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Cereal Cyst Nematodes

Biology and management in Pacific Northwest wheat, barley, and oat crops

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Nematodes are tiny but complex unsegmented roundworms that are anatomically differentiated for feeding, digestion, locomotion, and reproduction. These small animals occur worldwide in all environments. Most species are beneficial to agriculture. They make important contributions to organic matter decomposition and the food chain. Some species, however, are parasitic to plants or animals. One type of plant-parasitic nematode forms cysts that contain eggs from which juvenile nematodes hatch to damage and reduce yields of many agriculturally important crops.

The cyst nematode genus Heterodera contains as many as 70 species, including a complex of 12 species known as the Heterodera avenae group. Species in this group invade and reproduce only in living roots of cereals and many species of grasses. They do not reproduce on any broadleaf plant. Three species in the H. avenae group (H. avenae, H. filipjevi, and H. latipons) cause important economic losses in small grain crops worldwide and are known as the cereal cyst nematodes. Heterodera avenae and H. filipjevi are the only two species of cereal cyst nematode that are known to be present in the Pacific Northwest (PNW). Figure 1 shows the distribution and the year of first detection of cereal cyst nematodes in the western United States. Heterodera avenae is the most commonly encountered species. Heterodera filipjevi is also present in some areas of Oregon, Montana, and Washington. Where both species are known to be present, they may occur in individual fields either as only one of the species or as a mixture of the two species.

The infective stage of these cereal cyst nematodes can be seen only by using specialized methods to remove them from soil and view them through a microscope. The infective stage is transparent, eelshaped, and about 0.5 mm (½4 inch) long. These



Figure 1. Geographic distribution of *Heterodera avenae* (Ha) and *H. filipjevi* (Hf), and the year in which they were first detected in the western United States. Image by Dr. Richard Smiley, © Oregon State University.

nematodes puncture root cells, which reduces rooting depth, plant vigor, and uptake of water and nutrients.

Plant-parasitic nematodes are difficult to identify and to demonstrate as the cause of important crop damage. Symptoms expressed in the foliage are easily mistaken as symptoms of irregularities in soil depth, soil texture, soil pH, mineral nutrition, and water availability, or with diseases such as barley yellow dwarf. The foliar symptoms of cereal cyst nematode infestation also have many of the same characteristics as symptoms of root diseases such as Rhizoctonia root rot or take-all. Farmers, pest management advisors, and scientists routinely

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underestimate or fail to recognize the impact of cereal cyst nematodes on small grain cereals. It was conservatively estimated that this generally unrecognized pest reduces wheat profitability by at least \$3.4 million annually in the PNW states of Idaho, Oregon, and Washington.

Biology

The biology of cereal cyst nematodes is complex. There are several species, and each species may be composed of different pathotypes (i.e., strains or races). Pathotypes vary in their ability to reproduce on individual varieties and species of cereal crops. *Heterodera avenae* and *H. filipjevi* are the only cereal cyst nematodes currently known to occur in the PNW. Research in Europe and the Middle East has shown that combinations of these species and of several pathotypes of each species may occur within a region and even within an individual field. The reproductive capacity of the *Heterodera* population in each geographic region must be understood before successful sources of plant resistance can be introduced into those regions. Fortunately, the current situation in the PNW is simpler because multiple pathotypes have not yet been identified in this region. *Heterodera avenae* populations from Idaho, Oregon, and Washington reproduce nearly the same on individual indicator varieties of barley, oat, and wheat, which means that resistance data generated in one region should also be applicable to other PNW states and regions.

Heterodera species complete only one generation of their life cycle during each crop season (Figure 2). The first molt occurs inside the cyst, whereupon the egg hatches to transform into a second-stage juvenile. The infective second-stage juvenile (Figure 3, page 3) has a translucent, vermiform-shaped (like an eel) body that is about 0.5 mm ($\frac{1}{64}$ inch) long and 0.02 mm ($\frac{1}{1000}$ inch) in diameter. For comparison, the diameter of a human hair (0.1 mm) is about five times greater than the diameter of the infective juvenile stage of this nematode.



Figure 3. Second-stage juveniles of *Heterodera* species: A) This stage of *H. avenae* (1/1000 inch in diameter and 1/64 inch long) emerges from a cyst, migrates through soil, invades the root, and migrates through the root cortical tissue. B) A closer view of the feeding stylet of *H. trifolii* (clover cyst nematode).

A – Photo by Dr. Guiping Yan; Courtesy of Dr. Richard Smiley, Oregon State University; B – Courtesy of Dr. Lynn Carta, USDA-Agricultural Research Service, Beltsville, Maryland.

Second-stage juveniles emerge from openings (semifenestrae) in the vulval cone of the cyst (Figure 4), enter the soil, and migrate toward the roots. When they find a susceptible cereal host, they penetrate epidermal cells near the tips of new roots by rapidly thrusting the needle-like stylet (Figure 3). The juveniles initially migrate through the epidermis and into the root cortical cells until they select a permanent feeding site near the root phloem. They inject secretions into the cell cytoplasm that induce the formation of enlarged, specialized feeding cells called syncytia. The second-stage juveniles molt to become third-stage juveniles, at which time they differentiate into males or females. The third- and fourth-stage female juveniles become inflated (Figure 2, page 2) and sedentary. Males regain mobility, but females remain embedded in the root tissue and continue to feed from the syncytium. As the females enlarge they rupture the root surface and become exposed to the surrounding soil. The males migrate from the root to re-enter the soil, whereupon they copulate with the sedentary females. The female body becomes heavily swollen and white colored as it fills with 100 to 600 fertilized eggs.

