DISTRIBUTION OF *RHIZOCTONIA SOLANI* AG 2-2 INTRASPECIFIC GROUPS IN THE RED RIVER VALLEY AND SOUTHERN MINNESOTA

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Rhizoctonia root and crown rot (RRCR) of sugarbeet is caused by the soilborne fungus *Rhizoctonia solani*. The fungus is composed of genetically isolated populations called anastomosis groups or AGs (3). The AG population causing RRCR of sugarbeet is *R. solani* AG 2-2, which is further divided into the intraspecific groups (ISGs) AG 2-2 IIIB and AG 2-2 IV. Both ISGs cause RRCR on sugarbeet, but AG 2-2 IV is reported as the primary cause (3, 5), while AG 2-2 IIIB is the more aggressive population (4).

Rhizoctonia root and crown rot is becoming a more frequent and severe problem in the sugarbeet-growing regions of Minnesota and North Dakota. In the Red River Valley, this is partly due to increases in acreage of soybean and edible bean crops, which are susceptible to *R. solani* AG 2-2 (5) and are grown in rotation with sugarbeet. As these crop rotations continue, sugarbeet crops are likely to be exposed to higher soil populations of *R. solani* AG 2-2. Methods for controlling RRCR on sugarbeet include rotating to non-host crops (i.e. small grains), applying the fungicide azoxystrobin (Quadris), and planting resistant varieties.

Selection of rotation crops is influenced by a number of factors. The effect a particular crop may have on diseases of other crops in the rotation certainly is one important factor. Other overriding factors, however, are the marketability and commodity prices of different crops. Producers need to make money on crops they raise to stay in business, so economics often has the greatest influence on crops selected for rotation with sugarbeet. According to the USDA National Agriculture Statistics Service, acreage of some common crops grown in counties bordering Minnesota and North Dakota (north of Fargo) changed significantly from 1995 to 2005. Production of spring wheat (a non-host of *R. solani* AG 2-2) decreased 12.3% from 2,108,700 to 1,849,900 acres. Acreage of soybean (a host of *R. solani* AG 2-2) increased 177% from 487,000 to 1,346,800. Corn production also increased from 189,400 to 332,400 acres (76%).

The effect of corn on populations of *R. solani* AG 2-2 is not well known in the sugarbeet-growing areas of Minnesota and North Dakota. Corn is reported as susceptible to crown and brace root rot caused by *R. solani* AG 2-2 IIIB in Europe (2) and in the southeastern U.S.A (6). Recent reports indicate that *R. solani* AG 2-2 IIIB can infect corn roots without causing aboveground symptoms or yield loss (7,8,9).

There is no information on the prevalence and distribution of *R. solani* AG 2-2 ISGs in the sugarbeet-growing areas of Minnesota and North Dakota. Knowing where these ISGs occur, and how they are affected by crops rotated with sugarbeet, are important in learning how to reduce populations of *R. solani* AG 2-2 and manage RRCR on sugarbeet.

OBJECTIVES

Our objectives were to determine the prevalence and distribution of *R. solani* AG 2-2 ISGs in the sugarbeet growing areas of Minnesota and North Dakota.

MATERIALS AND METHODS

Collection of R. solani isolates. Sugarbeet seedlings or roots with symptoms of RRCR were submitted by industry agriculturists and crop consultants during the 2006 and 2007 growing seasons. If symptoms appeared to be caused by *R. solani*, plants were washed under running tap water or with a pressure washer. Pieces (1cm x 1cm) were cut from the margin of healthy and infected tissue, surface-treated for 15 sec in 0.5% NaOCl, rinsed twice in sterile deionized water, and placed in petri dishes containing sterile deionized water or acidified potato dextrose agar (PDA). Fungal growth consistent with *R. solani* was transferred to fresh PDA. Cultures then were sub-cultured on

thin plates of water agar where they were "hyphal-tipped" to ensure genetic purity and free them of contamination from other fungi or bacteria. Isolates with cultural characteristics typical of *R. solani* AG 2-2 were maintained on sterile barley grains and stored in an ultra-low freezer at -112 °F. The culture collection also included isolates of *R. solani* AG 2-2 from diseased sugarbeet samples received in 2005, with a few from prior growing seasons.

