be a separate species called *H. australis*, based on small pathogenic differences in cereals and differences in the ITS region of rDNA (Subbotin *et al*. 2002). No morphological differences have been recorded between European and Asian populations of *H. avenae* and *H. australis* and there has been no widespread acceptance of *H. australis* as a separate species (Smiley and Nicol, 2009).

Besides Ha13, several other races (defined by host plant resistance) of *H. avena*e have been recorded throughout the world (Rivoal and Cook, 1993).

Of the other economically important species of Cereal cyst nematode, both *H. latipons* and *H. filipjevi* are pathogens of wheat and barley and have a wide geographic distribution. Far less is known about yield loss caused by these species however, and few countries have active screening programs for resistance. *H. zeae* is a major pathogen of maize and is restricted to warmer tropical or sub-tropical regions.

In this contingency plan, specific pest information and status will be given for *H. latipons* (Mediterranean cereal cyst nematode), *H. avenae* (exotic strains) and *H. filipjevi*.

4.1.2 Life cycle

In temperate climates, the generic life cycle of cereal cyst nematodes (

Figure 8) contains four main stages:

- Juveniles emerge from the cysts in lower soil temperatures following rain (i.e. at plant emergence)
- Female nematodes then invade roots and establish a fixed feeding site (syncytium)
- Females begin to swell and rupture the root cortex as eggs are produced within the females body
- Female nematodes die and the egg filled body of the dead female is referred to as the cyst. Cysts change from white to mid/dark-brown after female death.

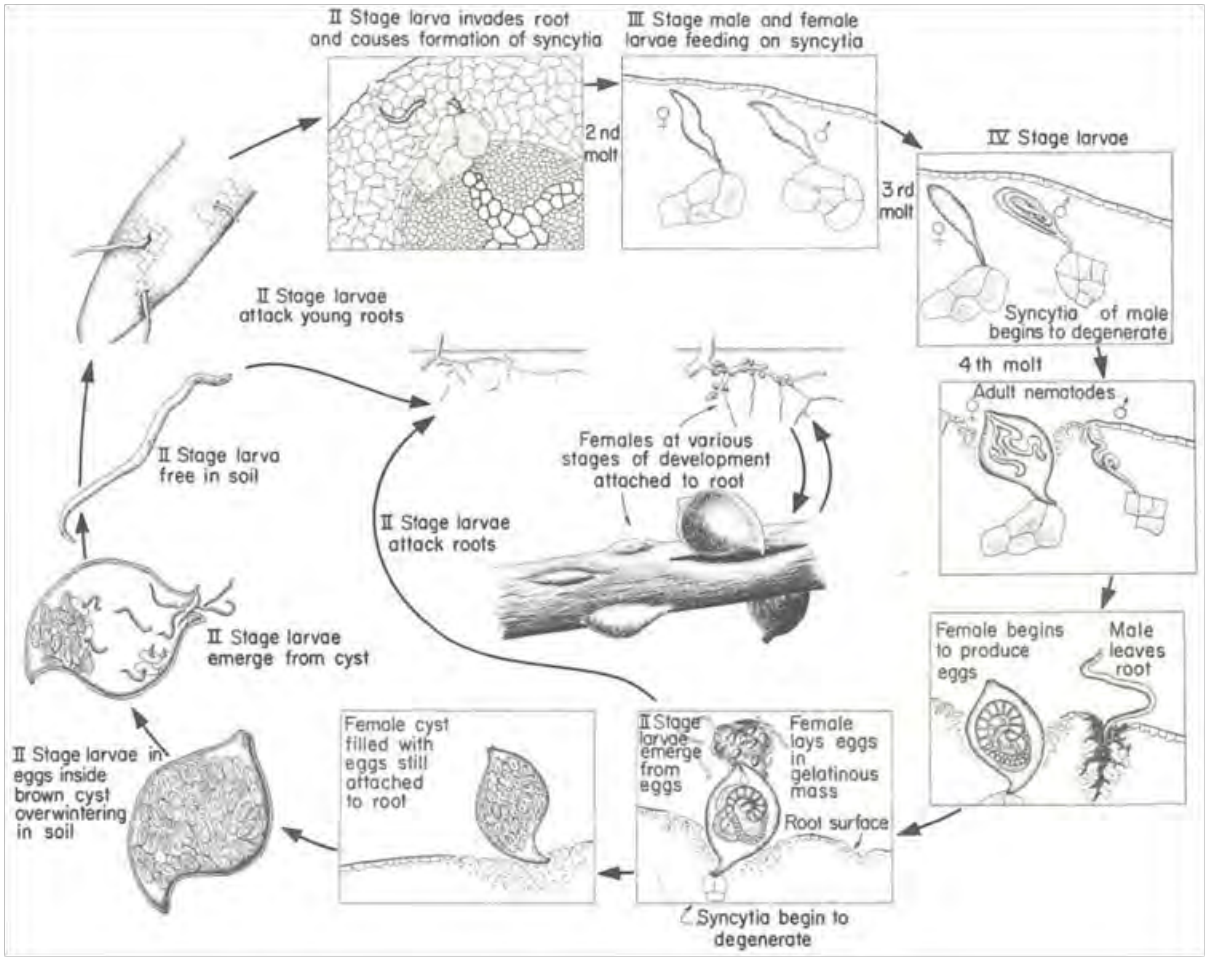

Figure 2. *Life cycle of cyst nematodes (reproduced from Agrios, 1978).*

For *H. avenae,* brown cysts are the survival stage over hot, dry summers. In Australia, cysts of *H. avenae* contain approximately 350 eggs/cyst (Meagher, 1968). Cysts undergo a facultative quiescence period and pre-treatment of older cysts at cooler temperatures (i.e. winter soil temperatures) coupled with moisture, improves egg hatch. In southern Australia, egg hatch occurs after opening rains in April - June, and when soil temperatures are approximately 10-15°C. Around 70-80% of *H. avenae* cysts hatch in the first year while remaining cysts can persist in soil over a number of seasons (Wherrett and Vanstone, 2010). Root exudates/leachates do not appear to be required for hatching.

The biology of *H. latipons* and *H. filipjevi* appear to be similar to *H. avenae*. Only one generation is completed per season and cysts are the survival stage for these species. It is believed that *H. filipjevi* cysts can survive in a desiccated state for several years in the absence of a host.