The presence of a white, swollen female embedded in a root but having a visibly globose body outside the root is diagnostic (Figure 2, page 2). A swollen female body is about the size of a pinhead, varying in diameter from 0.5 to 2 mm ($\frac{1}{64}$ to $\frac{1}{16}$ inch). It protrudes from the root surface, glistens when wet, is white to light gray, and can be seen most easily when the invaded plant is flowering. This structure is best viewed by gently washing the roots and observing the root mass under low magnification ($\times 20$). Among knotted roots, adhering soil often obscures white females, but one or more are generally visible at points where there is an abnormal proliferation of branch roots (Figure 5, page 4). Females are attached loosely and dislodge easily when soil is washed vigorously from roots.

When invaded roots mature and die, the female dies, and the outer membrane hardens into a leathery, light-brown cyst (Figure 2, page 2) that protects the eggs and is the same size and color



Figure 4. Structure of the vulval cones of cereal cyst nematodes, illustrating diagnostic features required to distinguish among species: A) Location of the vulval cone on an egg-filled cyst of *Heterodera avenae*, B) three-dimensional drawing with a cut-away to illustrate internal structures of *H. schachtii* (sugar beet cyst nematode) observable with a scanning electron microscope, C) photograph showing structures of the vulval cone of *H. filipjevi* that are observable using a standard light microscope.

A – Courtesy of Dr. Haiyan Wu, Agricultural College of Guangxi University, Nanning, China; B – Courtesy of Dr. Sergei Subbotin, California Department of Food and Agriculture, Sacramento; C – Image by Dr. Guiping Yan, Courtesy of Dr. Richard Smiley, Oregon State University





Figure 5. Shallow rooting (A, B) and abnormal branching (C) or root termination (D) at sites where *Heterodera avenae* juveniles invaded roots. Image A shows unwashed roots dug from soil; roots in the other images have been washed to remove soil. Image B compares roots of spring wheat that were lightly invaded (left) or heavily invaded (right) by the nematode.

Photos by Dr. Richard Smiley, © Oregon State University

as many soil particles. Most dry cysts dislodge into the soil as the wheat roots decompose and as soil is tilled. The cysts darken with age and protect eggs and juveniles from desiccation, cold, and heat between crop cycles. Eggs inside a cyst can remain viable for several years.

Infective *H. avenae* juveniles emerge from cysts after a necessary period

(2 months or more) of cold temperature. In the Willamette Valley of western Oregon, infective juveniles are found in soil from late January to late April, with peak numbers during late February (Figure 6). In colder regions of eastern Oregon (and presumably also in Idaho, Montana, and Washington), infective juveniles move into soil from late February to late May, with peak numbers during mid-April.

Emergence of *H. filipjevi* juveniles has not been fully investigated in the PNW. In a preliminary study conducted during spring in eastern Oregon, peak



Figure 6. Density of infective *Heterodera avenae* juveniles present in soil during the spring on farms in western Oregon (dotted line) and eastern Oregon (solid line).

Graph by Dr. Richard Smiley, © Oregon State University.



numbers of *H. filipjevi* juveniles in soil occurred at least 2 weeks earlier than peak numbers of *H. avenae* juveniles. However, recent reports from Asia, Europe, and the Middle East indicate that, in those regions, *H. filipjevi* juveniles did not require a period of cold temperature before emerging from cysts. The juveniles began to emerge during late fall from cysts developed on recently harvested crops of wheat or barley. The emergence rate of juveniles decreased over the winter then resumed and peaked during early spring. Research overseas has shown that root invasions during the spring have caused much greater damage than invasions during the autumn.

It is important to identify the species of cereal cyst nematode that occurs in each field or region because wheat, barley, and oat varieties may differ in their response to attack by *H. avenae* and *H. filipjevi*. Methods to detect and distinguish these species are complex and traditionally have been based on careful measurements of body features viewed under high magnification (Figure 4, page 3). Primary diagnostic features to distinguish among species include minute differences in the size and shape of the cyst's semifenestrae, the shape or absence of an underbridge, the length of the vulval slit, and the density or absence of bullae (Figure 4, page 3). Primary diagnostic features of second-stage juveniles (Figure 3, page 3) include an overall Heteroderalike appearance, the shape and size of the stylet,

the distance from the vulva to the tip of the tail compared to the length of the entire body, and the length of the translucent portion of the tail compared to the length of the entire tail. To circumvent these diagnostic challenges, particularly for those who are not skilled in nematode systematics, molecular (DNA-based) procedures are now available that identify these nematodes more accurately and rapidly (Figures 7 and 8).

Symptoms

Specific symptoms occur only on roots, and the type of symptom varies by host species. To detect root symptoms, gently wash the soil from the



Figure 7. Identification of *Heterodera avenae* (Ha) and *H. filipjevi* (Hf) using routine species-specific PCR assays of DNA extracted from soil: A) detection of *H. avenae* at the 242 bp band position, B) detection of *H. filipjevi* at the 170 bp band position.



Images by Dr. Guiping Yan; Courtesy of Dr. Richard Smiley, Oregon State University.

Figure 8. Identification of *Heterodera avenae* (Ha), *H. filipjevi* (Hf), and mixtures of these species from 11 wheat fields in Washington, using a more complex PCR-RFLP assay.