Determination of intraspecific group (ISG). Identification of ISG (IIIB and IV) of *R. solani* AG 2-2 cultures was done by testing growth at 95 °F. While AG 2-2 IIIB grows at 95 °F, AG 2-2 IV does not (5). Each isolate was transferred to eight, 4-inch diameter petri dishes containing 20 ml PDA. Four replicate dishes of each isolate were placed in an incubator at 77 °F (control) and four replicate dishes were placed in an incubator at 95 °F. After 24 hours, a line was drawn at the margin of culture growth to mark the "baseline". Fungal growth was measured from this baseline after an additional 24 and 48 hours of incubation. Growth at 95 °F was expressed as a percent of growth at 77 °F for each isolate and compared to known "check" isolates of each ISG. Isolates were identified as AG 2-2 IV if little or no growth occurred and as AG 2-2 IIIB if growth was similar to, or better than the known AG 2-2 IIIB check. Isolates were identified as "intermediate" if growth occurred, but was less than the AG 2-2 IIIB check.

RESULTS

Over 797 isolates of *R. solani* AG 2-2 were collected from diseased sugarbeet plants in 253 fields within Minnesota and North Dakota, including 12 counties of American Crystal Sugar Company (ACSC), 12 counties of the Southern Minnesota Beet Sugar Cooperative (SMBSC), and 1 county of the Minn-Dak Farmers Cooperative (Figure 1). Of these isolates, 369 (46%) were *R. solani* AG 2-2 IV, 320 (40%) were *R. solani* AG 2-2 IIIB, and 108 (14%) were interm

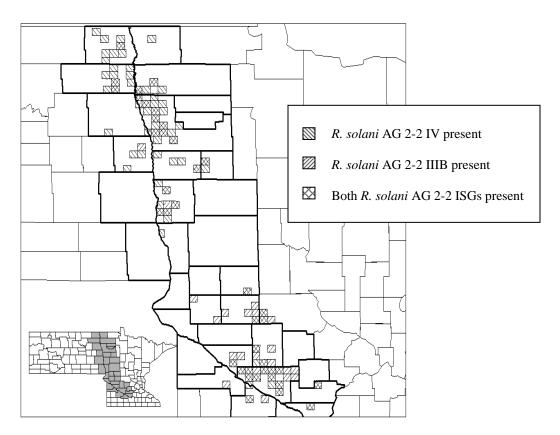


Figure 1. Distribution of *Rhizoctonia solani* AG 2-2 intraspecific groups (ISG, IV and IIIB) in the Red River Valley and southern Minnesota. The ISG was determined by growth of the fungus at 95 0 F; AG 2-2 IIIB grows at 95 0 F but AG 2-2 IV does not.

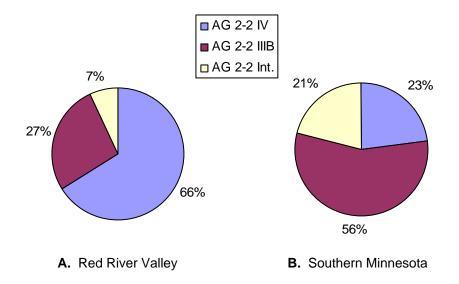


Figure 2. Percent incidence of *Rhizoctonia solani* AG 2-2 intraspecific group (ISG [IV and IIIB]) of cultures from sugarbeet with Rhizoctonia root and crown rot collected in **A.**) the Red River Valley of Minnesota and North Dakota (wheat and sugarbeet main crops in rotation) and **B.**) southern Minnesota (sugarbeet, corn, and soybean are major crops in rotation). The ISG was determined by growth of the fungus at 95 °F where AG 2-2 IIIB grows at 95 °F and AG 2-2 IV does not; growth of "intermediate" (int.) cultures occurred at 95 °F, but was weak. Values are based on 797 cultures of *R. solani* AG 2-2 (428 from the Red River Valley and 369 from southern Minnesota).