Cysts of *H. latipons* also appear to undergo a facultative quiescence period and pre-treatment of older cysts at 2-10°C, i.e. winter soil temperatures, has been shown to improve egg hatch from cysts. Late autumn breaks are therefore likely to delay hatching. Little information is available on whether *H. filipjevi* undergoes a period of quiescence once cysts are fully developed or whether improved hatching occurs following cold treatment or exposure to root leachates.

4.1.3 Affected hosts

Cereal cyst nematodes have relatively narrow host ranges, essentially limited to cereals (Table 1). Considerable variation in the tolerance or resistance of cultivars within cereal species can occur (see Section 6.3.4.

Table 1. *Hosts of Cereal cyst nematodes*

	Major hosts	**Minor hosts**	**Other potential hosts**[1]
H. latipons	Oats, wheat, barley, rye		Canary grass (*Phalaris* sp.)[2], sugarbeet[3], peanut[4]
H. avenae	Oats, barley, rye, triticale, wheat, wild oats	Poaceae (grasses), maize	
H. filipjevi	Wheat, barley	Oats	

4.1.4 Current geographic distribution

H. filipjevi is found throughout Europe with the main centre considered to be the East European-orient region (Rumpenhorst *et al*., 1996). This species has been recorded from the following countries:

- Russia, Sweden, Spain, Bulgaria, Turkey, Iran, India, England, Uzbekistan, Tadzhikistan, Estonia, Poland (Nicol *et al*. 2003, Subbotin *et al*., 1999)

H. latipons is present throughout Asia (Sikora, 1988), Europe (Scholz and Sikora, 2004), Africa and North America, as follows:

- Asia: Armenia, Iran, Israel, Japan, Jordan, Syria, Tajikstan, Turkey, Turkmenistan
- Europe: Bulgaria, Cyprus, former Czechoslovakia, Greece, Italy, Poland, Spain, Ukraine, United Kingdom
- Africa: Libya, Tunisia
- North America: Canada

H. avenae is present on all inhabited continents, though differences in distribution can occur with strains.

4.1.5 Symptoms

Above ground symptoms of *H. latipons, H. avenae* and *H. filipjevi* are similar, and include yellowing, poor tillering, stunting of plants and patchy growth (Figure 3). Leaves may be thin with a reddish-yellow colouring.

[1] The exact association with these hosts is difficult to reliably establish

[2] Greco *et al*. 2002

[3] Talatschian and Achyani 1976

[4] Fourie *et al*. 2001

Root symptoms of wheat and barley plants infested with *H. avenae* include elongation of the main root, bunched tips of rootlets and a knotted appearance due to cysts. Infected oat roots appear 'ropey' and swollen (Wherrett and Vanstone, 2010). White cysts (which turn brown as the season progresses) may be visible to the naked eye. *H. avenae* is characterised by the lemon shaped cysts formed (Bridge and Starr 2007): however, this is true of both endemic and exotic strains.

Root symptoms of *H. latipons* appear to be different to that seen with *H. avenae*, with no characteristic "knotting" (Figure 4) caused by excessive production of lateral roots at the site of infection (Mor *et al*., 1992).

Figure 3. *Patchy growth in cereal rye caused by H. filipjevi. Image source: Bonsak Hammeraas, Bioforsk - Norwegian Institute for Agricultural and Environmental Research, Bugwood.org*

Figure 4. *Characteristic 'knots' on wheat roots formed by proliferation of lateral roots around the H. avenae feeding site. Image source: South Australian Research and Development Institute*

4.2 Entry, establishment and spread

4.2.1 Entry pathways for Cereal cyst nematodes

Rating: Medium

Cereal cyst nematodes would be most likely to enter in soil contaminants in containers, machinery, plant bulbs, grain etc.

4.2.2 Establishment potential

Rating: Medium

Establishment potential is moderate-high if this species is introduced, based on the establishment of endemic strains of *H. avenae*, the suitable host range across large regions of Australia and favourable environmental conditions for development, particularly in southern regions of WA, SA, and NSW and in Victoria.

4.2.3 Spread potential

Rating: Medium

Once present in Australia, cysts could potentially be dispersed in high winds, in water or especially on machinery with infested soil.

4.2.4 Economic impact

Rating: High

Endemic strains of *H. avenae* can cause yield losses up to 80% and it is likely that exotic strains would cause similar yield losses, particularly where no resistance has been bred into Australian cereal cultivars. Losses of up to 50% have been observed in barley from *H. latipons* under similar environmental conditions to that found in southern Australia (i.e. a Mediterranean climate) (Philis,

1988) and losses appear to be greater in areas where water stress occurs (Greco *et al*., 2002). Yield losses caused by *H. filipjevi* have been observed in barley in glasshouse trials (Osipova *et al*., 1997) although little information is available on the impact of this species under field conditions.

4.2.5 Environmental impact

Rating: Unknown

There may be potential for *H. filipjevi*, *H. latipons* and exotic strains of *H. avenae* to affect native grass species, however nothing is known at present about possible impact of these species on Australian native grasses.

4.2.6 Social impact

Rating: Medium

The reduction in the value of production would be expected to cause moderate social impact with significant losses to local cereal producers and processors (livestock feed, malt producers) as well as flow on effects to the broader community.

4.2.7 Overall risk

Rating: Medium

The overall risk rating was calculated by combining the entry, establishment and spread potentials and the economic impact using the risk assessment framework applied in Industry Biosecurity Plans. A complete protocol can be found on the PHA website[5].

5 Diagnostic information

There are at least 10 species of cyst nematodes that infect cereals and grasses in the cyst nematode complex. Of these, only one species, *H. avenae* has been recorded in Australia, and this species is believed to be made up of only race, Ha13. In 2002, Subbotin *et al.* (2002) proposed that the Australian (Ha13) pathotype be a separate species called *H. australis.* This separation was based on small pathogenic differences in cereals and differences in the ITS region of rDNA. No morphological differences have been recorded between European and Asian populations of *H. avenae* and *H. australis* and there has been no widespread acceptance of *H. australis* as a separate species (Smiley and Nicol, 2009).