Image by Dr. Guiping Yan; Courtesy of Dr. Richard Smiley, Oregon State University.

roots, and then closely observe the root branching pattern. Abnormal rooting patterns are generally not recognizable until a month or more after infective juveniles have invaded the root. Wheat and barley roots branch excessively at locations where cereal cyst nematode females have established a feeding syncytium. The result is a bushy or knotted appearance on the root (Figure 5, page 4). Invaded roots often fail to continue growing deeply into soil at sites where nematodes have caused abnormal branching. The roots of invaded plants are shallower and less capable of extracting soil water and nutrients than roots of healthy plants. When H. avenae invades oats, the roots become shortened and larger in diameter but do not develop the knotted symptom.

Plants with heavily damaged roots often appear initially as unthrifty, pale green seedlings that occur in patches (Figure 9, page 6). Damage may become widespread and uniform over entire fields when susceptible cereals are planted frequently in the crop rotation. Symptoms become more pronounced when affected plants are also exposed to a stress such as inadequate nutrition, shallow soil, or a shortage of available water (Figure 10, page 7). Affected plants generally do not respond well to additional applications of fertilizer or water. Plants with heavily damaged roots may be severely stunted and may mature early, similar to plants affected by root and crown diseases such as take-all.

Root tissues invaded by cereal cyst nematodes have a greater risk of additional damage by rootrotting fungi and saprophytic bacteria, fungi, and other nematodes. The rotting and discoloration caused by these secondary organisms, however, are not direct symptoms of root invasion by cereal cyst nematodes.

Yield Reduction

Grain yields are often negatively correlated with the number of cereal cyst nematodes in soil during the spring time (Figure 11, page 7). However, a definite relationship between the number of nematodes and the magnitude of yield suppression is difficult to generalize because yield responses are strongly influenced by interactions between climate, crop variety, management practice, and nematode distribution and density within the field as well as chemical, biological, and physical properties of soil.



Figure 9. Symptoms of wheat injury caused by the cereal cyst nematodes Heterodera avenae or H. filipjevi in winter wheat (A-C), oats (D), or spring wheat (E-F). Patches were caused by *H. avenae* in Union County, Oregon (A-B & E-F), by H. avenae in Washington County, Oregon (D), or by H. filipjevi in Whitman County, Washington (C). Images E and F are from fields of a single farm but on opposite sides of a road and with different management practices; the crop in (E) shows a combined effect of *H. avenae* plus inadequate plant nutrition on a third consecutive crop of irrigated annual spring wheat (symptoms in the foreground were masked by a doubled rate of fertilizer applied along the field border), and the crop in (F) shows the same variety of spring wheat on an adjacent field managed as a spring wheat-cultivated fallow rotation. Photos by Dr. Richard Smiley, © Oregon State University.



Figure 10. Combined stresses from cereal cyst nematode and nutrient deficiency, showing distinct patches caused by *H. avenae* in a single field of winter wheat in Whitman County, Washington (A–B), with the patch 'A' having more intense yellowing on the eroded hilltop where the soil is more shallow and lower in fertility compared to patch 'B' that occurred on the deeper and more fertile soil on the flat in the background, and (C) a crop of spring wheat in Fremont County, Idaho showing where an equipment malfunction caused plugging of the starter fertilizer tubes on the outer two rows of a 4-row plot drill as compared to normal functioning of tubes for the inner two rows.

Photos by Dr. Richard Smiley, © Oregon State University.



Figure 11. Relationship between yield of irrigated winter wheat (left) or dryland spring wheat (right) and the number of *Heterodera avenae* before planting in two typically infested fields in Union County, Oregon. Note that greater densities of eggs + juveniles (J2) were required to reduce yield of irrigated winter wheat compared to dryland spring wheat.

Graphs by Dr. Richard Smiley, © Oregon State University.

For instance, the importance of a given density of nematodes at the time of planting will become greater if affected plants are later subjected to drought, inadequate nutrition, impediments to root penetration into soil, or adverse temperature. The potential for damage may also differ among varieties if, for example, those varieties have different abilities to replace damaged roots. Winter wheat typically has a well-established root system at the time juveniles emerge from cysts during the spring. In contrast, when spring grains are planted, juveniles are already present in soil and continue to emerge from cysts as the spring grain seedlings become established. For that reason, spring cereals are often more severely affected than winter cereals by a given density of juveniles in soil during the spring.

In general, research in the PNW has demonstrated that reduced wheat yields may occur when the number of *H. avenae* eggs plus juveniles from cysts plus juveniles already present within the soil matrix exceeds three nematodes per gram of soil, which is approximately 1,400 nematodes per pound (or pint) of soil. This density is often exceeded in at least some portions of infested fields in Idaho, Oregon, and Washington. Research in Iran, under climatic conditions similar to those of the inland PNW, has shown that *H. filipjevi* densities of 0.7 nematodes per gram of soil (320 eggs plus juveniles per pound of soil) were capable of reducing the yield of winter wheat.

Crop Management

After cereal cyst nematodes have been detected in a field, damage can be minimized by reducing the density to fewer than 1,000 eggs plus second-stage juveniles per pound of soil. Commercial labs report nematode densities in a given volume or weight of soil. Approximate conversions are as follows:

- Reports based on "500 cc of soil" represent approximately 1 pound or 1 pint of soil.
- Reports based on "100 g of soil corrected for moisture" represent approximately ¼ of a pound.