The ISGs of *R. solani* AG 2-2 were distributed throughout the Red River Valley and southern Minnesota, but differed in prevalence between the two regions (Figure 1). Of 428 cultures from the Red River Valley (Figure 2A), 283 (66%) were AG 2-2 IV, 114 (27%) were AG 2-2 IIIB, and 31 (7%) were intermediate. By contrast, of 369 cultures collected in southern Minnesota (Figure 2B), 86 (23%) were AG 2-2 IV, 206 (56%) were AG 2-2 IIIB, and 77 (21%) were intermediate.

Within the Red River Valley, several crops preceded sugarbeet and prevalence of ISGs differed among previous crops (Figure 3). There were 211 isolates collected from 73 fields where wheat was the previous crop. Of these isolates, 162 (77%) were AG 2-2 IV, 36 (17%) were AG 2-2 IIIB, and 13 (6%) were intermediate (Figure 3A). In contrast, of 69 isolates from 17 fields where edible bean was the previous crop, 29 (42%) were AG 2-2 IV, 35 (51%) were AG 2-2 IIIB, and 5 (7%) were intermediate (Figure 3B). When soybean was the previous crop (42 isolates collected from 13 fields), 24 isolates (57%) were AG 2-2 IV, 12 (29%) were AG 2-2 IIIB, and 6 (14%) were intermediate (Figure 3C). There were 16 isolates from 6 fields where potato was the previous crop and 13 (81%) were AG 2-2 IV, only 1 (6%) was AG 2-2 IIIB, and 2 (13%) were intermediate (Figure 3D). There were nine isolates from four fields where the previous crop was either corn or rye grass or the field was fallow. Information on previous crop was unavailable for 81 isolates collected from 30 fields.

In southern Minnesota, previous crops were less diverse compared to the Red River Valley, and prevalence of ISGs of *R. solani* AG 2-2 isolated from diseased sugarbeet varied somewhat (data not shown). There were 178 isolates from 54 fields where corn was the previous crop and 45 (25%) were AG 2-2 IV, 96 (54%) were AG 2-2 IIIB, and 37 (21%) were intermediate. Soybean was the previous crop for 30 isolates collected from 9 fields, and 11 (37%) were AG 2-2 IV, 19 (63%) were AG 2-2 IIIB, and there were no intermediate isolates. When edible bean was the previous crop (16 isolates from 4 fields), 4 isolates (25%) were AG 2-2 ISG IV, 11 (69%) were AG 2-2 IIIB, and 1 (6%) was intermediate. There were 18 isolates from 5 fields where the previous crop was alfalfa, peas, potato, or wheat. Information on previous crop was unavailable for 127 isolates from 38 fields.

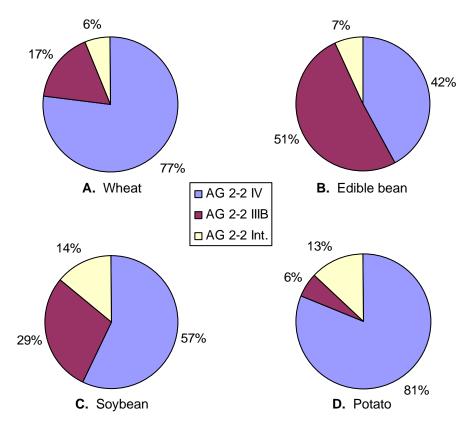


Figure 3. Percent incidence of *Rhizoctonia solani* AG 2-2 intraspecific groups (ISG [IV and IIIB]) of cultures from sugarbeet with Rhizoctonia root and crown rot collected in the Red River Valley with previous crops of **A.**) wheat (211 isolates from 73 fields), **B.**) edible bean (69 isolates from 17 fields), **C.**) soybean (42 isolates from 13 fields, and **D.**) potato (16 isolates from 6 fields). The ISG was determined by growth of the fungus at 95 °F where AG 2-2 IIIB grows at 95 °F and AG 2-2 IV does not; growth of "intermediate" (int.) cultures occurred at 95 °F, but was weak.