Since the cyst structure is usually the most readily available life stage, taxonomic characters are often related to the cyst. Morphologically, cyst nematodes are identified by assessing differences in the vulval cone structure of the adult cyst. In *Heterodera*, the cuticle in the area surrounding the vulva (of the mature cyst) ruptures and is termed the fenestra. The fenestra is used as a diagnostic feature and in *H. avenae* is termed bifenestrate, meaning the holes are each more than one half circle.

In some cyst nematode species, the cuticle thickens at the end of the vagina and forms an underbridge (Figures 5 and 6).

Measurements of juvenile nematodes can also be diagnostic including the lateral lines, stylet length and hyaline (clear) part of the tail.

[5] Available from **www.planthealthaustralia.com.au/go/phau/biosecurity/general-biosecurity-information**

Samples of suspected *Heterodera* spp. would require identification by a nematode taxonomist, as species identification based on morphology requires expertise for the preparation and measurement of juveniles and female vulval cone structures. Exotic strains of *H. avenae* (i.e. those other than Ha13) could only be distinguished using host range testing or molecular tests.

This identification may need to be confirmed by taxonomists overseas and by molecular techniques. Molecular tests have been developed in Australia for *H. avenae* and sequence data is available at CSIRO Plant Industries, Canberra.

5.1 Diagnostic protocol

H. filipjevi is morphologically similar to *H. avenae*, and much confusion has occurred in identifying these species. In 1997, the Swedish "Gotland" strain of *H. avenae* was identified as being *H. filipjevi* (Ferris *et al*., 1999; Yan and Smiley 2010). *H. filipjevi* (Figure 5) has a strong underbridge under the vulval slit, while *H. avenae* (Figure 6) has no underbridge. *H. latipons* can be differentiated by the presence of a strong underbridge under the vulval slit with a schlerotised enlargement (Figure 7).

Figure 5. *Cyst shape, vulval underbridge and cone tip structure of H. filipjevi. Photos courtesy of Roger Rivoal, INRA, France*

Figure 6. *Cyst shape, vulval underbridge and cone tip structure of H. avenae. Photos courtesy of Roger Rivoal, INRA, France*

Figure 7. *Cyst shape, vulval underbridge and cone tip structure of H. latipons. Photos courtesy of Roger Rivoal, INRA, France*

6 Pest management

6.1 Response checklist

The following checklist (Table 2) provides a summary of generic requirements to be identified and implemented within a Response Plan.

Table 2. *Checklist of requirements to be identified in a Response Plan*

Checklist item	Further information
Destruction methods for plant material, soil and disposable items	Section 7.1.1, 7.1.2
Disposal issues	Section 7.1.5
Quarantine and movement controls	Section 7.3
Decontamination and hygiene	Section 7.5
Diagnostic information	Section 5
Surveillance and tracing	Section 7.6
Surveys and epidemiology	Section 6.2
Zoning	Section 7.4
Communication strategy	Section 9.3

A range of specifically designed procedures for the emergency response to a pest incursion and a general communication strategy refer to PLANTPLAN (Plant Health Australia, 2013).

6.2 Surveys and epidemiology

Information provided in Section 6.2.1 to 6.2.3 provides a framework for the development of early detection and delimiting surveys for Cereal cyst nematodes.

Personnel should avoid moving infested soil between paddocks and properties. Shoes, tools and vehicle tyres should be thoroughly washed of soil and then sanitised with a registered disinfectant. Extra precaution should be taken when working in areas known to be infested, including rubber boots that may be left on site or disinfected.

6.2.1 Technical information for planning surveys

When developing surveys for presence and/or distribution of Cereal cyst nematode, the following characteristics of the pest provide the basic biological knowledge that informs the survey strategy:

- Host species in Australia are likely to be numerous and widely dispersed.
- An endemic strain of *H. avenae* is widespread which needs to be considered when diagnosing samples.

- The risk of Cereal cyst nematode movement on machinery and equipment contaminated with soil is high.
- Significant proportions of Australia have favourable climatic conditions for Cereal cyst nematode spread and establishment.

6.2.2 Surveys for early detection of an incursion

No active surveys for Cereal cyst nematodes occur at present. Passive surveillance for the Ha13 strain of *H. avenae* occur through routine nematode testing through submission of samples to laboratories in Western Australia and Victoria and through the Root Disease Testing Service at SARDI.

Plant symptoms may not be reliable indicators in a survey situation because the symptoms of infestation by exotic nematode species are similar in appearance to damage caused by the endemic race of *H. avenae*. Thus, targeted surveillance using soil or plant sampling and molecular or morphological diagnosis of nematodes will be required for a suspected incursion of a new species of Cereal cyst nematode.

General points to consider in effective surveillance for the presence of Cereal cyst nematodes are noted in Davis and Venette (2004) and Bridge and Starr (2007). Points to consider include:

- The chances of detecting nematodes if they are present depends on many factors including soil type, crop and cultivar, time of year, vertical distribution of nematodes in the soil, number of samples collected, size of samples etc.
- Nematodes are typically distributed in non-uniform, non-random patterns in fields (i.e. they form aggregated clusters that are distributed irregularly across fields). Numerous smaller samples provide a higher chance of detection: composite samples of at least 20 cores minimise variation and optimise time and effort.
- Narrow cores of 2.5 cm width are sufficient.
- Systematic sampling such as zig-zag patterns can be used within a field to obtain sub-samples for a composite sample.
- Samples should be collected from a depth of 10-30 cm (i.e. within the root zone).
- Samples of host plant roots or surrounding soil should be collected to obtain cysts.
- Samples must be processed to separate cysts from soil and debris.[6]
- Cysts must be prepared for identification using molecular or morphological techniques.

6.2.3 Delimiting surveys in the event of an incursion

- In the event of an incursion, delimiting surveys are essential to inform the decision-making process.

[6] Note that a molecular diagnostic test is available for detecting the presence and abundance of *H. avenae (*strain Ha13) in soil (Root Disease Testing Service, SARDI)

- The size of the survey area will depend on the size of the infected area and the severity of the infection, as well as potential movement of plant (particularly root) material and soil during the period prior to detection. It is recommended delimiting surveys should comprise local surveys around the area of initial detection concentrating on areas of poor crop growth.
- A high intensity of field sampling is needed for a high degree of confidence.
- For Potato cyst nematode, note that to achieve a high degree of confidence in probability of detection (>90%) of this species, Been and Schomaker (1996) proposed sample units of 50 cores collected on a 5m x 6 m grid to form a 2 kg composite sample, repeated 6-7 times per ha.