Management of cereal cyst nematodes involves an integrated approach that includes field sanitation, crop rotation, weed control, genetic resistance, timing of planting, crop nutrition, and water supply.

Field sanitation. Once cereal cyst nematodes have been introduced into a field, eradication is nearly impossible. Therefore, efforts to minimize the transmission of soil from infested to noninfested

fields or regions are of critical importance. These nematodes are transmitted in all manners in which soil is moved from location to location. A common means of transport is soil that adheres to equipment, vehicles, animals, humans (boots), and plant products such as root and tuber crops, turfgrass sod, and some horticultural nursery crops. Cysts with viable eggs are also carried in dust blowing from infested fields and in soil carried by water moving from infested fields either by erosion or in return ditches at the discharge end of flood-irrigated fields. Viable cysts are also transmitted though the gut of birds that have fed on grain or on insects in infested soils.

Crop rotation. Damage from cereal cyst nematodes is greatest when susceptible crops are produced annually. Yield losses can also become very high in 2-year rotations (cereals with summer fallow or a crop such as potato) and 3-year rotations (e.g., winter wheat, a spring cereal, and a nonhost broadleaf crop or fallow). Crop rotations that include several years of broadleaf crops, corn, fallow, or resistant wheat, barley, or oat varieties can greatly reduce the nematode density. In general, growing a susceptible host only once during a 3- to 4-year period can dramatically reduce the density of *H. avenae* in soil.

Cereal cysts nematodes are also capable of reproducing on a wide range of economically important grasses that include bentgrass, bluegrass, fescue, ryegrass, brome, orchard grass, canary grass, timothy, and sorghum. These crops should not precede wheat, barley, or oat in crop rotations on fields where cereal cyst nematodes are known to be present.

Weed control. Cereal cysts nematodes may also persist on a wide range of weed grasses. Grassy weeds such as quackgrass, crabgrass, brome grass, foxtail, wild oat, rat-tail fescue, and others should not be allowed to grow during any phase of a crop rotation in a field that is infested with a cereal cyst nematode.

Resistance and tolerance of wheat and barley. International research with *H. avenae* shows that the susceptibility of small grain crops and the potential for yield reduction occurs in the following order of increasing susceptibility and yield reduction: rye and winter barley, spring barley, winter wheat, spring wheat, winter oats, and spring oats. Research in the PNW has been focused on spring wheat, spring barley, and winter wheat, which are currently the crops of primary importance in the region.

Production of varieties that are both resistant and tolerant is an effective method of controlling cereal cyst nematodes. Resistance is defined as the ability of the host to reduce or inhibit nematode multiplication (Figure 12). The opposite term for resistant is susceptible. Susceptible crops enable the nematodes to multiply prolifically. The benefit of



Figure 12. Reproduction of *Heterodera avenae* on wheat roots in naturally infested soil. The *Cre 1* and *Cre 5* resistance genes in wheat are effective against populations of this nematode in the PNW.

Graph by Dr. Richard Smiley, © Oregon State University.



Figure 13. Density of *Heterodera avenae* in soil after harvesting two susceptible varieties (Alpowa and Louise) and two resistant varieties (Ouyen and WB Rockland) at St. Anthony, Idaho; the dashed line approximates the number of residual eggs in cysts produced on previous susceptible crops of wheat or barley, and the bars extending above that line are an estimate of eggs contained within cysts formed on the most recently harvested susceptible varieties, either treated with nematicide (blue) or not treated (red) at the time of planting. resistance is that it reduces the density of nematodes remaining in soil (Figure 13), thereby reducing the risk of nematode damage to the next crop of barley, oats, or wheat. However, even when reproduction is prevented or suppressed by resistance, infective juveniles usually invade and injure roots of resistant plants, which can reduce yield of the current resistant crop.

Tolerance is defined as the ability of plants to produce acceptable yields even when cereal cyst nematodes are present. Tolerance can be estimated by comparing grain yields in adjacent trial plots that have either a high or low (or nil) population of nematodes. These conditions can be established either by growing varieties in naturally infested soil with alternating strips of nematicide-treated and untreated soil (Figure 14) or by growing



Figure 14. Moderately intolerant Alpowa (top) and more tolerant Louise (bottom) spring wheat growing in soil naturally infested by *Heterodera avenae* at Steptoe, Washington. The 4-row drill strip on the left within each image was not treated with nematicide and the drill strip on the right was treated with a non-registered nematicide for this research.

Photos by Dr. Richard Smiley, © Oregon State University.

Chart by Dr. Richard Smiley, © Oregon State University.

varieties in adjacent blocks of non-infested soil that are left natural or inoculated with nematodes. It is important to note that tolerance estimates the yielding capacity of the current crop only and does not indicate the yielding capacity of the following crop. Intolerant plants may incur significant yield declines in fields infested by cereal cyst nematode.

Tolerant and intolerant plants alike can be either resistant or susceptible, and resistant plants can be either tolerant or intolerant. Tolerance and resistance are genetically independent, and therefore all combinations of resistance and tolerance are possible within a collection of varieties. For example, tolerant varieties that are susceptible may produce normal yields but allow the nematode to multiply, posing a higher risk to a subsequent crop. Alternately, roots of some resistant wheat varieties may be very sensitive (intolerant) to the initial invasion by the cereal cyst nematode, resulting in reduced growth and yield.

