Identifying and Managing Root Rot of Pulses on the Northern Great Plains

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Pulse crops have been an important component of the cropping rotation across the Canadian prairies for more than 30 years and are an increasing component of the cropping system in many regions of the northern Great Plains of the United States, with an estimated farm-gate value of \$3 billion per year (Table 1). Pulses are annual dry grain legume crops that fix atmospheric nitrogen, are drought- and heat-tolerant, and have a positive impact on the yield of subsequent crops through changes in soil microbial communities and soil nutrient levels. The main pulse crops produced in the northern Great Plains are field pea (*Pisum sativum* L.), lentil (*Lens culinaris* Medikus), dry bean (*Phaseolus vulgaris* L.), and small acreages of chickpea (*Cicer arietinum* L.), faba bean (*Vicia faba* L.), and lupine (*Lupinus angustifolius* L.). Canada is the world's largest exporter of lentil (Risula 2010) and dry pea (Saskatchewan Ministry of Agriculture 2012). In the United States, production of dry bean far exceeds that of the other pulse crops. Over the last 20 years, the acreage of the major pulse crops in Canada has increased slowly, but production of field pea and lentil in the United States has increased dramatically since the late 1990s (Table 1).

The root rot disease complex of pulse crops on the northern Great Plains consists of *Fusarium* spp., *Pythium* spp., *Rhizoctonia solani* Kühn (teleomorph, *Thanatephorus cucumeris* (Frank) Donk), and *Aphanomyces euteiches* Drechs. This complex causes damping-off, seedling blight, root rot, and reduces stand establishment, nitrogen fixation (Hwang et al. 2001), root distribution, and root vigor. Uneven plant stands resulting from poor germination and seedling

blight frequently result in subsequent difficulty in managing weeds in those areas (Hwang et al. 2007). The root rot complex has become an important constraint to pulse production in the northern Great Plains, but root symptoms are often overlooked and their importance underestimated. In recent years, there has been a substantial research effort in North America to expand our understanding of this disease complex and to improve management recommendations.

Root diseases are generally most severe where susceptible crops are grown in short cropping rotation, because pathogen inoculum can build up quickly when environmental conditions are conducive for disease development (Abawi and Ludwig 2005; Estévez de Jensen et al. 2002). Yield loss is often difficult to assess, in large part because roots are rarely examined unless there is a substantial impact on top-growth, and even severe symptoms are often under-reported or go unnoticed because affected plants are unevenly distributed within a field (Moussart et al. 2009; Navarro et al. 2008). Damping-off and seedling blight can result in thin, uneven plant stands. Plant compensation can counteract losses in stand caused by seedling blight (van Bruggen et al. 1986), but weed management issues and uneven plant development (Hwang et al. 2007) associated with a thin, patchy crop can affect harvestability and quality even when total yield is not affected.

The life cycles of the pathogens in the root rot complex are similar (Fig. 1). Primary inoculum, which consists of resistant resting spores or sclerotia in soil and/or crop residue, germinates in the presence of seed or root exudates and infects seedling roots. The pathogen proliferates in the root system of infected plants, spreads from plant to plant in the soil via secondary spores or mycelium (Fig. 2), and produces new resting spores or sclerotia at the end of the season. Infested soil and organic matter can be spread by wind and water (Palmero et al. 2011), farming equipment, and many other human activities.

In the early years of pulse crop introductions on the Canadian prairies, root rot was not an issue in most fields (B. D. Gossen, unpublished data). However, over the last 15 years, there have been numerous observations of pulse fields with severe root rot (e.g.,

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Chang et al. 2003, 2004a, 2007), and increasing numbers of experienced pulse producers have reported unexpectedly low yields in and around the main centers of pulse production. At the same time, root rot has become ubiquitous across the region, present at damaging levels in almost every field each year (e.g., McLaren et al. 2013, 2015). This indicates that the root rot complex has become an important constraint to pulse crop production throughout the northern Great Plains. To date, no strong sources of resistance to the components of the root rot complex have been identified. Although some sources of partial

resistance (reduced disease intensity relative to susceptible lines) or tolerance (maintains yield despite symptom development) have been identified, commercial cultivars with useful levels of resistance are not yet available.

Until recently, the identity of root rot pathogens was determined by associating symptoms in host plants with the results of inoculation studies. The importance of specific pathogens was based on culturing infected roots and identifying the isolates growing from infected samples based on morphological characteristics. This approach is

Table 1. Estimates of the hectareage and value of selected pulse crops in Canada and the United States at about 5 year intervals since 1991^a

Crop	Canada					United States				
	1991	1996	2001	2006	2011	1991	1996	2001	2006	2010
Area of production (K ha)										
Pea	201	536	1,340	1,264	976	76	83	78	358	288
Bean	92	91	181	185	98	775	709	506	620	746
Lentil	239	303	704	522	1,044	52	60	81	174	257
Chickpea	0	0	482	130	40	1.4	1.6	52	55	58
Value of Production (\$M)										
Pea	\$47	\$220	\$304	\$367	\$842	\$29	\$30	\$21	\$86	\$139
Bean	\$51	\$64	\$101	\$153	\$120	\$528	\$652	\$427	\$554	\$899
Lentil	\$78	\$135	\$178	\$182	\$665	\$26	\$23	\$28	\$40	\$219
Chickpea	0	0	\$149	\$78	\$54	0	\$15	\$26	\$39	\$56

^a Sources: Statistics Canada, Census of Agriculture, 1991, 1996, 2001, 2006, and 2011, and USDA, Economic Research Service, Vegetables and Pulses Yearbook, 1991, 1996, 2001, 2006, 2011. N.B. Production values reported in \$US for the United States and \$CDN for Canada.

Fig. 1. A generalized life cycle diagram for the dominant root rot pathogens of pulse crops on the northern Great Plains of North America.

