VARIABILITY IN AGGRESSIVENESS BETWEEN AND WITHIN INTRASPECIFIC GROUPS OF *RHIZOCTONIA SOLANI* AG 2-2 ON SUGARBEET AND ROTATION CROPS

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Rhizoctonia crown and root rot (RCRR) of sugarbeet, caused by the soilborne fungus *Rhizoctonia solani*, is increasing in prevalence in the United States, Europe, and other countries (1,2). The fungus is composed of 13 genetically isolated populations called anastomosis groups or AGs (8). The primary population attacking sugarbeet is *R. solani* AG 2-2, which is further divided into the intraspecific groups (ISGs) AG 2-2 IV and AG 2-2 IIIB. Both ISGs occur in Minnesota and North Dakota (1) and produce identical symptoms of RRCR on sugarbeet. The ISGs of AG 2-2 are identified by growth on culture media at 95 °F; AG 2-2 IIIB grows at this temperature but AG 2-2 IV does not (8). According to the literature, AG 2-2 IIIB is more aggressive and has a wider host range (e.g., bean crops, corn) than AG 2-2 IV (3-6, 8).

The Sugarbeet Plant Pathology Laboratory at the University of Minnesota, Northwest Research and Outreach Center (NWROC), Crookston, has collected nearly 1,000 cultures of *R. solani* AG 2-2 from sugarbeet with symptoms of RCRR throughout the Red River Valley (RRV) and southern Minnesota. The collection has been identified to ISG by differential growth at 95 °F. In 2009, collaborations were initiated with plant pathologists Dr. Frank Martin (USDA-ARS, Salinas, CA) and Dr. Linda Hanson (USDA-ARS, Michigan State University, East Lansing) to develop molecular markers to analyze genetic population structure of the collection. A subset of 48 cultures was selected to represent maximum diversity and preliminary evidence indicates considerable genetic variability <u>within</u> each ISG. This variability likely occurs because *R. solani* has multiple nuclei in each cell. Population structure of the cultures (based on molecular markers), will be correlated with their aggressiveness/pathogenicity on sugarbeet and rotation crops.

Previous results of pathogenicity of *R. solani* AG 2-2 IV and AG 2-2 IIIB on seedlings and adult plants of sugarbeet, bean, and wheat have been previously reported (12,13) and also are included here for comparison purposes.

OBJECTIVES

Experiments were conducted to evaluate 48 cultures of *R. solani* AG 2-2 (24 cultures each of AG 2-2 IV and AG 2-2 IIIB) collected in Minnesota and North Dakota for: 1) aggressiveness/pathogenicity on seedlings and adult plants of sugarbeet and several rotation crops and 2) variability in pathogenicity between and within both populations.

MATERIALS AND METHODS

A subset of 48 cultures of *R. solani* from sugarbeet with RCRR were confirmed as AG 2-2 by polymerase chain reaction (PCR) and identified to ISG by growth on agar media at 95 °F; 24 cultures were AG 2-2 IV and 24 were AG 2-2 IIIB. Within each ISG, cultures originated in different geographic areas of Minnesota and North Dakota, from near the Canadian border to the Southern Minnesota Beet Sugar Cooperative (~280 miles, Fig. 1). Fields had been sown to various crops the previous season (corn, sweet corn, edible bean, potato, soybean, spring wheat). Cultures were isolated from sugarbeet varieties differing in susceptibility to RCRR and from different parts of the root (crown or tap root).

<u>Pathogenicity tests.</u> Each culture of *R. solani* was tested for pathogenicity on seedlings and roots/basal stems of adult plants of sugarbeet, pinto bean, soybean, sunflower, hard red spring wheat, field corn, and sweet corn. Inoculum for seedling pathogenicity tests was prepared by growing each culture of *R. solani* AG 2-2 on sterilized barley grain for 3 weeks. Then, the grain was dried and ground in a coffee grinder (Proctor Silex). Inoculum for pathogenicity tests on adult rotation crops was prepared by growing each culture on sterilized whole corn kernels for 3 weeks. All pathogenicity tests were arranged in a randomized complete block design with four replicates and included a non-inoculated control. Each test was repeated, except for the second test on sunflower seedlings, which will be completed in 2012.