Neither resistance nor tolerance alone is considered an effective long-term management strategy. Management of cereal cyst nematodes will require development of wheat and barley varieties that are both resistant and tolerant to both *H. avenae* and *H. filipjevi*.

Wheat and barley varieties with both resistance and tolerance are being identified to increase productivity in fields with dense populations of cereal cyst nematodes. However, varieties with resistance to *H. avenae* are not necessarily resistant to *H. filipjevi*, and vice versa (Table 1). Likewise, varieties that are tolerant to *H. avenae* are not necessarily tolerant to *H. filipjevi*, and vice versa. In general, spring barley has exhibited greater stability (more resistance and more tolerance) than spring wheat in fields infested with *H. avenae*. Although not tested, it is likely that the same relationship exists for winter barley and winter wheat.

Varieties of wheat and barley that are resistant, tolerant, or both resistant and tolerant are being identified in the PNW (Tables 2-5, pages 11-14). Genes that greatly suppress reproduction of *H*. avenae populations in the PNW have been identified and are being transferred into wheat varieties that are agronomically adapted to climatic conditions in the PNW. The most effective of the currently identified wheat resistance genes for controlling pathotypes of *H. avenae* in the PNW is the Cre1 gene (Table 6, page 15; Figure 12, page 9, and Figure 15, page 13), and that gene has been crossed into locally adapted varieties. The Cre5 gene is another gene of potential value in the PNW, particularly if pyramided with the Cre1 gene within a single variety. Several varieties with unknown sources of resistance have been identified in the PNW using variety trials planted into naturally infested fields or soils collected from those fields (Tables 1-5, pages 10–14; Figure 15, page 13). Additional resistance tests for both H. avenae and H. filipjevi are currently underway in Idaho and Washington, and several more promising resistant varieties and breeding lines have been identified. Resistance in locally adapted oat varieties has not been studied in the PNW.

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Variety	White females ¹		Resistance rating ²			
	H. avenae	H. filipjevi	H. avenae	H. filipjevi		
Louise	33	60	VS	VS		
Ouyen	1	19	R	S		
WB 936	22	46	S	VS		
WB-Rockland	1	31	R	VS		
Sönmez	24	3	S	R		
SY Steelhead	26	6	VS	MR		

Table 1. Number of *Heterodera avenae* or *H. filipjevi* white females produced on roots, and nematode resistance ratings of six spring wheat cultivars growing in naturally infested soils collected from fields in eastern Washington.

¹ Average number of *H. avenae* white females produced on roots of each plant in the control (no-nematicide) treatment.

² Resistance ratings were very resistant (VR; \leq 1 white female produced on roots of one plant), resistant (R; 1.1–3), moderately resistant (MR; 3.1–6), moderately susceptible (MS; 6.1–12), susceptible (S; 12.1–25), or very susceptible (VS; > 25).

Table 2. Evaluation of 20 spring wheat varieties for resistance and tolerance to *Heterodera avenae*; data are means of field screening trials on two farms in eastern Washington and southeast Idaho, using three replicates per entry on each of two treatments (with or without application of a nematicide).

Market class	Variety	White females ¹	Resistance rating ²	Yield increase (%) ³	Tolerance rating⁴	MR + MT⁵
Hard red	WB-Rockland	1	VR	5	Т	Х
	Cabernet	9	MS	9	Т	
	Bullseye	10	MS	19	MI	
	Jedd	12	MS	8	Т	
	Buck Pronto	13	S	4	VT	
	Westbred 936	16	S	12	MT	
	Kelse	17	S	8	Т	
	Glee	18	S	7	Т	
	Jefferson	29	VS	6	Т	
Hard white	Klasic	8	MS	18	MI	
	Otis	14	S	17	MI	
Soft white	Alturas	9	MS	12	MT	
	Louise	10	MS	6	Т	
	Alpowa	12	MS	19	MI	
	UI Pettit	12	MS	9	Т	
	WB 1035Cl+	15	S	11	MT	
	Babe	18	S	6	Т	
	UI Stone	20	S	3	VT	
Soft white club	JD	9	MS	9	Т	
Australian standard white	Ouyen	1	VR	11	MT	Х

¹ Average number of *H. avenae* white females produced on roots of each plant in the control (no-nematicide) treatment.

² Varieties were rated as very resistant (VR; ≤ 1 white female/plant), resistant (R; 1.1–3), moderately resistant (MR; 3.1–6), moderately susceptible (MS; 6.1–12), susceptible (S; 12.1–25), or very susceptible (VS; >25).

³ Percentage increase in grain yield due to application of a nematicide that is used in research but is not registered for use in commercial fields; the non-fumigant type nematicide was metered into or below the seed row at the time of planting. ⁴ Varieties were rated as very tolerant (VT; <5% yield increase by application of nematicide), tolerant (T; 5–10%), moderately tolerant (MT; 10–15%), moderately intolerant (MI; 15–30%), intolerant (I; 30–50%), or very intolerant (VI; >50%).

⁵ Entries having at least moderate resistance (≤6% white females/plant) plus moderate tolerance (≤15% yield increase).

Table 3. Evaluation of 39 spring wheat varieties for resistance and tolerance to <i>Heterodera avenae</i> ; data are
means of field screening trials during two years on a farm in southeast Idaho, using four replicates per entry on
each of two treatments (with or without application of a nematicide).