Fig. 2. Spreading patch of stunted and yellowing field pea plants prior to flowering, caused by severe root rot near Drumheller, Alberta, Canada.

inherently problematic because i) it excludes fastidious organisms, ii) it tends to favor pathogen species that grow quickly on media and underestimates slow-growing pathogens, iii) it may be adversely affected by other microorganisms, iv) species identification based on morphology in culture often requires specialized expertise, and v) the studies are time consuming and resource intensive. Molecular techniques have largely replaced culturing approaches for pathogen identification and quantification, and have the potential to provide valuable insights into the population dynamics of a wide range of soil microbes, including plant pathogens.

The purpose of this review is to provide a brief description of the important pathogens that make up the root rot complex on pulses, to evaluate strategies for root rot management across the region, and highlight recent contributions to our understanding of the root rot complex on pulses on the northern Great Plains of North America.

Components of the Root Rot Complex

***Fusarium* spp.** *Symptoms, life cycle, and impact.* Symptoms include reddish-brown to blackish brown lesions on roots (Fig. 3) and a red discoloration of the root vascular system (Chang et al. 2005). The taproot may remain discolored but intact, but fine roots can be completely destroyed, resulting in fewer nodules (Hwang et al. 1994, 1995). Proliferation of adventitious roots can occur when the taproot is severely damaged. When soil moisture is high, conidia may form near the soil surface on infected plants. As infected tissues disintegrate, hyphae and conidia are converted to thick-walled, long-lived chlamydospores (Nash et al. 1961). Losses in yield can be up to 60% loss in field pea (Chang et al. 2004b, 2005) and 84% in dry bean (Knodel et al. 2007; Steadman et al. 1975).

Some pathogenic *Fusarium* spp. are generalists, while others are highly host specific (Gordon and Martyn 1997). For example, *F. avenaceum* is a pathogen of all of the most common crops (cereals, canola, pulses) grown on the northern Great Plains (Abdellatif et al. 2010; Fernandez et al. 2008; Hwang et al. 2000b; Satyaprasad et al. 2000). In contrast, *F. solani* f. sp. *pisi* from pea and f. sp. *phaseoli* from bean cause severe disease on the primary host but only mild necrosis on the other crop (Reinking 1950; Suga et al. 2002).

Fusarium spp. are the predominant root rot pathogens on pulse crops in the region. On dry bean, *F. solani* f. sp. *phaseoli* was highly destructive (Conner et al. 2014; Goswami et al. 2010; Henriquez et al. 2014, 2015). On field pea, *F. avenaceum* was the species most frequently isolated in western Canada (Chang et al. 2004a, 2007, 2013; Chatterton et al. 2015b; McLaren et al. 2013, 2015), and *F. oxysporum* and *F. avenaceum* predominated in North Dakota (Chittam 2012; Chittam et al. 2015). On chickpea, *F. solani* and *F. redolens* were most common (Chang et al. 2003; Esmaeili Taheri et al. 2011; Harveson 2011), and *F. avenaceum* was predominant on lentil (Bailey et al. 2000; Hwang et al. 1994; Lin and Cook 1977). Also, *F. graminearum* has been associated with root rot of several pulse crops (Bilgi et al. 2011; Esmaeili Taheri et al. 2011; Chongo et al. 2001).

Favorable conditions. *Fusarium* spp. are generally weakly aggressive or opportunistic pathogens on pulse crops, requiring some type of plant stress to cause severe infections (Leslie et al. 1990). Conditions that produce plant stress, such as soil compaction, high soil temperatures, extreme soil moisture levels, and flooding/drought, favor disease development (Estévez de Jensen et al. 2004; Harveson et al. 2005; Tu 1994; Tu and Tan 1991). *Fusarium* spp. often overwinter in and on crop residues, and so become more important under tillage systems that increase retention of residues on the soil surface (Fernandez et al. 2008; Larkin 2015). The presence of infected crop residue is especially important for survival of *F. avenaceum*, which does not produce chlamydospores (Leslie and Summerell 2006). Severity of root rot caused by many *Fusarium* species and subsequent increased pathogen inoculum in the soil is highest in warm, moist soil (Hall and Phillips 1992). For example, the most severe symptoms of *F. avenaceum* develop between 25 and 30°C (Hwang et al. 2000b; Tu 1994).

Identification and quantification. Until recently, identification of *Fusarium* spp. was based primarily on morphological characteristics

on specialized media (Leslie and Summerell 2006). However, high variability among isolates resulted in reclassification of many isolates and even species over time as a result of misidentification (Kristensen et al. 2005; Leslie and Summerell 2006). For example, phylogenetic studies of *F. solani* have identified it as a complex consisting of three groups (O'Donnell et al. 2008; Zhang et al. 2006).

The translation elongation factor 1 alpha (EF1- α) gene (Geiser et al. 2004) has become the preferred region of the pathogen genome for isolate identification, but the internal transcribed spacer (ITS) region (O'Donnell et al. 1998) and the ribosomal intergenic spacer (IGS) (Appel and Gordon 1995) have also been utilized. These techniques have been used to identify *F. redolens* from chickpea, pea, and lentil (Esmaeili Taheri et al. 2011) and *F. cuneirostrum* from dry bean (Henriquez et al. 2014). Real-time PCR for quantification of *Fusarium* spp. has been developed for cereal crops (Atoui et al. 2012; Preiser et al. 2015; Scaufflaire et al. 2012) and primers and probes for the rapid detection and quantification of *F. oxysporum* f. sp. *phaseoli* are available (de Sousa et al. 2015). Also, multilocus genotyping assays (MLGT) have been used to identify *F. cuneirostrum* (O'Donnell et al. 2010) and members of the *F. graminearum* complex (Ward et al. 2008). Molecular techniques to quantify *Fusarium* spp. from pulse roots have been developed recently (S. Chatterton, unpublished data; D. L. McLaren, unpublished data).