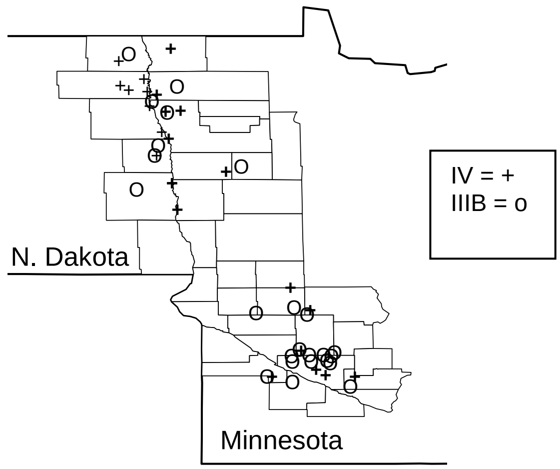

Fig. 1. Source of 24 cultures of *Rhizoctonia solani* AG 2-2 IV (noted by "+") and 24 cultures of AG 2-2 IIIB (noted by "o").

Sugarbeet seedlings. Seed of 'Beta 87RR38' treated with fungicides (standard rates of Apron + Thiram + 20 g Tachigaren/unit) was sown in 4.5 x 4.5 x 4.5 inch plastic pots (25 seeds/pot) filled with a commercial soil (Berger BM2, fertilized with 2g/Liter Osmocote 14-14-14 slow release fertilizer) and mixed with ground barley inoculum of *R. solani* (15 mg/pot). Soil was watered to keep moist and pots were incubated 4 weeks in a controlled environment chamber at 75 °F with a 14-hr photoperiod. Seedling stands were counted three times per week and dying seedlings were removed and assayed in the laboratory to verify infection by *R. solani*. After 4 weeks, remaining seedlings were removed from soil, washed, and rated on a 0 to 3 scale where 0 = no disease and 3 = root completely rotted and plant dead. These ratings, along with the number of dying seedlings, were used to calculate a root rot index (0 to 100 scale; 0 = no disease, 100 = all plants dead at 4 weeks after planting).

Rotation crop seedlings. Seed of 'LaPaz' pinto bean, 'NK05RM304030' soybean, 'SO614U' sunflower (an oil variety), 'Fuller' hard red spring wheat, 'Pioneer 139D81' field corn, and Burpee 'Silver Choice Hybrid' sweet corn were sown in 4.5 x 4.5 x 4.5 inch plastic pots (10 seeds/pot) filled with a commercial soil (Berger BM2, fertilized with 2g/Liter Osmocote 14-14-14 slow release fertilizer) that was mixed with ground inoculum of *R. solani* at a rate of 1:500 by volume (modified from Sumner and Bell [10]). Soil was watered to keep moist and pots were incubated in a controlled environment chamber at 75 °F with a 14-hr photoperiod. After 12 days, plants were removed from soil, washed, and rated. Pinto bean, soybean, and sunflowers were rated on a 1 to 5 basal stem rot scale where 1 = no symptoms, 2 = one or a few pin-point dark spots, 3 = necrotic lesions < 0.5 cm long with 25-49% of stem girdled, 4 = necrotic lesions > 0.5 cm long with 50-74% of stem girdled, and 5 = shoot dead with 75-100% of stem girdled (7). Field corn and sweet corn were rated on a 1 to 5 scale where 1 = < 2% root surface rotted, 2 = 2-10% discoloration and decay, 3 = 11-50% discoloration and decay, 4 = > 50% discoloration and decay, and 5 = plant dead (10). Wheat subcrown internodes were rated on a 0 to 3 scale where 0 = healthy, 1 = 1-25% surface with lesions, 2 = 26-50% surface with lesions and brown discoloration, and 3 = more than 50% of surface with lesions and brown discoloration (11).