Market class	Variety	White females ¹	Resistance rating ²	Yield increase (%) ³	Tolerance rating⁴	MR + MT⁵
Hard red	WB-Rockland	1.5	R	14.5	MT	Х
	Jefferson	7.7	MS	38.7	I	
	WB9576	7.8	MS	5.8	Т	
	UI Platinum	8.9	MS	20.7	MI	
	WB9229	10.4	MS	26.6	MI	
	Choteau	11.9	MS	18.3	MI	
	Glee	12.4	S	16.9	MI	
	Kelse	13.0	S	18.3	MI	
	Cabernet	15.2	S	21.7	MI	
	IDO862T	15.2	S	16.1	MI	
	LCS Iron	16.1	S	24.1	MI	
	IDO12025	18.3	S	27.6	MI	
	UI Winchester	19.3	S	19.7	MI	
	Alzada	21.4	S	12.5	MT	
	IDO862E	23.9	S	25.0	MI	
	Bullseye	24.2	S	16.6	MI	
	SY Basalt	24.7	S	23.8	MI	
	Volt	31.2	VS	35.4	I	
	Westbred 936	60.8	VS	41.9	I	
Hard white	LCS Star	2.6	R	15.5	MI	(X)
	Klasic	4.4	MR	15.0	MT	Х
	Dayn	7.5	MS	14.3	MT	
	WB-Idamax	16.2	S	22.4	MI	
	Blanca Grande	20.8	S	10.4	MT	
	Snow Crest	26.0	VS	23.7	MI	
	WB-Paloma	26.2	VS	25.8	MI	
Soft white	Cataldo	5.2	MR	39.1	I	
	Alpowa	8.9	MS	7.1	Т	
	IDO 852	10.6	MS	17.8	MI	
	UI Stone	11.1	MS	18.7	MI	
	Babe	15.7	S	20.4	MI	
	UI Pettit	19.7	S	17.7	MI	
	WB6121	20.5	S	11.6	MT	
	Penawawa	21.7	S	17.0	MI	
	IDO 854	23.0	S	27.9	MI	
	Alturas	25.0	S	16.1	MI	
	IDO 851	26.6	VS	18.0	MI	
	Seahawk	26.6	VS	17.2	MI	
	WB6430	28.9	VS	14.5	MT	

¹⁻⁵ Footnotes 1–4 are described under Table 2. (X) indicates a variety that very nearly met the limits for resistance plus tolerance, and can therefore be considered to be functionally equivalent to that dual-trait criteria in commercial agriculture.

Market class	Variety	White females ¹	Resistance rating ²	Yield increase (%) ³	Tolerance rating⁴	MR + MT⁵
2-row	Lenetah	3	R	10	Т	Х
feed barley	Xena	3	MR	5	Т	Х
	CDC Fibar (hull-less)	4	MR	18	MI	
	Idagold II	5	MR	11	MT	Х
	Transit	5	MR	11	MT	Х
	Julie	6	MS	2	VT	
	RWA 1758	6	MS	5	VT	
	Baronesse	6	MS	9	Т	
	Champion	6	MR	7	Т	Х
	Sawtooth	6	MS	16	MI	
	Harriman	7	MS	12	MT	
	Clearwater	7	MS	23	MI	
	CDC McGwire	9	MS	12	MT	
	Vespa	10	MS	3	VT	
	Tetonia	13	S	5	VT	
	Spaulding	15	S	12	MT	
6-row feed barley	Millennium	5	MR	3	VT	Х
	Steptoe	5	MR	17	MI	
	Goldeneye	6	MR	14	MT	Х
	Herald	11	MS	6	Т	

Table 4. Evaluation of 20 spring feed barley varieties for resistance and tolerance to *Heterodera avenae*; data are means of field screening trials during two years on a farm in southeast Idaho, using four replicates per entry on each of two treatments (with or without application of a nematicide).

 $^{\rm 1-5}$ Footnotes are described under Tables 2 and 3.



Figure 15. Number of *Heterodera avenae* white swollen females produced on 20 spring wheat varieties tested in two experiments near St. Anthony, Idaho and Cashup, Washington. Varieties with * have significantly fewer white females than the check variety (Louise). Varieties with ** have significantly greater numbers of white females than Louise. Ouyen carries the *Cre1* resistance gene, but the source of resistance in WB-Rockland has not yet been identified.

Chart by Dr. Richard Smiley, © Oregon State University.

Market class	Variety	White females ¹	Resistance rating ²	Yield increase (%) ³	Tolerance rating⁴	MR + MT⁵
2-row	Odyssey	1	VR	16	MI	
malt barley	LCS1820	8	MS	9	Т	
	Conrad	10	MS	17	MI	
	2Ab07-X031098-31	11	MS	6	Т	
	ABI Balster	12	S	2	VT	
	Merem	12	MS	6	Т	
	Meredith	14	S	14	MT	
	Merit	15	S	4	VT	
	Overture	17	S	9	Т	
	Metcalf	17	S	17	MI	
	Copeland	19	S	4	VT	
	Pinnacle	19	S	7	Ţ	
	Harrington	21	S	10	MT	
	2Ab04-X001084-27	22	S	3	VT	
	Hockett	24	S	17	MI	
	B1202	24	S	30	I	
	Merit 57	26	VS	7	Т	
	LCS Genie	34	VS	11	MT	
	ABI Voyager	39	VS	19	MI	
6-row	Legacy	4	MR	27	MI	
malt barley	Quest	6	MS	3	VT	
	Tradition	6	MR	21	MI	
	Celebration	6	MS	31	I	
	Menan	8	MS	1	VT	
	Morex	10	MS	18	МІ	

Table 5. Evaluation of 25 spring malt barley varieties for resistance and tolerance to *Heterodera avenae*; data are means of field screening trials during two years on a farm in southeast Idaho, using four replicates per entry on each of two treatments (with or without application of a nematicide).