6.2.4 Collection and treatment of Cereal cyst nematode samples

Protocols for the collection, transport and diagnosis of suspect Emergency Plant Pests (EPPs) must follow PLANTPLAN (Plant Health Australia, 2013). Details are provided in the Standard Operating Procedure (SOP) for *Collection and transport of EPPs* available as a supporting document of PLANTPLAN (Plant Health Australia, 2013) (**www.planthealthaustralia.com.au/wp-content/uploads/2013/12/SOP-Collection-and-transport-of-EPPs.pdf**). Any personnel collecting samples for assessment should notify the diagnostic laboratory prior to submitting samples to ensure expertise is available to undertake the diagnosis.

All sample containers should be clearly labelled with the name, address and contact phone number of both the sending and receiving officers. In addition containers should be clearly labelled in accordance with the requirements of PLANTPLAN (Plant Health Australia, 2013). Containers should be carefully sealed to prevent loss, contamination or tampering of samples. The Chief Plant Health Manager will select the preferred laboratory. Additional labelling includes the identification of plant species/parts affected, location of site of sample collection in the paddock (preferably with a GPS reading) as well as symptoms and an image if available.

Refer to PLANTPLAN (Plant Health Australia, 2013) for packing instructions under IATA 650.

6.2.5 Collection of specimens

6.2.5.1 SAMPLING PROCEDURES

Samples should preferably be taken from roots of living plants or soil in which host plants have been grown. If soil is sampled, cores of soil (including root material) should be taken from within the root zone (10-30 cm deep) from numerous sites across a field to form a composite sample of around 2 kg soil (Bridge and Starr 2007). Ideally, samples should contain as much of the root system as possible, which can be lost if plants are simply pulled from the field (as opposed to using soil sampling equipment).

To obtain adult, dark cysts, sampling should be done close to harvest or over the summer period.

6.2.5.2 NUMBER OF SPECIMENS TO BE COLLECTED

A composite sample of around 20 cores over a 5 ha area is recommended (Bridge and Starr 2007). Record the location, preferably as GPS co-ordinates, or alternatively, a map reference or distance and direction from a suitable landmark. If the land is privately owned, record the owner's details including contact telephone numbers.

6.2.5.3 HOW TO COLLECT AND SEND SOIL/ROOT SAMPLES

If plant roots are sampled, samples should be treated in a manner that allows them to arrive at the laboratory in a fresh, well-preserved state. An esky with ice packs or portable fridge should be carried when sampling crops. Samples need to be delivered to the appropriate laboratory immediately and should be transported in cool, moist conditions that avoid extremes of temperature (freezing or above 35°C). If samples are not processed within a day or two, they should be kept in a refrigerator. Samples should be considered perishable in 3-4 days (Bridge and Starr 2007).

If soil is sampled, dry soil can be taken (cysts are resistant to desiccation and very hardy). Composite soil samples should be kept in appropriately labelled plastic bags containing the name, address and contact phone number of both the sending and receiving officers. In addition bags and containers to be sent in should be clearly labelled in accordance with the requirements of PLANTPLAN (Plant Health Australia, 2013). For details consult the SOP for the *Collection and transport of EPPs* available as a supporting document of PLANTPLAN (Plant Health Australia, 2013) (**http://www.planthealthaustralia.com.au/wp-content/uploads/2013/12/SOP-Collection-and-transport-of-EPPs.pdf**).

6.2.5.4 EXTRACTION OF NEMATODES FROM SOIL

Mature (dead) nematode cysts of *H. avenae, H. latipons* and *H. filipjevi* occur in the soil and, because they float when dry, can be extracted from dry soil. The protocol for extraction using flotation can be undertaken using a Fenwick can. Alternatively a simple flotation extraction method comprises placing a sample of air dried soil (200 – 500 g) in a larger container. A strong jet of water is used to separate light organic matter fraction (containing cysts) to the surface without flowing over the container. Soil is allowed soil to settle for 30 seconds then the water containing the organic matter and cysts is poured through two nested sieves (250 µm on top and 150 µm on bottom). The process is repeated at least twice.

Material on the sieves is gently rinsed to the bottom of the sieve and then washed into a beaker. Organic matter in the beaker is concentrated onto filter paper or fine mesh material and examined for dry cysts.

White (immature) nematode cysts can also be observed by direct examination of plant roots however care must be taken as cysts can easily be dislodged as roots are washed free of soil.

6.2.6 Epidemiological study

The number of infected patches within a crop will depend on the amount of inoculum available and whether conditions have been favourable for the nematodes to spread from initial foci.

Sampling of crops within a district and beyond will be based upon the origins of the initial suspect sample(s). Factors to consider will be:

- Whether contaminated soil has moved from the infected paddock on vehicles or machinery.
- Whether dry cysts could have blown between paddocks.
- The proximity of other susceptible crops to the initial infected crop, both in the current growing season and previous season. This will include the growers own crops and those on neighbouring properties.
- It is likely that by the time an infestation of a new species of cereal cyst nematode (or a new strain of *H. avenae*) has been detected, the infestation will have been present for months or years.

6.2.7 Models of spread potential

No models of spread potential have been developed for Cereal cyst nematode. Cereal cyst nematodes would primarily be spread within and between crops by water, wind (cysts can be blown in soil) or especially in contaminated soil on boots, vehicles and machinery.

6.2.8 Pest Free Area guidelines

Determination of Pest Free Areas (PFAs) should be completed in accordance with the International Standards for Phytosanitary Measures (ISPMs) 8 and 10 (IPPC 1998a, 1999).