DISCUSSION

The ISGs of *R. solani* AG 2-2 (IV and IIIB) are distributed throughout the sugarbeet-growing areas of Minnesota and North Dakota, with AG 2-2 IV predominating in the Red River Valley and AG 2-2 IIIB in southern Minnesota. These differences likely are attributed to historical crop rotations in the two locations. Crop rotations in the Red River Valley historically included mostly small grains (primarily spring wheat) but now are changing to increased production of soybean, edible bean, and corn. This trend may result in a shift in prevalence from *R. solani* AG 2-2 IV to AG 2-2 IIIB. Data from our survey suggests this may be happening because prevalence of AG 2-2 IIIB following previous crops of wheat, soybean, and edible bean averaged 17, 29, and 51%, respectively. Only six isolates from two fields had corn as the previous crop so it is impossible to determine the influence of corn on prevalence of ISGs.

Southern Minnesota has grown corn and soybean in rotation with sugarbeet since the cooperative was established in the 1970s. Although both ISGs infect soybean, AG 2-2 IIIB is more aggressive than AG 2-2 IV (1), so populations of AG 2-2 IIIB likely increased over the years. Interestingly, there was a higher percentage of "intermediate" isolates of *R. solani* AG 2-2 in southern Minnesota than in the Red River Valley. This population indicates increased potential for disease because it is as aggressive as *R. solani* AG 2-2 IIIB on sugarbeet (4). In addition, corn may also have favored build-up of AG 2-2 IIIB since it is reported as susceptible to this ISG in Europe (2), southeast U.S.A. (6) and recently, in the Red River Valley (7,8,9). Research is underway to determine the effect of corn varieties with different traits on soil populations of *R. solani* AG 2-2 IV and IIIB and severity of RRCR on a subsequent sugarbeet crop In light of the long-term rotation of bean crops, corn, and sugarbeet in southern Minnesota, RRCR of sugarbeet is not as severe as expected. This may be the result of early planting of sugarbeet, which reduces RRCR (1) or that corn does not result in build-up of populations of *R. solani* AG 2-2.

In both the Red River Valley and southern Minnesota, isolates of *R. solani* AG 2-2 IIIB were most common in sugarbeet following edible bean. Production of edible bean in rotation with sugarbeet appears to be very favorable for build-up of populations of *R. solani* AG 2-2 IIIB.

Evaluation of cropping history the year before sugarbeet production may not be the best way to judge effects of rotation crops on prevalence of *R. solani* AG 2-2 ISGs. The effect of rotation crop on sugarbeet RRCR could likely be affected by crops grown two or more previous growing seasons. In a typical rotation, there are at least two growing seasons between sugarbeet crops. Also, sugarbeet was the source of isolates of *R. solani* identified in this survey. If other crops had been sampled, perhaps different percentages of ISGs of *R. solani* AG 2-2 IV and IIIB would have been identified.

It is unknown how differences in prevalence of *R. solani* AG 2-2 ISGs in the Red River Valley and southern Minnesota, and among fields with different previous crops, affect prevalence and severity of RRCR on sugarbeet and root disease of other crops. Research is currently underway to test pathogenicity of some isolates of each ISG from both growing areas on different varieties of corn, edible bean, soybean, and sugarbeet. Also, field trials are established to determine effects of different crop rotations (wheat/soybean/sugarbeet, wheat/corn/sugarbeet, and wheat/wheat/sugarbeet) on populations of *R. solani* AG 2-2 IV and IIIB.

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