^{1–5} Footnotes are described under Tables 2 and 3.

Table 6. Reproduction of *Heterodera avenae* populations from western (Hillsboro) and eastern (Union) Oregon on international indexing entries of barley, oat, and wheat, showing that resistance to these populations was conferred to barley by the *Rha2* and *Rha3* resistance genes, to oat by multiple or unknown genes in two entries, and to wheat by the *Cre1* resistance gene, with additional partial resistance by *Cre5*.

Crop	Test entry and [resistance gene, if any]	White females ¹	Resistance rating ²
Barley	Martin 403-2 [<i>Rha2</i>]	0	VR
	Morocco [<i>Rha3</i>]	0–1	VR
	Ortolan [<i>Rha1</i>] ³	10–14	S
	Emir [<i>RhaE</i>] ³	11–27	S
	Varde	22–45	VS
Oat	l376 [multiple genes]	0–1	VR
	IGV.H.72-646 [unknown]	0–1	VR
	Sun II [multiple genes] ³	20–43	VS
	Nidar II	58–73	VS
Wheat	AUS10894 [Cre1]	0–1	VR
	Loros [Cre1]	0–1	VR
	Ouyen [<i>Cre1</i>]	4	MR
	VPM1 [Cre5]	12	MS
	T-2003 [Cre7] ³	17	S
	Stephens	29	VS
	Frame [<i>Cre8</i>] ³	31	VS
	Zak	39	VS
	ID-2150 [Cre2] 3	48	VS
	VL125 [Cre3] ³	65	VS

¹ Average number of *H. avenae* white females produced on roots of each plant.

² Resistance ratings are shown under Table 1.

³ The designated resistance genes are very effective for restricting reproduction of certain populations of *H. avenae* elsewhere in the world but are shown here to be ineffective against the populations of this nematode in Oregon.

Timing of planting. Planting winter wheat rather than a spring crop of wheat, barley, or oat can favor strong, deep root development before the majority of second-stage juveniles emerge from cysts during the spring. Also, where sufficient water is available, planting a susceptible host as a trap crop during the fall or early spring can reduce cereal cyst nematode densities in soil. The trap crop is invaded when second-stage juveniles migrate from the cyst into the soil during early spring. The trap crop is then killed during mid-spring before new egg-bearing cysts can be developed. This strategy is particularly useful where growers plan to produce a warm-season crop such as chickpea or bean that can be planted during late spring after the trap crop has been killed in an infested field that will be planted to wheat or barley the following year.

Crop nutrition and water supply. Since the greatest crop loss occurs when nutrients or water become limiting for maximum plant growth at any stage, supplying optimal plant nutrition and, where possible, supplemental water during intervals of drought can minimize (mask) crop damage, particularly when the nematode damage is only slight or moderate. However, crops that are severely damaged by cereal cyst nematodes usually do not respond well to additional applications of nutrients or water. If severe damage becomes evident early enough in the spring, it may be more profitable to destroy the crop and plant a non-host (broadleaf) crop in its place.

Biofumigation. Some green manure crops such as brown mustard, rapeseed, or sudangrass can be used as biofumigants to reduce numbers of soilborne plant-pathogenic fungi, plant-parasitic nematodes, root-feeding insects, and weed seeds. These crops are used mostly where water is not a limiting factor for wheat growth. When green tissue from a biofumigant crop is thoroughly macerated and immediately incorporated into soil, the toxic products from the degradation of that tissue may reduce the nematode density in soil. It is important to realize, however, that some of the crops used as biofumigants may increase the density of root-lesion nematodes (Pratylenchus species), particularly when those crops are grown to maturity for the harvest of seed. Incorporation of mature stalks does not initiate the biofumigation process. If, however, young plants susceptible to root-lesion nematodes are macerated and incorporated into soil, any nematodes that multiplied on the biofumigant crop are killed in the same manner as weed seeds and other nematodes and soil-dwelling organisms. After rototilling a green manure crop into the soil to serve as a biofumigant crop, it is important to wait several weeks before planting another crop because the biofumigant action may cause phytotoxicity to seedlings if the seed is planted too quickly.

Nematicides. Pre-plant nematicides are registered for suppressing nematode populations in fields that will be planted to many commercial crops. These nematicides are mostly fumigants that must be shanked deeply into soil, after which the soil surface is sealed by tilling, applying water, or covering it with a plastic barrier tarp. Most fumigants are restricted to application in soils having few or no clods, little or no surface residues from previous crops, and a specific range of temperature and moisture. Further restrictions apply to weather conditions involving atmospheric inversion or significant rainfall forecasted within a given number of days. Most of these conditions are far too restrictive for applying pre-plant fumigants to large acreages of small grain crops, particularly in rainfed settings where supplemental irrigation is not possible. Also, the high expense of pre-plant fumigants usually restricts their use to crops that are of a higher value than small grain crops. However, when used on high-value crops that are rotated with small grains, the application will likely also reduce the density of cereal cyst nematodes and thereby provide partial protection for subsequent crops of wheat or barley.