***Pythium* spp.** *Symptoms, life cycle, and impact.* *Pythium* spp. infect seeds or seedlings before emergence, causing pre-emergence damping-off (Hendrix and Campbell 1973), or infect the seedling root and hypocotyl, causing post-emergence damping-off (Fig. 4). Seedlings may survive root infection after emergence (Larkin et al. 1996; Mellano et al. 1970), but typically exhibit reduced vigor and growth (Hancock 1991; Hodges and Coleman 1985). As a result, moderate to severe disease pressure often results in patchy plant

Fig. 3. Root rot symptoms on field pea caused by *Fusarium* spp.

stands. Seed vigor affects host susceptibility because leakage of solutes from seed with low vigor increases seed infection (Hwang et al. 2001). *Pythium* spp. also can infect the roots of mature plants, causing necrotic lesions on root tips or fine feeder roots and (less commonly) on tap roots that reduce plant growth and yield even when necrotic symptoms are not present (Lee and Hoy 1992; Stanghellini and Kronland 1986).

Pythium spp. can overwinter as hyphae or sporangia, and can survive for many years of unfavorable conditions as oospores (Allen et al. 2004; Hendrix and Campbell 1973; Onokpise et al. 1999). Under favorable conditions, oospores germinate directly via a germ tube that may infect the plant or form sporangia. Sporangia formed on plant tissue can germinate in the same fashion as the oospores, either directly or by forming zoospores. The sporangia produce flagellated zoospores that swim for a short time before encysting on host roots and forming a germ tube.

Pythium spp. are important root pathogens of pulse crops, with estimates of losses as high as 50% (Hwang et al. 2000a). Some species have a wide host range, while others are narrow (Abad et al. 1994; Chamswang and Cook 1985; Hancock 1985; Ingram and Cook 1990; Lee and Hoy 1992; Moulin et al. 1994). Where detailed assessments have been made, large numbers of *Pythium* spp. are often associated with root rot on pulses (Li et al. 2014; Matoba et al. 2008; Nzungize et al. 2012; Zitnick-Anderson and Nelson 2015), but a much smaller number of species generally predominate. For example, *P. ultimum* Trow and *P. irregulare* Buisman are the principal species causing seed rot and damping-off of field pea on the Canadian prairies (Hwang and Chang 1989).

Favorable conditions. *Pythium* spp. are found in wet or moist environments, including soils, marshes, and in water (Van der Plaats-Niterink 1981). They are most damaging when soils are waterlogged, where large numbers of motile zoospores are produced and released. Even one day of waterlogging can substantially enhance root infection, damping off, and subsequent root rot development (Li et al. 2015; Yanar et al. 1997). Soil temperature can affect spore germination, germ tube growth, and zoospore discharge (Tedla and Stanghellini 1992), but optimum temperatures can be species-specific. For example, *P. ultimum* and *P. dissotocum* (Drechs.) thrived as saprophytes on crop residues in cool (10 to 15°C) soils, but other species were favored by warmer (25 to 36°C) soil (Owen-Going et al. 2008).

Acidic soil pH can reduce the formation of oospores and sporangia, so populations of *Pythium* spp. tend to be higher at pH ≥ 7.0

(Lumsden et al. 1987; Martin and Loper 1999) and were more prevalent in cultivated than uncultivated soil (Hendrix et al. 1971).

Identification and quantification. Presence/absence, morphology, and size of sporangia, oogonia, and antheridia are the main morphological criteria used for species identification in *Pythium* (Matsumoto et al. 1999). However, these characteristics vary with cultural conditions and growth media (Dick 2001), making identification problematic (Kageyama et al. 2005; Uzuhashi et al. 2010). Dilution plating onto a selective medium has been used in studies of population dynamics (Ali-Shtayeh et al. 1986; Conway 1985; Mircetich and Kraft 1973). These methods have been largely replaced by DNA barcoding with cytochrome c oxidase subunit I (COI) from mitochondria (Hajibabaei et al. 2006; Seifert et al. 2007; Ward et al. 2005), which is more reliable than internal transcribed spacer (ITS) sequences for species identification (Robideau et al. 2011). Other molecular approaches include PCR ELISA (Bailey et al. 2002), sequencing (Martin 2000; Matsumoto et al. 1999), and qPCR for pathogen quantification (Bates et al. 2001; Schroeder et al. 2006).

***Rhizoctonia solani*. Symptoms, life cycle, and impact.** Multinucleate isolates of *R. solani* have been divided into 14 anastomosis groups (AG) that are considered to be isolated, non-interbreeding populations (Anderson 1982; Tewoldemedhin et al. 2006). AG-4 is the most important group on pulses on the northern Great Plains (Conner et al. 2014; Hwang et al. 2003, 2007; Mathew et al. 2012; Sharma-Poudyal et al. 2015; Zhou et al. 2009), but AG-2-2 is also highly pathogenic on dry bean and faba bean (Engelkes and Windels 1996) and AG-2-1 reduced seeding emergence and caused severe root rot on pea (Sharma-Poudyal et al. 2015).

The pathogen can survive as hyphae in infected crop debris, as sclerotia in the soil (Hanson 2005; Howard et al. 1994; Porter et al. 2011), and thick-walled hyphae can function as chlamydospores (McCoy and Kraft 1984a; Porter et al. 2011). It can also persist in soil as a saprophyte (Papavizas et al. 1975). It can be spread by blowing infested soil or crop debris (Dillard 2001), and can even be transmitted, to a limited extent, via infected seed (Howard et al. 1994; Porter et al. 2011). There are conflicting reports on the importance of the role of basidiospores of *Thanatephorus cucumeris* (teleomorph of *R. solani*) in dispersal of the pathogen (Hanson 2005; Howard et al. 1994; Mordue 1974; Ogoshi 1987; Porter et al. 2011).

Seedling infection often results in seed rot, damping-off, and seedling blight (Kaiser and Horner 1980). Infection of older plants results in sunken, reddish-brown, penetrating lesions on tap roots,

Fig. 4. Root rot symptoms caused by *Pythium* spp. on seed, germinating seed and young field pea seedlings (healthy plant on left).

hypocotyls, epicotyls, and stem bases (Hanson 2005; Howard et al. 1994), soft rot of stems and roots in pea and faba bean (Lamari and Bernier 1985; Hwang et al. 2007; McCoy and Kraft 1984a, b; Shehata et al. 1981, 1984), stunting, wilting, and girdling of the stem in pea (Navarro et al. 2008; Porter et al. 2011). Severe symptoms on the roots can also stimulate the formation of adventitious roots close to the soil surface (O'Brien et al. 1991) and reduce root nodulation (Chang et al. 2008). Small, spherical or irregularly-shaped, brown to black sclerotia can develop in old lesions and inside infected stems (Howard et al. 1994; Porter et al. 2011).