General points to consider are:

- Given the presence of an existing strain of *H. avenae* in Australia and the length of time it would take for an incursion of a new cereal cyst nematode strain or species to build up to noticeable levels, a new incursion may be present for several years prior to detection
- Given it will not be possible to declare paddock or area freedom on visible symptoms in crop growth, surveillance requirements to assess plant roots or soil for cysts would be extensive and may be cost prohibitive
- Design of a statistical delimiting field survey on host plants (see Section 6.2 for points to consider in the design)
- Soil sampling should be completed as described in 6.2.5.3
- Information (including absence of the pest) should be recorded

Additional information is provided by the IPPC (1995) in Requirements for the Establishment of Pest Free Areas. This standard describes the requirements for the establishment and use of pest free areas as a risk management option for phytosanitary certification of plants and plant products. Establishment and maintenance of a PFA can vary according to the biology of the pest, pest survival potential, means of dispersal, availability of host plants, restrictions on movement of produce, as well as PFA characteristics (size, degree of isolation and ecological conditions).

6.3 Availability of control methods

6.3.1 General procedures for control

- Keep traffic out of affected areas and minimise movement in adjacent areas
- Adopt best-practice property hygiene procedures to retard the spread of plant material (particularly plant roots) and soil containing nematodes between fields and adjacent properties
- After surveys are completed, and permission has been obtained from the Chief Plant Health Manager or OCPPO, destruction of the infested plant material is an effective control
- On-going surveillance of infected areas to ensure the pathogen is eradicated

Controlling Cereal cyst nematode populations before they reach large numbers in crops is crucial for any chance of eradication. However, due to the similarity of *H. filipjevi, H. latipons* and exotic strains of *H. avenae* with endemic *H. avenae* in both symptoms and morphology, the difficulty of identifying root disease symptoms and the relatively slow build up to levels where significant damage would be

observed, it is likely that an incursion would not be identified for several years. Probably as a result of these factors, there is no evidence of eradication of these pests overseas.

6.3.2 Chemical control

The nematicides aldicarb and dibromochloropropane have been shown to reduce levels of Cereal cyst nematodes and therefore improve cereal yields in infested paddocks southern Australia (Rovira *et al*. 1981). However, the only currently registered nematicide for control of endemic Cereal cyst nematode is carbofuran (registered as the product Furadan 360) for use on wheat and barley in South Australia and Victoria. In the event of an incursion, nematicides may be a key component of an eradication program depending on the extent of the incursion. However, the economics of using nematicides in ongoing management of Cereal cyst nematodes is questionable and other cultural and breeding options provide more economically viable control options.

6.3.3 Cultural Control

Strategies incorporating non-cereal crops (canola or grain legumes) into crop rotations may reduce the load of viable eggs in paddocks. In an eradication program, crop rotation (or fallowing) could be used in conjunction with chemical control to effectively control the nematodes if the area of the initial incursion was sufficiently small. The removal of cereal hosts for a minimum of 5 years would be required to ensure loss of viability in the majority of eggs. Advice may need to be sought from a Scientific Advisory Panel to confirm this length of time as it may be dependent on the type and scope of the incursion. Crop rotation forms an integral part of management strategies for the endemic strain of *H. avenae* (Wherrett and Vanstone 2010) and would be a key strategy in the management of other species of Cereal cyst nematode should they become endemic.

6.3.4 Host-Plant Resistance

Other than crop rotation, the use of resistant cultivars is the only economic method of control available for Cereal cyst nematodes. Resistance sources to endemic *H. avenae* have been identified (Ogbonnaya *et al*. 2001). While some sources of resistance currently used for *H. avenae* in wheat and barley in Australia have been found to be effective against *H. latipons* (Moklabi *et al*., 2002), substantial increased efforts would be required in breeding for resistance and tolerance of *H. filijevi, H. latipons* or new strains of *H. avenae*. Sources of genetic resistance in wheat are outlined in Table 3.

Table 3. *Sources of genes for resistance in wheat to Heterodera avenae (source: Smiley and Nicol 2009)*

Cereal	Genotype	Resistance gene	Use in cultivars
Triticum aestivum	Loros, AUS10894	*Cre1* (formerly *Ccn1*) on chromosome 2BL	NW Europe, Australia, NW USA – under evaluation
	Katyil	*Ccn*	Australia
	Festiguay	*Cre8* (formerly *CreF*) on chromosome 7L or 6B	Australia
	AUS4930 = Iraq48	Possible identical genetic location as *Cre1*; also resistant to *P. thornei*	Under evaluation in Australia, France and CIMMYT
	Molineux	Chromosome 1B	Australia
	Raj MR1	One dominant gene	Released cv in India
Triticum durum	Psathias 7654,7655, Sansome, Khapli	Not known	Not known
Triticosecale	T701-4-6	*CreR* on chromosome 6RL	Australia
	Drira = Ningadhu	Not known	Australia
	Tahara	Not known	Not known
	Salvo	Not known	UK
Secale cereale	R173 family	*CreR* on chromosome 6RL	Australia
Aegilops tauschii	CPI 110813	*Cre4* on chromosome 2DL	Australian synthetic hexaploid lines
	AUS 18913	*Cre3* on chromosome 2DL	Australian advanced breeding lines
Aegilops peregrina	1	*Cre(3S)* with Rkn2 on chromosome 3S; *CreX* not yet located	Not known
Aegilops longissima	18	Not known	France
Aegilops geniculate	79, MZ1, MZ61, MZ77, MZ124	Not known	Under evaluation in France
Aegilops triuncialis	TR-353	*Cre7* (formerly *CreAet*)	Under evaluation in France
Aegilops ventricosa	VPM 1	*Cre5* (formerly *CreX*) on chromosome 2AS	Under evaluation in Spain
	11; AP-1, H-93-8,	*Cre2* (formerly *CreX*) on genome N^v	
	11; AP-1, H-93-8, H-93-35	*Cre6,* on genome $5N^v$	

7 Course of action

Additional information is provided by the IPPC (1998b) in Guidelines for Pest Eradication Programmes. This standard describes the components of a pest eradication programme which can lead to the establishment or re-establishment of pest absence in an area. A pest eradication programme may be developed as an emergency measure to prevent establishment and/or spread of a pest following its recent entry (re-establish a pest free area) or a measure to eliminate an established pest (establish a pest free area). The eradication process involves three main activities: surveillance, containment, and treatment and/or control measures.

7.1 Destruction strategy

7.1.1 Destruction protocols

- General protocols:
    - No plant material should be removed from the infested area unless part of the disposal procedure.
    - Disposable equipment, infested plant material or soil should be disposed of by autoclaving, high temperature incineration or deep burial.
    - Any equipment removed from the site for disposal should be double-bagged.
    - Machinery used in destruction processes need to be thoroughly washed, preferably using a detergent or farm degreaser.