Non-fumigant, in-crop nematicides are effective and widely used in research to demonstrate yield constraints caused by cereal cyst nematodes (Figures 13 and 14, page 9). However, this use of nematicides is not permitted for use in commercial crops, meaning that all grain from research trials treated with a non-registered pesticide must be destroyed.

Biological agents. Applications of currently available biological nematicides have not been effective for increasing the productivity of wheat in the PNW. However, in some locations, naturally occurring fungal or bacterial parasites invade and kill some of the *Heterodera* eggs that are still inside the cyst. These natural parasites of eggs reduce the density of *Heterodera*, but even in fields where they are known to be present and active, like in many eastern Oregon fields, reduced yields of wheat and barley continue to occur. Ways to amplify the benefit of these natural biological agents in commercial agriculture have not been identified.

Tillage. Tillage does not have an appreciable effect on the density of *Heterodera* species. Populations are likely to be similar in both cultivated and no-till fields.

Sampling and Identification

Nematode detection and identification requires the services of a professional nematologist. Population densities of cereal cyst nematodes are determined by extracting eggs and juveniles still confined within cysts, and by using a second extraction procedure that can detect juveniles that may have already emerged from cysts and are now free within the soil matrix. Since samples taken during the spring and early summer include both life stages, both types of extraction are required to determine the density of cereal cyst nematodes. Samples taken from the time of harvest through fall, particularly when the soil is dry, contain only eggs and juveniles still inside a cyst. Samples collected during the mid-summer through fall require only one extraction procedure and are less expensive to process.

Two commercial and two university labs that provide nematode testing services in the PNW are listed under "Nematode Testing Labs" (page 17). Both commercial labs provide a courier service to transport samples from many locations throughout the region. Testing labs provide specific guidance as to how they wish to have nematode samples packaged and shipped, which may vary slightly depending on the lab. It is strongly recommended that you contact the lab of your choice in advance for their specific sampling and handling instructions. For this reason, only general guidelines are provided in this bulletin.

Sampling. Collect soil samples for cereal cyst nematodes to a depth of 12 inches. Most cysts are located in the upper 4 to 6 inches of soil. Detection of these parasites is often more successful and less expensive when samples are collected after plant maturation or harvest, when the soil is dry and there are no juveniles present in it. It is also generally more effective to collect soil samples from specific areas where patches of stunted plants occurred during crop growth rather than sampling randomly or in a grid-sampling pattern across entire fields, as is typically done for assays of soil fertility.

To increase the reliability of the test result, at least 20 soil cores must be collected and composited into a single sample for testing. Soil can be collected from the field in a bag or bucket and then mixed very well before transferring some of it into a standard soil-sampling bag for delivery to the lab. Samples collected with a shovel are generally not adequate because they typically include much more soil from near the surface than from the deepest part of the 'shovel slice,' making such samples nonrepresentative of the soil profile. Collect and handle samples carefully because nematodes can be killed by improper handling, such as leaving samples in direct sunlight or in a car trunk on a hot day.

Identification. Distinguishing between *H. avenae* and *H. filipjevi* and species of *Heterodera* that occur on other crops, such as pea or sugar beet, is an essential prerequisite for using a control tactic based on the selection of a resistant or tolerant plant variety. However, identification is difficult because there are few morphological characteristics of taxonomic value that are clearly visible, even with the aid of a standard laboratory microscope (Figure 4, page 3). Identification of these species using traditional methods is therefore slow and often prone to error if the diagnostician lacks experience in differentiating *Heterodera* species. Consequently, commercial diagnostic labs usually do not attempt to differentiate individual species as a regular component of the testing service.

For this reason, molecular procedures were developed in our laboratory and are now available to precisely differentiate and quantify individual species of cereal cyst nematodes using a single DNA extract from soil or from an individual egg, juvenile, or cyst that has been extracted from soil. Our speciesspecific PCR (polymerase chain reaction) assays (Figure 7, page 5) are being used routinely in many nematology laboratories. Western Laboratories, one of the commercial nematology labs in the following list, now also provides this DNA-based diagnostic procedure to differentiate species such as H. avenae and H. filipjevi. Additionally, that lab uses other DNA-based tests to identify many species of root-infecting fungal pathogens and root-lesion nematodes, all of which are important on small grain crops in the PNW. A more precise PCR-RFLP (polymerase chain reaction - restriction fragment length polymorphism) assay (Figure 8, page 5) developed in our laboratory is now also being used in many research laboratories to distinguish among Heterodera species.

Nematode Testing Labs

- Kuo Testing Labs (2 locations): 1300 6th Street, Umatilla, Oregon 97882 and 337 South 1st Avenue, Othello, Washington 99344. 800-328-0112. <u>http://kuotesting.com</u>
- 2. OSU Nematode Testing Service, 1089 Cordley Hall, Corvallis, Oregon 97331. 541-737-5255. <u>http://plant-clinic.bpp.oregonstate.edu/</u> <u>nematodes</u>
- University of Idaho, Parma Research and Extension Center, Parma, Idaho 83660.
 208-722-6701. <u>http://extension.uidaho.edu/</u> parma/tag/nematology/
- Western Laboratories, 211 Highway 95, Parma, Idaho 83660. 208-649-4360. <u>http://www.westernlaboratories.com</u> (services include DNA-based species identification)
- Agnema Analytical Laboratories, 350 Hills Street, Suite 103, Richland, Washington 99354. 509-255-3744. <u>http://agnema.com</u>

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