Sugarbeet adult plants. Seed of the same variety used in the seedling trial also was sown in 6-inch diameter plastic pots (three seeds/pot) filled with commercial soil mix (Berger BM2). Pots were watered as needed and incubated in the greenhouse from 70 to 80 °F with a 14-hr photoperiod. After 3 weeks, plants were thinned to one per pot and fertilized with Osmocote 14-14-14 slow release fertilizer (6 g/pot). At 8 weeks after planting, soil was scraped from the root surface to a 1-inch depth; one-half teaspoon of ground barley inoculum of *R. solani* was placed on the root surface; and then roots were re-covered with soil. An additional 250 cc of soil was added around the sugarbeet crown. Pots were watered and placed in a controlled environment chamber at 75 °F with a 14-hr photoperiod. After 12 days, roots were removed from soil, washed, and rated on a 0 to 7 scale: 0= no disease; 1 = superficial, scattered non-active lesions; 2 = shallow, dry rot cankers on $\leq$ 5% of root surface; 3 = deep dry rot cankers at crown or extensive lateral lesions affecting 6-25% root surface; 4 = extensive rot affecting 26-50% of root, with cracks and cankers up to 5 mm deep; 5 = 51-75% of root blackened with rot extending into interior; 6 = entire root blackened except extreme tip; and 7 = root 100% rotted and foliage dead)..

Rotation crop adult plants. The same varieties of pinto bean, soybean, sunflower, wheat, field corn, and sweet corn sown in the seedling trials also were used in adult plant pathogenicity trials. Seeds of all crops (except wheat) were sown in 6-inch diameter plastic plots (four seeds/pot) filled with commercial soil mix (Berger BM2). Pots were watered as needed and incubated in the greenhouse from 70 to 80 °F with a 14-hr photoperiod. Wheat seed was sown in 4 x 4 x 5-inch pots (four seeds per pot) and placed in a controlled environment chamber at 75 °F with a 14-hr photoperiod.

After 3 weeks, plants were thinned to two per pot (one per pot for sunflower, sweet corn, and field corn) and fertilized with Osmocote 14-14-14 slow release fertilizer (6 g/pot). At 8 weeks after planting (5 weeks after planting in the repeated trial for pinto bean), crops were inoculated by scraping soil from the root surface to a 1-inch depth; one *R. solani*-infested corn kernel was placed along the root surface; and soil was pushed back to cover the root. Pots were returned to the greenhouse and watered regularly to maintain high soil moisture. Wheat plants were inoculated 6 weeks after planting by the same method and returned to a controlled environment chamber. At 12 days after inoculation, all rotation crops were removed from pots, roots were washed free of soil, and then were rated for disease as previously described.

For each trial, subcrown internodes of wheat and basal stems/roots from the other rotation crops (from at least one replicate) were assayed in the laboratory to verify infection with *R. solani*. Pieces of roots or basal stems were excised from the margin of diseased and healthy tissue, surface-treated in 0.5% bleach, rinsed twice in sterile deionized water, and placed on modified tannic acid medium. Plates were examined for growth of *R. solani* AG 2-2 from 7 to 14 days later.

Statistical analysis. For each crop (seedling and adult tests), pathogenicity data were combined for repeated experiments and subjected to analysis of variance to determine if AG 2-2 IV cultures differed from AG 2-2 IIIB (P = 0.05).

RESULTS

Seedling Pathogenicity Tests:

Sugarbeet seedlings. Overall, cultures of *R. solani* AG 2-2 IIIB were significantly more aggressive than AG 2-2 IV in causing disease (Table 1). Root rot ratings (0 to 100 scale) for AG 2-2 IIIB averaged 78 and for AG 2-2 IV averaged 51. There was considerable variability in aggressiveness, however, within each ISG. Cultures of AG 2-2 IV fell into nearly a full spectrum of root rot severities, ranging from 5 to 100 (Table 1) and 12 cultures resulted in root rot ratings between >50 to 100 (Fig 1A). Cultures of AG 2-2 IIIB ranged in root rot ratings from 42 to 100 (Table 1) and 10 of the 24 cultures were very aggressive and caused root rot ratings of >90 to 100 (Fig. 2A). The non-inoculated control was disease-free and averaged a rating of 0.8. Cultures of *R. solani* were isolated from 85% of sugarbeet seedlings in *Rhizoctonia*-infested soil and were not isolated in the control.