Losses in yield in inoculated trials have been as high as 79% in field pea (Hwang et al. 2007), 88% in dry bean (Burke and Kraft 1974; Tan and Tu 1995), 70% in chickpea (Chang et al. 2004b), 69% in lentil (Chang et al. 2008; Hwang et al. 2003), and 19% in faba bean (Chang et al. 2014).

Favorable conditions. Cropping history and environmental conditions strongly influence the incidence and severity of root rot. Hyphae spread most rapidly at 24 to 30°C, temperatures above 17.5°C favored symptom development; and severity increased with increased inoculum concentration (Dillard 2001; Hwang et al. 2007). Late seeding into warm soil resulted in the greatest reduction in seedling establishment (Chang et al. 2004b, 2008; Hwang et al. 2007). Deep seeding (5-cm versus 2-cm depth) also resulted in reduced seedling emergence of field pea and chickpea, but seeding depth had no effect on lentil. Finally, poor soil conditions, such as low fertility (Howard et al. 1994), compaction (Tan and Tu 1995; Tu and Tan 1991), and poor drainage (O'Brien et al. 1991) predispose pulses to *Rhizoctonia* seedling blight and root rot (Harveson et al. 2005).

Identification and quantification. *R. solani* produces broad (8 to 12 µm), multinucleate, hyaline hyphae that typically branch at right angles and anastomose frequently. They darken with age, lack clamp connections, and have conspicuous dolipore septa (Barron 1977; Mordue 1974; Ogoshi 1987). The pathogen produces no asexual spores, but forms irregular-shaped, spongy, brown to black sclerotia (Barnett and Hunter 1972; Barron 1977).

Baiting methods and selective media have been developed for detection and quantification of inoculum (Ko and Hora 1971; Trujillo et al. 1987; Vincelli and Beaupré 1989), but these methods are labor intensive and require considerable expertise (Thornton et al. 1999). Pectic enzyme pattern have also been used to examine intraspecific groups (Brisbane et al. 1995). In recent years, molecular approaches have been used for detection, diagnosis, and identification of AGs (Dubey et al. 2014; Mathew et al. 2012; Sharma-Poudyal et al. 2015;

Zhou et al. 2009). Real-time PCR has been used for the quantification of inoculum in other crops (Abbas et al. 2014; Okubara et al. 2008), but its use has not yet been reported in pulse crops.

***Aphanomyces euteiches*. Symptoms, life cycle, and impact.** *Aphanomyces euteiches* is widespread in North America, Europe, Japan, Australia, and New Zealand (Hughes and Grau 2013; Papavizas and Ayers 1974; Wicker et al. 2001). Its host range includes dry bean, field pea, lentil, faba bean, and various forage legumes. Soybean, chickpea, and some faba bean lines are resistant (Malvick et al. 2009; Moussart et al. 2008, 2013; Vandemark and Porter 2010). Until recently, *A. euteiches* was not recognized as a pathogen of concern on pulses in most parts of the northern Great Plains. That changed when molecular methods were used to assess pathogen prevalence in infected roots (Banniza et al. 2013), and it is now considered to be widespread across the region (Chatterton et al. 2014, 2015a, b; Zitnick-Anderson and Pasche 2016).

Infected pea root tissues exhibit a water-soaked, honey-brown discoloration that eventually extends throughout the root system and into the epicotyl (Fig. 5). Oospores are visible under moderate magnification in the root cortex and in the hypocotyl or epicotyl (Hughes and Grau 2013). Infected tissues often turn black as a result of colonization by other soilborne fungi (Papavizas and Ayers 1974). In advanced stages, the entire root mass can be decayed, with only strings of vascular bundles remaining. Shoot symptoms (chlorosis, wilting, and eventually death) only become obvious when root symptoms are advanced.

Oospores can remain dormant in infested soils for many years (Papavizas and Ayers 1974; Hughes and Grau 2013). In the presence of a host, they germinate to produce sporangia that in turn release hundreds of biflagellate zoospores (Shang et al. 2000). Zoospores can swim short distances, guided by chemical signals released from host roots. They rapidly attach to roots, encyst, germinate, and penetrate cells of the root cortex. Hyphae grow inter- and intracellularly throughout the cortex, and differentiate to form haploid antheridia and oogonia (Papavizas and Ayers 1974). Antheridia penetrate and fertilize the oogonia, resulting in formation of diploid oospores.

Favorable conditions. The pathogen is favored by high soil moisture, associated with poor drainage, heavy clay soils, and soil compaction (Grath and Hakansson 1994; Hossain et al. 2012). Zoospores are released when soil water potential is between -2.5 and -1.0 kPa, or close to saturation (0 kPa) as this results in water-filled pores of at least 200 µm diameter that are required for zoospore motility (Allmaras et al. 2003). Conducive soils had 35 to 40% clay content, and so were

Fig. 5. Root rot symptoms (healthy plant on left) of field pea caused by *Aphanomyces euteiches* (A) and oospores (B) in infested roots.

prone to compaction and low water permeability. Severity decreased with increasing soil calcium concentrations in greenhouse studies, which was attributed to inhibition of zoospore production (Heyman et al. 2007). Two types of suppressive soils have been reported; one with high sand content, low clay content, and higher than average organic carbon content, and another with high clay content and high calcium and pH levels (Persson and Olsson 2000).

Short cropping rotations with susceptible species rapidly increased the inoculum potential in a field, whereas inclusion of nonhost species and partially resistant cultivars reduced inoculum levels over time (Moussart et al. 2013; Oyarzun et al. 1993; Papavizas and Ayers 1974; Pfender and Hagedorn 1983). The estimated half-life of *A. euteiches* inoculum was 1 year, so many years without pea production may be required to reduce even moderate levels of inoculum below yield-reducing levels (Moussart et al. 2009; Pfender and Hagedorn 1983). Severity increased in the presence of other root rot pathogens (Peters and Grau 2002; Xue 2003b).