7.1.2 Decontamination protocols

Machinery, equipment and vehicles should be washed to remove plant material (roots) and soil using high pressure water in a designated wash down area. Scrubbing with products such as a degreaser or a bleach solution (1% available chlorine) may also assist with decontamination especially where juvenile forms of cysts nematodes may be expected to be present.

Steam heat treatment has been shown to be effective for disinfesting equipment from the related species, Potato cyst nematode. In studies undertaken by the United States Department of Agriculture, cysts that had been pre-soaked in water for 24 hours were killed when exposed to 30 seconds of 55^{o}C. This was in contrast to dry cysts which could tolerate 75^{o}C for brief periods (Brodie 1997).

When using high pressure water to remove infested soil from machinery and equipment, care should be taken not to spread plant material or soil. High pressure water should be used in wash down areas which meet the following guidelines:

- Located away from crops or sensitive vegetation.
- Readily accessible with clear signage.
- Access to fresh water and power.
- Mud free, including entry and exit points (e.g. packed gravel, concrete or rubber matting).

- Gently sloped to drain effluent away. Effluent must not enter water courses or water bodies.
- Allow adequate space to move larger vehicles and keep away from hazards such as power lines.
- Waste water, growing media/soil or plant residues should be contained (see PLANTPLAN supporting document *Disinfection and Decontamination guidelines* [Plant Health Australia, 2013]).
- Disposable overalls and rubber boots should be worn when handling infested plant material or soil in the field. Boots, clothes and shoes in contact with infested plant material or soil should be disinfected at the site or double-bagged to remove for cleaning.
- Skin and hair in contact with infested soil should be washed.

Procedures for the sterilisation of plant containers and growing media are provided within the BioSecure *HACCP* Guidelines, however, in the event of a Cereal cyst nematode incursion, additional or modified procedures may be required for the destruction of the pest. Any sterilisation procedure must be approved for use in the endorsed Response Plan.

7.1.3 Priorities

- Confirm the presence of the pest.
- Limit movement or people and prevent movement of vehicles and equipment through affected areas.
- Stop the movement of any plant material that may be infested with the pest.
- Determine the strategy for the eradication/decontamination of the pest and infested host material.
- Determine the extent of infestation through survey and plant material trace back and trace forward.

7.1.4 Plants, by-products and waste processing

- Any soil or infected plant material removed from the infected site should be destroyed by (enclosed) high temperature incineration, autoclaving or deep burial.
- Plant debris from the destruction zone must be carefully handled and transported.
- Infested areas or paddocks should remain free of susceptible host plants until the area has been shown to be free from nematodes.

7.1.5 Disposal issues

- Particular care must be taken to minimise the transfer of infected plant material (roots) or soil from the area.

7.2 Containment strategies

For some exotic pest incursions where eradication is considered impractical, containment of the pest may be attempted to prevent or slow its spread and to limit its impact on other parts of the state/territory or country. Containment is currently being considered for inclusion within the Emergency Plant Pest Response Deed (EPPRD). The decision on whether to eradicate or contain the pest will be made by the National Management Group, based on scientific and economic advice. Emergency interim containment measures are possible under EPPRD arrangements to gather information to determine if eradication is technically feasible.

7.3 Quarantine and movement controls

Consult PLANTPLAN (Plant Health Australia, 2013) for administrative details and procedures.

7.3.1 Quarantine priorities

- Plant material (especially plant roots) and soil at the site of infestation to be subject to movement restrictions. While it is less likely plant products such as hay and grain will be a source of movement of cysts, assessment should be made of plant products to ensure potential contamination of soil containing cysts does not occur.
- Machinery, equipment, vehicles and disposable equipment in contact with infested plant material (roots) or growing media/soil, or present in close proximity to the site of infestation to be subject to movement restrictions.

7.3.2 Movement controls

Movement controls need to be put in place to minimise the potential for transport of the pest, and this will apply to all plant material (especially roots), soil and other items within the quarantined area.

Movement of people, vehicles, equipment and plant material, from and to affected properties or areas, must be controlled to ensure that the pest is not moved off-property. Movement controls can be achieved through the following, however specific measures must be endorsed in the Response Plan:

- Signage to indicate quarantine area and restricted movement into and within these zones.
- Fenced, barricaded or locked entry to quarantine areas.
- Movement of equipment, machinery, or soil by permit only.
- Where no dwellings are located within these areas, strong movement controls should be enforced.
- Where dwellings and places of business are included within the Restricted and Control Areas movement restrictions are more difficult to enforce, however limitation of contact with infested soil should be enforced.
- Residents should be advised on measures to minimise the inadvertent transport of Cereal cyst nematode from the infested area to unaffected areas.

- Clothing and footwear worn at the infested site should either be double-bagged prior to removal for decontamination or should not leave the site until thoroughly cleaned, washed and disinfected.
- All machinery and equipment should be thoroughly cleaned down with a high pressure cleaner (see Section 7.1.2) or scrubbing with products such as a farm degreaser or a 1% bleach (available chlorine) solution or steam treated, prior to leaving the affected area. Machinery could be treated with methyl bromide fumigation or cleaned with steam to eliminate the chance of further spread. The clean down procedure should be carried out on a hard surface, preferably a designated wash-down area, to avoid mud being re-collected from the affected site onto the machine. When using high pressure water, care should be taken to contain all plant root material and mud dislodged during the cleaning process.

7.4 Zoning

The size of each quarantine area will be determined by a number of factors, including the location of the incursion, biology of the pest, climatic conditions and the proximity of the infested property to other infested properties. This will be determined by the National Management Group during the production of the Response Plan. Further information on quarantine zones in an Emergency Plant Pest (EPP) incursion can be found in the Section 4.1.4 of PLANTPLAN (Plant Health Australia, 2013). These zones are outlined below and in

Figure 8.

7.4.1 Establishing Quarantine Zones

Delimiting surveillance will inform the establishment of quarantine zones and identify the Restricted Area(s) (RA), Control Area (CA) and Pest Free Area (PFA). The size of each quarantine zone will be determined by a number of factors including location of the incursion, climatic conditions, pest biology and proximity of an Infected Premises (IP) to other IPs.