Beans. On both pinto bean and soybean, *R. solani* AG 2-2 IIIB was significantly more aggressive in causing basal stem rot and root rot than AG 2-2 IV (Table 1). Disease ratings (1 to 5 scale) for pinto bean and soybean averaged 4.4 and 4.1 for AG 2-2 IIIB, respectively, and 2.7 and 3.2 for AG 2-2 IV, respectively. On both bean crops, individual cultures of *R. solani* AG 2-2 IV and AG 2-2 IIIB ranged from low to high aggressiveness in causing disease (Table 1). On pinto bean, 19 cultures of AG 2-2 IIIB and only 3 cultures of AG 2-2 IV caused disease ratings >4-5; no cultures of AG 2-2 IIIB caused disease ratings less than 3, whereas 16 cultures of AG 2-2 IV caused disease ratings of 3 or less (Fig. 3A). On soybean, 15 cultures of AG 2-2 IIIB and 3 cultures of AG 2-2 IV caused disease ratings of >4-5; three cultures of AG 2-2 IIIB caused disease in the >2-3 category, whereas 10 cultures of AG 2-2 IV caused disease in this category (Fig. 3B). Cultures of *R. solani* were isolated from 88% of pinto beans and 54% of soybean basal stems in inoculated soil; the fungus was not isolated in non-inoculated controls.

Table 1. Pathogenicity of 24 cultures of *Rhizoctonia solani* AG 2-2 IV and 24 cultures of AG 2-2 IIIB on seedlings and adult plants of sugarbeet and several rotation crops. Each value is averaged across two trials of the 24 cultures (range = minimum to maximum disease ratings for individual cultures).

Crop (Disease scale)	Average disease ratings (range: minimum-maximum) — Seedlings[u]			Adult plants[v]		
	Control	AG 2-2 IV	AG 2-2 IIIB	Control	AG 2-2 IV	AG 2-2 IIIB
Sugarbeet (0-100 scale)[w] adults (0-7 scale)	0.8	51 a (5-100)	78 b (42-100)	0.3	5.0 a (3.3-5.6)	4.9 a (3.8-5.9)
Pinto bean (1-5 scale)[x]	1.0	2.7 a (1.8-4.5)	4.4 b 3.5-5.0)	1.0	2.9 a (2.5-3.4)	3.1 b (2.3-3.8)
Soybean (1-5 scale)[x]	1.0	3.2 a (1.9-4.6)	4.1 b (2.7-5.0)	1.0	3.5 a (3.0-4.3)	3.5 a (2.3-4.1)
Sunflower (1-5 scale)[x]	1.0	1.3 a (1.0-1.8)	1.9 b (1.0-4.0)	1.0	2.3 a (1.5-3.4)	2.8 b (1.0-3.6)
Wheat (0-3 scale)[y]	0.0	0.7 a (0.4-1.1)	1.0 b (0.5-1.6)	0.0	0.1 a (0.0-0.7)	0.3 b (0.1-0.7)
Corn (1-5 scale)[z]	1.0	2.1 a (1.2-3.0)	3.1 b (1.8-4.1)	1.0	1.5 a (1.1-2.6)	2.1 b (1.3-2.6)
Sweet corn (1-5 scale)[z]	1.0	2.2 a (1.2-4.3)	2.7 b (1.2-4.3)	1.1	2.1 a (1.6-2.5)	2.6 b (1.9-3.4)

[u] For seedlings of each crop, values followed by the same letter are not significantly different (*P*=0.05); sunflower seedling trial has been conducted once.

[v] For adult roots of each crop, values followed by the same letter are not significantly different (*P*=0.05).

[w] Sugarbeet seedling root rot index = 0 to 100 scale, 0 = no disease, 100 = all plants dead at 4 weeks after planting; adult root rot index 0 to 7 scale, 0 = no disease, 7 = root completely rotted and plant dead.

[x] Pinto bean, soybean, and sunflower (seedlings and adult plants) rated on a 1 to 5 scale, 1 = no symptoms, 5 = shoot dead with 75-100% of stem girdled (7).

[y] Wheat (seedlings and adult plants) subcrown internodes rated on a 0 to 3 scale, 0 = healthy and 3 = more than 50% of surface with lesions and discoloration (11).

[z] Corn and sweet corn (seedlings and adult plants) rated on a 1 to 5 scale, 1 = <2% root surface rotted and 5 = plant dead (10).