Identification and quantification. The pathogen is difficult to isolate directly from symptomatic roots (Vandemark et al. 2000), but it can be recovered using semiselective agar (Pfender et al. 1984). It has traditionally been identified by the presence of sporangia bearing primary spores that are loosely clustered on sporangial tips, which give rise to biflagellate zoospores. Dilution plating to quantify oospore levels in soil is ineffective due to inconsistent germination in vitro and the lack of a highly selective media for *A. euteiches* (Malvick et al. 1994). Direct quantification of oospores is time consuming and labor intensive (Chan and Close 1987; Papavizas and Ayers 1974). Therefore, quantification of inoculum has been primarily based on soil indexing, using a bioassay with a susceptible host grown in field soil (Chan and Close 1987; Malvick et al. 1994; Oyarzun 1993).

Molecular tests have been developed to identify (Vandemark and Grünwald 2005; Vandemark et al. 2000) and quantify *A. euteiches* in soil, with a detection threshold of 10 oospores/g dry soil (Gangneux et al. 2014; Sauvage et al. 2007).

Management

***Fusarium* spp. Seed treatment.** Seed treatments with synthetic fungicides represent a consistent and effective management option for *Fusarium* seedling blight of pulse crops (Broders et al. 2007; Chang et al. 2013; Hwang et al. 2000b; Munkvold and O'Mara 2002), but generally have little effect on root rot. Biocontrol seed treatments with activity against *Fusarium* root rot have been identified, and several have shown some promise (Estévez de Jensen et al. 2002, 2004; Wang et al. 2003; Xue 2003a, b), but none have been widely adopted.

Cultural practices. On lentil, seeding in mid-May generally improved seedling establishment compared with early or late May planting dates in trials inoculated with *F. avenaceum*, even though the most severe root rot symptoms occurred at temperatures between 20 and 27.5°C under controlled conditions (Hwang et al. 2000b). Seeding date did not affect emergence or yield of field pea inoculated with *F. avenaceum* (Chang et al. 2013).

Extended crop rotation and minimal tillage have been recommended for maintaining soil health and reducing the effects of soilborne pathogens (Larkin 2015). However, moldboard tillage increased yield of dry bean compared with no-till, likely as a result of burial and rapid breakdown of infested crop residues (Estévez de Jensen et al. 2004). Similarly, zone tillage reduced root rot in compacted soils, and also minimized soil compaction and loss of soil moisture (Harveson et al. 2005). On field pea, fields with an extended rotation interval (1 in 4 to 5 years) experienced severe root rot injury associated with *F. solani* f. sp. *pisi* and *F. avenaceum* (Chatterton et al. 2014, 2015b; S. Chatterton, unpublished data), which indicated that rotation was not effective in reducing *Fusarium* root rot on pulses on the northern Great Plains.

Resistance. On dry bean, no source of complete resistance to *F. solani* f. sp. *phaseoli* has been identified, but partial resistance has been reported (Bilgi et al. 2008; Conner et al. 2014; Nicoli et al. 2011, 2012; Ronquillo-Lopez et al. 2010).

In field pea, four quantitative trait loci associated with resistance to *F. avenaceum* have been identified (Li et al. 2012). Also, partial resistance to *F. solani* f. sp. *pisi* has been reported in germplasm lines (Grünwald et al. 2003; Hwang et al. 1995; Infantino et al. 2006; Porter 2010) and markers linked to quantitative trait loci (QTL) for resistance have been identified (Feng et al. 2011). Breeding lines with improved partial resistance to *F. solani* f. sp. *pisi* have been developed (Porter et al. 2014). Partial resistance to both *F. solani* and *F. avenaceum* has been reported in the commercial cv. Franklin (Chittam 2012), associated with pigmented seed-coat (Kraft 1975; Porter 2015).

In lentil, the inheritance of resistance to vascular wilt has been investigated (Eujayl et al. 1998), but little research has been conducted on sources of resistance to root rot/wilt. In chickpea, sources of resistance to *Fusarium* wilt, dry root rot, and foot rot have been summarized previously (Infantino et al. 2006).

***Pythium* spp. Seed treatment.** Seed treatment with fungicides can reduce seedling blights for 2 to 3 weeks after sowing (Abawi et al. 2006; Hwang et al. 2000a; Leisso et al. 2009). Soil fumigants are also highly effective against *Pythium* spp. (Abawi et al. 2006). Several commercial biofungicides based on strains of the fungal genera *Trichoderma* and *Gliocladium* provided effective reduction in seed rot and seedling blight (Fravel 2005; Howell et al. 1993). Bacteria and actinomycetes with activity against *Pythium* spp. have also been identified (Bardin et al. 2004; Chin-A-Woeng et al. 2003; Hynes et al. 2008; Naseby et al. 2001), but are not yet commercially available. Seed treatment with *Rhizobium leguminosarum* bv. *viceae* reduced damping-off and increased plant growth and seed yield of pea and lentil (Huang and Erickson 2007).

Cultural practices. Low soil temperature did not increase the severity of seedling blight or root rot on field pea, and the optimum temperature for infection was 15 to 22.5°C for *P. ultimum* and 17.5 to 27.5°C for *P. irregulare* (Hwang et al. 2000a). However, emergence was 10 to 15% lower and seed yield was 20 to 50% lower when the crop was seeded into warm soil in late May to early June. This indicated that, on the northern prairies, the crop should be seeded early to maximize yield, even where soils are infested with *Pythium* spp.

For most crops, well-drained soils and balanced irrigation reduce the risk of infection by species such as *P. aphanidermatum* (Edson) Fitzp., likely via reduced potential for dispersal of zoospores. For example, deep plowing and the use of raised ridges increased aeration and drainage, and reduced root rot on bean, which is favored by high moisture (Rosado May et al. 1985). Application of organic soil amendments and including nonhost crops in the cropping rotation can also reduce root rot severity (Buruchara 1991; Rosado May et al. 1985; Volland and Epstein 1994).