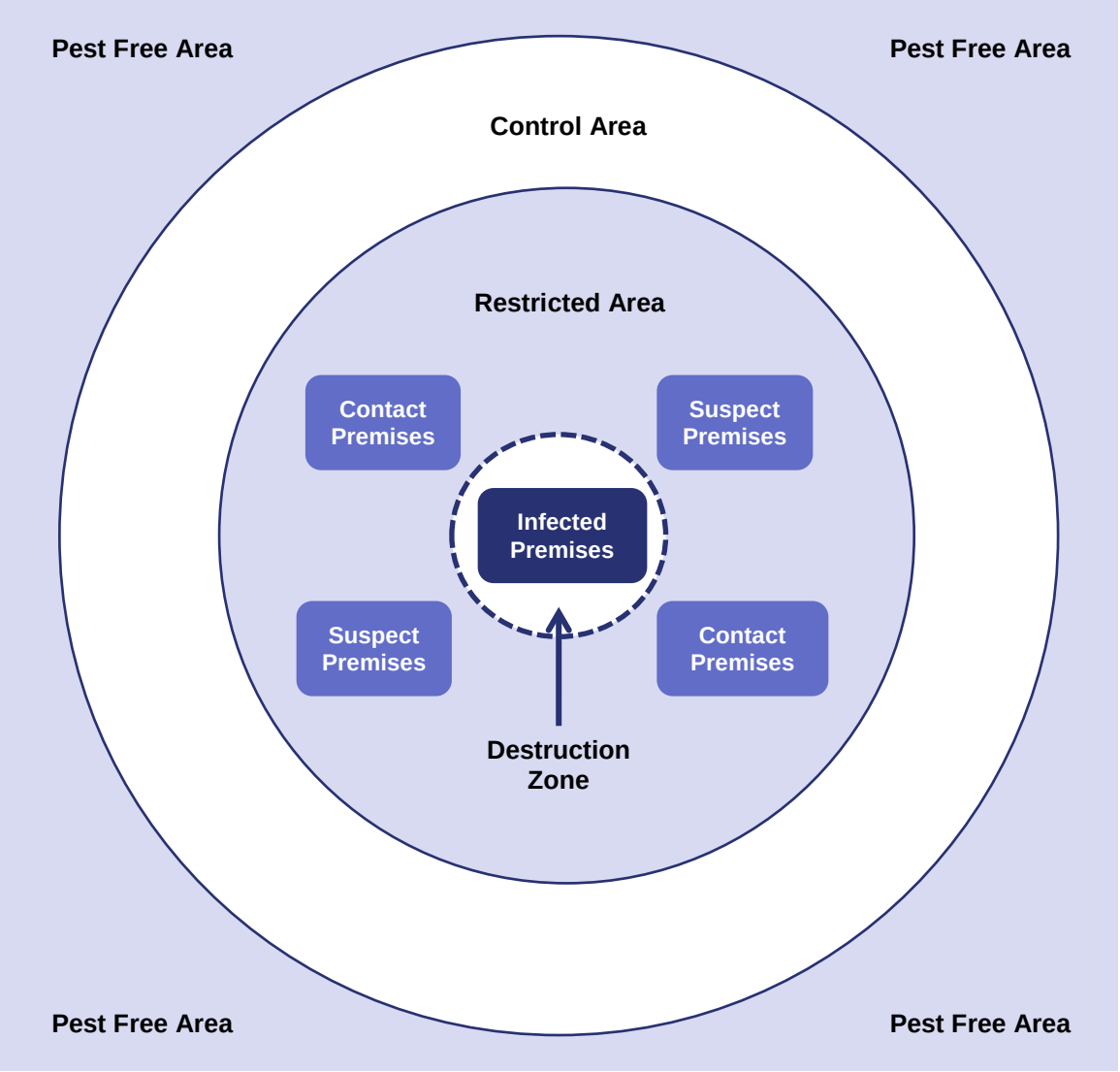

Figure 8. *Schematic diagram of quarantine zones used during an EPP incursion (not drawn to scale)*

7.4.2 Destruction Zone

The size of the Destruction Zone (i.e. zone in which the pest and all host material is destroyed) will depend on, distribution of the pest (as determined by delimiting surveys), ability of the pest to spread, factors which may contribute to the pest spreading and the time of season.

All host plants should be destroyed after the level of infection has been established. The delimiting survey will determine whether or not neighbouring host crops are infected and need to be destroyed. If spread is likely to have occurred prior to detection, the Destruction Zone may include contiguous areas that have been in contact with, or are associated with the same management practices as, the infected area Particular care needs to be taken to ensure that plant material and soil are not moved into surrounding areas that are not showing symptoms of the pest. Where possible, destruction should take place in dry conditions to limit mud being spread within the field on boots and protective clothing.

7.4.3 Restricted Area

Data collected from surveys and tracing (trace back and trace forward) will be used to define the RA, which comprises all properties where the pest has been confirmed (Infected Premises or IP), properties which have come into direct or indirect contact with an IP or infected plants (Contact Premises or CP) and properties which may have been exposed to the pest (Suspect Premises or SP). The RA will be subject to intense surveillance and movement control, with movement out of the RA to be prohibited and movement into the RA to occur by permit only.

7.4.4 Control Area

A Control Area or Buffer Zone is established around a Restricted Area to control the movement of susceptible hosts and other regulated materials until the extent of the incursion is determined. There may be multiple Restricted Areas within one Control Area. When the extent of the EPP Incident has been confidently defined, the Restricted Area and Control Area boundaries and movement controls may need to be modified, and where possible reduced in size commensurate with appropriate controls.

Additional zones can be utilised as required for operational purposes.

7.5 Decontamination and hygiene

Decontaminant practices are aimed at eliminating the pathogen thus preventing its spread to other areas.