Resistance. In field pea, three kinds of genetic resistance have been identified; lines where the juvenile susceptibility of seed is lost within 48 h after imbibition begins, which reduces seed rot; lines with round, unwrinkled seed that exude lower amounts of substances stimulatory to *Pythium*; and lines with anthocyanin pigment in the seed coat, which is fungistatic to *P. ultimum* (Muehlbauer and Kraft 1978; Stasz et al. 1980). Lines with resistance to both *P. ultimum* and *F. solani* f. sp. *pisi* have been identified (Ali et al. 1994; Kraft and Roberts 1970).

In bean, resistance to *P. ultimum* was controlled by a single dominant gene (Mahuku et al. 2005; Otsyula et al. 2003). In faba bean, white-flowered lines were more susceptible to infection by *P. debaryanum* Hesse than those with colored flowers (Kantar et al. 1996), likely due to a linkage between reduced pigmentation in flowers and reduced tannin content in the seed coat (Cabrera and Martin 1989). Also, the *P* gene for flower and seed coat color in chickpea is closely linked to resistance (Kumar et al. 1991).

***Rhizoctonia solani*. Seed treatment.** Treatment with systemic fungicides generally improved seedling establishment and yield in inoculated trials more than contact fungicides (Kuznia et al. 1993; Harveson et al. 2005; O'Brien et al. 1991; Tu 1992). Seed treatments that contained combinations of active ingredients were highly effective, but the most effective combinations were not the same across

crops (Chang et al. 2014; Hwang et al. 2003, 2007). Seed treatment with biocontrol agents, often in combination with synthetic fungicides or plant defense inducers, showed promise (Abdel-Monaim 2013; Estévez de Jensen et al. 2002; Xue 2003b). Also, application of rhizobia improved seedling health and increased subsequent plant dry weight in chickpea (Hemissi et al. 2013).

Cultural practices. Across a range of pulse crops, deep seeding reduced seedling establishment and increased root rot severity when inoculum pressure was high (Chang et al. 2004b, 2008; Hwang et al. 2007). Late seeding generally produced the lowest yield of field pea, lentil, and chickpea (Chang et al. 2004b, 2008; Hwang et al. 2007). Increasing seeding rates of field pea up to 90 seeds m⁻² increased stand establishment and seed yield (Hwang et al. 2007). Soil compaction and the depletion of soil organic matter also reduced seedling emergence and vigor and increased root rot severity (Tu 1992; Tu and Tan 1991). Reduction in soil compaction also favored root development and vigorous plants, so deep tillage reduced root rot severity (Tan and Tu 1995).

The initial inoculum density of *R. solani* had a major impact on seedling emergence, root rot severity, and yield of pulse crops (Chang et al. 2008, 2014). Rotation with nonhost crops for at least 3 years reduced inoculum density (Dillard 2001; Hanson 2005; Porter et al. 2011). However, other susceptible crops include canola, mustard, soybean, sunflower (Porter et al. 2011), and potato (Hanson 2005; Mathew et al. 2012), so cereals represent the only nonhost crops grown on a large acreage on the northern Great Plains.

Resistance. In field pea, partial resistance or tolerance has been identified (McCoy and Kraft 1984a, b; Hwang et al. 2007; Shehata et al. 1984), but no lines were highly resistant (Shehata et al. 1981). Resistance to seed rot appears to be under separate genetic control from stem rot, linked to purple flower color and a dark seed coat. Resistance in pea was strongly correlated with epicotyl diameter, but showed little relationship to anthocyanin pigmentation of the seed or seedling emergence rate (McCoy and Kraft 1984b). Interestingly, the media used to produce inoculum affected the expression of stem rot resistance in pea (McCoy and Kraft 1984a).

In bean, partial resistance has been identified in lines from Latin America (Burke and Miller 1983; Conner et al. 2014; Peña et al. 2011), but is more common in Mesoamerican than Andean genotypes (Nicoli et al. 2011). However, root rot resistance was not always associated with resistance to seedling blight (Conner et al. 2014). Resistant bean plants produced phytoalexins that made the host tissue more resistant to pectic enzymes and caused fungistasis in *R. solani* (Bateman et al. 1969). Symptoms in resistant genotypes were most severe under high temperatures and saturated soil conditions, which enhanced the growth of the pathogen more than the host (Shehata et al. 1984).

In lentil, no lines have been identified as resistant (Kaiser and Horner 1980; Kraft et al. 1994), but lines with tolerance or partial resistance have been reported (Kannaiyan and Nene 1976; Wang et al. 2006). In chickpea, lines with partial or even high levels of resistance have been identified (Gupta and Babbar 2006; Oad et al. 1995; Parashar and Indra Hooda Sindhan 1992). In faba bean, lines with partial resistance have been identified (Assunção et al. 2011) but previous attempts at selection over several generations did not result in improvements in resistance (Rashid and Bernier 1993). In lupin, only a few lines have been tested, but one breeding line appeared to be tolerant (Thiele et al. 2008).

***Aphanomyces euteiches*.** **Seed treatment.** Ethaboxam is registered in the United States and Canada for suppression of early-season root rot on many pulse crops except field pea (Anonymous 2014), but published reports of its efficacy are not yet available. Seed treatment with fosetyl-Al reduced root rot on pea in greenhouse trials (Oyarzun et al. 1990). Seed treatment with hymexazol and soil drenches with azoxystrobin and propomocarb reduced severity in Australia (Watson et al. 2013).

Application of Phostrol (sodium, potassium, and ammonium phosphites) reduced the impact of *A. euteiches* in some studies (Porter 2006; Porter and Coffman 2007), but not others (Conner et al. 2013; Gossen et al. 2016; Gundersen et al. 2006). Several biological

agents have been reported to reduce severity (Bødker et al. 2002; Thygesen et al. 2004; Wakelin et al. 2002; Xue 2003a, b), but none are commercially available.