7.5.1 Decontamination procedures

General guidelines for decontamination and clean up:

- Keep traffic out of affected area and minimise it in adjacent areas.
- Adopt best-practice property hygiene procedures especially for equipment to retard the spread of the pest between growing areas/fields and adjacent properties.
- Machinery, equipment, vehicles in contact with infested plant material or growing media/soil present within the Quarantine Zone, should be washed to remove soil and plant material using high pressure water or scrubbing with products such as a degreaser or a bleach solution or treated with steam in a designated wash down area as described in Section 7.1.2.
- Only recommended materials are to be used when conducting decontamination procedures, and should be applied according to the product label.
- If removed from the site, infested plant root material should be disposed of by autoclaving, high temperature (enclosed) incineration or deep burial.

For further information, refer to *Disinfection and decontamination* guidelines available as a supporting document of PLANTPLAN (Plant Health Australia, 2013) (**www.planthealthaustralia.com.au/wp-content/uploads/2013/12/Guidelines-Disinfection-and-decontamination.pdf**).

7.5.2 General safety precautions

For any chemicals used in the decontamination, follow all safety procedures listed within each MSDS.

7.6 Surveillance and tracing

7.6.1 Surveillance

Detection and delimiting surveys are required to delimit the extent of the outbreak, ensuring areas free of the pest retain market access and appropriate quarantine zones are established.

Initial surveillance priorities include the following:

- Surveying all host growing properties and businesses in the pest quarantine area.
- Surveying all properties identified in trace-forward or trace-back analysis as being at risk.
- Surveying all host growing properties that are reliant on trade with interstate or international markets which may be sensitive to pest presence.
- Surveying other host growing properties.

7.6.2 Survey regions

Establish survey regions around the surveillance priorities identified above. These regions will be generated based on the zoning requirements (see Section 7.4), and prioritised based on their potential likelihood to currently have or receive an incursion of this pest. Surveillance activities within these regions will either allow for the area to be declared pest free and maintain market access requirements or establish the impact and spread of the incursion to allow for effective control and containment measures to be carried out. Detailed information regarding surveys for Cereal cyst nematode have been outlined elsewhere in this plan (refer to Section 6.2).

Steps outlined in Table 4 form a basis for a survey plan. Although categorised in stages, some stages may be undertaken concurrently based on available skill sets, resources and priorities.

Table 4. *Phases to be covered in a survey plan*

Phase 1	Identify properties that fall within the buffer zone around the infested premise Complete preliminary surveillance to determine ownership, property details, production dynamics and tracings information (this may be an ongoing action)
Phase 2	Preliminary survey of host crops in properties in buffer zone establishing points of pest detection
Phase 3	Surveillance of an intensive nature, to support control and containment activities around points of pest detection
Phase 4	Surveillance of contact premises. A contact premise is a property containing susceptible host plants, which are known to have been in direct or indirect contact with an infested premises or infected soil. Contact premises may be determined through tracking movement of materials from the property that may provide a viable pathway for spread of the disease. Pathways to be considered are: • Items of equipment and machinery which have been shared between properties including vehicles and equipment • The producer of infected material if this is suspected to be the source of the incursion • Labour and other personnel that have moved from infested, contact and suspect premises to unaffected properties (other growers, tradesmen, visitors, salesmen, crop scouts, harvesters and possibly beekeepers) • Movement of plant material and soil from controlled and restricted areas • Storm and rain events that may spread the pest
Phase 5	Surveillance of paddocks, gardens and public land where plants known to be hosts of pest are being grown
Phase 6	Agreed area freedom maintenance, post control and containment

7.6.3 Post-eradication surveillance

The period of pest freedom sufficient to indicate that eradication of the pest has been achieved will be determined by a number of factors, including growth conditions, the previous level of infestation, the control measures applied and the pest biology.

Specific methods to confirm eradication of Cereal cyst nematode may include:

- Monitoring for symptoms in host plants
- If symptoms are detected, samples are to be collected and stored and plants destroyed
- Surveys comprising soil/root sampling should be undertaken for a minimum of 3 years after eradication has been achieved
- Alternate non-host crops should be grown on the site and any self-sown plants sprayed out with a selective herbicide

7.7 Technical debrief and analysis for stand down

Refer to Section 4.3 of PLANTPLAN (Plant Health Australia, 2013) for further details.

The emergency response is considered to be ended when either:

- Eradication has been deemed successful by the lead agency, with agreement by the Consultative Committee on Emergency Plant Pests and the Domestic Quarantine and Market Access Working Group.
- Eradication has been deemed impractical and procedures for long-term management of the disease risk have been implemented.

A final report should be completed by the lead agency and the handling of the incident reviewed.

Eradication will be deemed impractical if, at any stage, the results of the delimiting surveys lead to a decision to move to containment/control.

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8.1 Related Websites

CABI 2007 **www.cabicompendium.org/cpc/home.asp**

IPPC website **www.ippc.int**

9 Appendices

9.1 Appendix 1: Standard diagnostic protocols

For a range of specifically designed procedures for the emergency response to a pest incursion refer to Plant Health Australia's PLANTPLAN (**www.planthealthaustralia.com.au/plantplan**).

9.2 Appendix 2: Resources and facilities

Formal diagnostic services for plant pests in Australia are delivered through a network of facilities located in every state and territory. These services are provided by a range of agencies, including state and territory governments, the Australian Government, commercial and private diagnostic laboratories, museums, CSIRO and universities. A current listing of these facilities can be found at **www.npbdn.net.au/resource-hub/directories/laboratory-directory**.

The national network is supported by the Subcommittee on Plant Health Diagnostic Standards (SPHDS), which was established to improve the quality and reliability of plant pest diagnostics in Australia. SPHDS also manages the production of National Diagnostic Protocols.

For more information on the diagnostic services, or to identify an appropriate facility to undertake specific pest diagnostic services, refer to **www.npbdn.net.au** or contact the SPHDS Executive Officer through **SPHDS@daff.gov.au**.

9.3 Appendix 3: Communications strategy

A general Communications Strategy is provided in Section 4.1.5 of PLANTPLAN (Plant Health Australia, 2013).

9.4 Appendix 4: Market access impacts

Within the Department of Agriculture MICoR (plants) data base export of some material may require an additional declaration regarding freedom from some species of Cereal cyst nematode. Should exotic species of Cereal cyst nematode be detected or become established in Australia, some countries may require specific declaration. Latest information can be found with MICoR (plants) using a search for the particular nematode. For further information on MICoR see website at **http://www.daff.gov.au/micor/plants**.