Cultural practices. Where *A. euteiches* is endemic, the recommended practice is avoidance based on the inoculum potential in field soil. Inoculum potential is an index of disease activity, and soil factors that inhibit or promote infection, which until recently has been estimated based on greenhouse grow-out tests (Harveson et al. 2014; Hughes and Grau 2013; Malvick et al. 1994; Sauvage et al. 2007). There is a strong correlation between severity in the greenhouse and the field, so this assay has been used as a predictive test, but is slow and labor-intensive. Recently, several seed testing labs in Canada have utilized an *A. euteiches* detection test based on DNA using PCR, but it does not provide information on inoculum levels in the soil.

Precropping with oat or Brassica green manures (reviewed by Hossain et al. 2012) have shown potential for reducing severity (Fritz et al. 1995; Hossain et al. 2014; Smolinska et al. 1997; Williams-Woodward et al. 1997).

Resistance. Soybean, chickpea, and some faba bean lines carry resistance (Malvick et al. 2009; Moussart et al. 2008, 2013; Vandemark and Porter 2010). Lines of field pea with partial resistance have been developed and released (McGee et al. 2012), and a line with tolerance has been identified (Conner et al. 2013). Partial resistance and tolerance were found to be controlled by multiple genes with complex inheritance (Hamon et al. 2013; McGee et al. 2012; Moussart et al. 2007) and QTLs contributing to partial resistance have been described (Hamon et al. 2013; Pilet-Nayel et al. 2005, 2009). Two races have been described based on an alfalfa differential set (Malvick and Grau 2001; Malvick et al. 2009), but screening isolates from pea against a set of pea differentials failed to characterize distinct groupings (Wicker and Rouxel 2001; Wicker et al. 2003).

Conclusions

Cultivars that carry broad resistance to the root rot complex would be important tools for the cost-effective management of root rot for the pulse industry. However, resistance to each pathogen is generally only partial, based on multiple genes or QTLs. Marker-assisted selection is likely to be the only effective breeding strategy for developing commercial cultivars with broad resistance.

In the absence of commercial cultivars with high levels of resistance, use of seed treatments has been and will continue to be an important component of root rot management on pulses. Over the last decade, there has been substantial progress in identifying combinations of active ingredients with efficacy against *Fusarium* spp., *Pythium* spp., and *R. solani* on pulse seedlings. However, seed treatments generally do not provide useful levels of reduction in root rot severity later in the season. For *A. euteiches*, fungicide options are even more limited. Concern has been expressed that widespread use of synthetic fungicides to manage root diseases could have adverse effects on the environment or result in development of insensitivity in pathogen populations (Djébalí et al. 2014). However, the relatively low application rate, low nontarget toxicity, and below-ground target for most seed treatments makes them a desirable disease management option on the northern Great Plains (Gossen et al. 2014).

Seed treatments with biological agents represent a potentially powerful tool for root rot management in pulses. Biologicals may provide season-long root rot suppression by colonizing the root and stimulating host defense responses (Xue 2003a, b), and some may carry over from one year to the next. They might also provide options for management of *A. euteiches*. To date, however, few consistently effective biofungicides are commercially available for seedling blight and root rot of pulses. Progress in this area would be welcome.

The development of molecular techniques for identifying the agents causing seedling blight and root rot of pulses and quantifying inoculum in the field is an exciting development whose potential is only just starting to be realized (e.g., Esmaeili Taheri et al. 2015; Henriquez et al. 2014; Zitnick-Anderson et al. 2016). Molecular approaches have largely replaced culturing of pathogens of

high value crops, and these techniques are being developed rapidly for field crops such as pulses as well. The data generated using these new techniques is changing our understanding of the root rot complex on pulses. An important example is the change in the understanding of the importance of *A. euteiches*, which has only recently been recognized as an important component of the disease complex on pulses across the northern Great Plains (Banniza et al. 2013; Chatterton et al. 2014, 2015a, b; Zitnick-Anderson and Pasche 2016). Its wide distribution is a clear indication that this pathogen has been present for many years. Similarly, molecular techniques are driving a reassessment of the distribution and importance of the individual species that make up the root rot complex on pulse crops across the region (Esmaili Taheri et al. 2015; S. Chatterton, unpublished data). Techniques are being developed to utilize next-generation sequencing for rapid quantification of root rotting pathogens in host tissue (Zitnick-Anderson et al. 2015). This technique has the potential for use in large-scale population studies that will enhance our knowledge of host-pathogen and pathogen-pathogen interaction in the root rot disease complex.

Cultural approaches for managing root rot generally focus on minimizing inoculum pressure and optimizing crop health and vigor. Minimizing inoculum pressure is achieved by maintaining diverse crop rotations with nonhosts (not always possible, e.g., *F. avenaceum*), and through avoidance of heavily infested fields. Crop health aspects include using high seeding rates of vigorous seed, minimizing soil compaction to ensure adequate drainage and root growth, seeding early to maximize yield potential, and seeding shallow to ensure rapid germination and seedling establishment. Individually, these approaches are only partially effective, so they need to be used in combination with seed treatment and resistance (where available).

The next steps forward will likely involve quantifying root pathogen levels in individual fields and developing specific management recommendations for each field. The capability to quantify the main root rot pathogens in plant or soil samples using molecular tools is improving rapidly and costs are coming down. However, an important drawback to implementing assessment of individual fields is the difficulty of collecting representative samples of soilborne pathogens. Most root rot pathogens have an inherently uneven distribution in each field, so the risk of developing severe root rot may differ dramatically among portions of a field. Information on field characteristics (soil type and texture, slope, catchment areas), together with yield data (e.g., from yield monitors) and images from drones, planes, or possibly even satellites, may be required to develop sampling protocols that identify areas of a field at risk of root rot and facilitate collection of representative samples of each risk area. Once representative data on pathogen levels within individual fields is available, a decision support system could be developed to assess the disease risk in a field and recommend a combination of seed treatments, host genetics, agronomic inputs, and cultural management practices to minimize disease risk.

The impact and management of root rot diseases has almost always been conducted in studies inoculated with a single pathogen. Studies of the interaction of root rot pathogens are rare. However, knowledge of the interaction among root rot pathogens under a range of weather conditions will likely be required to optimize any decision support system for individual hosts.

In the short term, development of new tools, such as partially resistant cultivars and pathogen avoidance based on risk assessment of individual fields, will be important additions to existing tools such as seed treatment and early, shallow seeding to manage root rot risk to maintain or even increase pulse production across the region.

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Dr. Gossen received a Ph.D. from the University of Saskatchewan in 1985. Since then, he has worked as a research scientist with Agriculture & Agri-Food Canada in Canada, specializing in diseases of field crops on the northern Great Plains. His research on disease management of pulse, canola, forage, and other crops has been recognized with awards including the Golden Harvest Award from AAFC in 2008 and the Award for Outstanding Research from the Canadian Phytopathological Society in 2010. He has also had a leadership role in professional organizations, including president of the Canadian Phytopathological Society. He currently leads studies on root rot of field pea and clubroot of canola.

Dr. Conner has worked with Agriculture and Agri-Food Canada for over 34 years. Bob received his Ph.D. in plant pathology in 1981 from the University of Manitoba. From 1982 to 2001, he was employed as a cereal pathologist at Lethbridge Research Centre where he primarily studied foliar and root diseases of soft white spring wheat and hard red winter wheat. Currently, he is the pulse crop pathologist at the Morden Research and Development Centre, where he focuses on diseases of beans and other pulse crops. His research includes studies on foliar and root diseases of dry bean, field pea, faba bean, and soybean. Dr. Conner has served as associate editor for three scientific journals, participated on the advisory committee of several graduate students, and supervised seven post-doctoral fellows. During his career, Bob has published many scientific manuscripts and review articles. In 2016 as part of a team of researchers, Bob received the Achievement in Plant Disease Management Award from the Canadian Phytopathological Society.

Dr. Chang obtained his M.Sc. degree from Washington State University and Ph.D. degree from the University of Alberta, Canada. He has been associated with Alberta Agriculture and Forestry since 1988 as a plant pathologist working on diseases of greenhouse and horticultural crops and their management. Since 1995, Dr. Chang has devoted his time to conducting research on diseases of special crops, medicinal herbs, and pulses at three Provincial Research Centres in southern and central Alberta. He has been highly successful in identifying and characterizing new plant disease organisms of significance to Alberta and published numerous papers. Dr. Chang also serves as an adjunct professor at the University of Alberta and has supervised several graduate students and post-doctoral fellows working on pulse crop diseases. In 2016, Dr. Chang, along with his research colleagues in the Pulse Crop Research Team, received the Achievements in Plant Disease Management Award from the Canadian Phytopathological Society.

Dr. Pasche joined North Dakota State University as an assistant professor and pulse crops pathologist in 2012. The mission of her research program is to develop management tools and strategies for economically important diseases of dry edible beans, dry edible peas, lentils, and chickpeas. Currently, Dr. Pasche's focus is to develop genetic and chemical management tools for root rot and bacterial pathogens, characterize emerging pathogens like *Pea seedborne mosaic virus*, and monitor other pathogens for evolutionary changes that impact management, such as fungicide

resistance development and pathogen race changes. Dr. Pasche currently leads the North Central Region - Pulse Crops Working Group (NCIPM Center – USDA-NIFA) and is involved with several pulse crop organizations and multistate research projects.

Dr. McLaren received her B.Sc. from the University of Manitoba after which she worked as a technician at Agriculture and Agri-Food Canada (AAFC) in Morden, MB, where Dr. H. C. Huang introduced her to *Sclerotinia sclerotiorum* and there was no turning back. She completed her M.Sc. and Ph.D. in plant pathology from the University of Manitoba with input from the University of Minnesota (coursework) and AAFC-Lethbridge (research studies). During her postdoctoral position at AAFC-Beaverlodge, she continued her study of soilborne root pathogens. Since 1998, she has worked as a research scientist with AAFC in Brandon, MB, specializing in crop production pathology. She has led numerous studies on management strategies for pulse crop, canola, and potato diseases and currently leads studies on soybean root rot. Recently, she accepted the position of editor for the oilseeds, pulses, forages, and special crops section of the Canadian Plant Disease Survey published by the Canadian Phytopathological Society. As a member of a pulse crop research team, Debra was awarded the Achievements in Plant Disease Management Award from the Canadian Phytopathological Society in 2016.

Dr. Henriquez has over 15 years of plant pathology research experience. She completed her Ph.D. in plant pathology from the University of Manitoba in 2011. Prior to taking up her Ph.D., she worked as research assistant and laboratory manager for five years in the bean pathology program at the International Center for Tropical Agriculture (CIAT). During her postdoctoral research at Agriculture and Agri-Food Canada (AAFC), she worked with root rot pathogens in dry bean, soybean, and field peas. Currently, Dr. Henriquez is a research scientist in wheat pathology at Agriculture and Agri-Food Canada (AAFC).

Dr. Chatterton received her Ph.D. in biological sciences at Simon Fraser University in 2010, with a specialization in biological control of root diseases of greenhouse crops. She joined Agriculture and Agri-Food Canada's Lethbridge Research and Development Centre in 2011 as a research scientist in special crops pathology. She is an adjunct professor at the University of Lethbridge. Her research focus is on management of root and foliar diseases of pulse crops (field pea, lentil, dry bean, and faba bean), and molecular diagnostics and quantification of soilborne pathogens.

Dr. Hwang graduated from Washington State University and worked for over 20 years at Alberta Research Council in Vegreville on soilborne diseases of field crops, particularly pulses. She became an adjunct professor at the University of Alberta in 2005 and since then she has supervised many graduate students and produced numerous publications. She joined Alberta Agriculture in 2006 to initiate an intensive research program to develop methods of controlling clubroot of canola. In 2011, she received an award from the Canadian Phytopathological Society (CPS) for Outstanding Research in Plant Pathology. In 2014 and 2016, along with her research colleagues, she received awards for Achievement in Plant Disease Management from the CPS for the progress in both canola clubroot research and pulse disease research.

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