Sunflower. Cultures of *R. solani* AG 2-2 IIIB were not very aggressive, but caused significantly more disease than AG 2-2 IV and averaged disease ratings of 1.9 and 1.3, respectively (Table 1, 1 to 5 scale). Only 5 cultures of AG 2-2 IIIB caused root rot ratings between >3-4 and 18 caused low disease ratings of 1-2 (Fig. 3C). On the other hand, all 24 cultures of AG 2-2 IV caused negligible disease with ratings of 1-2 (Fig 3C). Cultures of *R. solani* where not isolated from sunflower seedlings in the *Rhizoctonia*-infested soil or the control.

Wheat. Overall, disease levels were relatively low, but *R. solani* AG 2-2 IIIB caused significantly more disease than AG 2-2 IV and averaged ratings of 1 and 0.7, respectively (Table 1). Individual cultures of AG 2-2 IV averaged disease ratings of 0.4 to 1.1 and AG 2-2 IIIB averaged ratings from 0.5 to 1.6 (Table 1). Twenty-two cultures of AG 2-2 IV and 14 of AG 2-2 IIIB resulted in virtually no disease (0-1 category, Fig. 4A). Two cultures of AG 2-2 IV and 10 of AG 2-2 IIIB caused ratings >1-2, where 25 to 50% of subcrown internodes had lesions and were discolored (Fig. 4A). *R. solani* was re-isolated from 96% of subcrown internodes in inoculated soil and not isolated in the non-inoculated control.

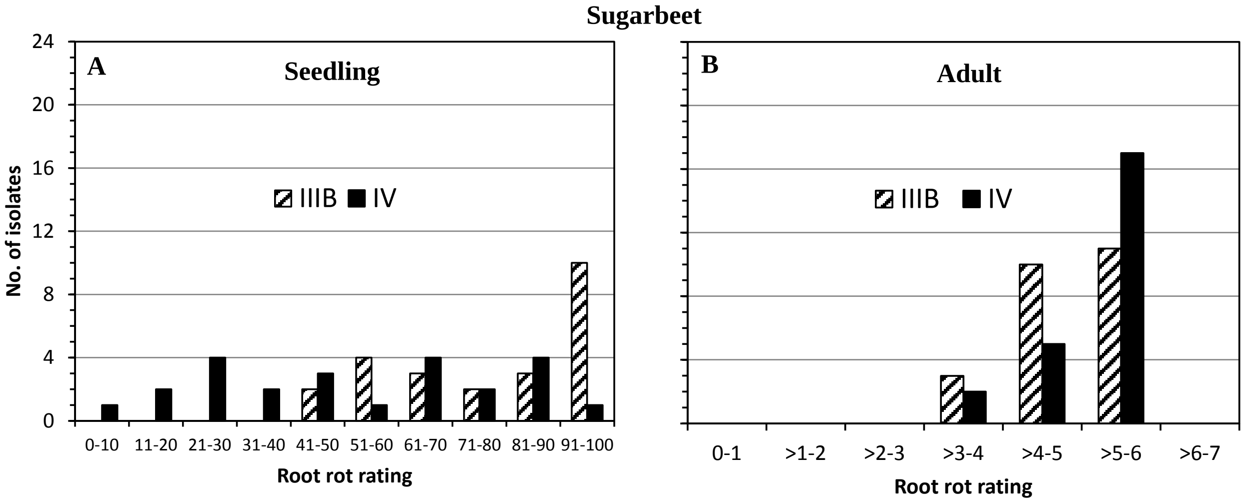

Fig. 2. Number of cultures of *Rhizoctonia solani* AG 2-2 IV (24 cultures) and AG 2-2 IIIB (24 cultures) in various root rot disease categories based on pathogenicity tests. **A)** **<u>Sugarbeet Seedling trials</u>:** a root rot rating from 0-10 denotes healthy roots; disease severity increases with increasing higher intervals and at 90-100, plants are dead. **B)** **<u>Sugarbeet Adult plant trial</u>:** a root rot rating of 0 denotes healthy roots; disease severity increases with increasingly higher root rot values and at a value of 7, roots are completely rotted and foliage is dead. Data for each culture are averaged across two trials, four replicates per trial.

<u>Field corn</u>. Significantly more root rot was caused by *R. solani* AG 2-2 IIIB than by AG 2-2 IV, resulting in average disease ratings of 3.1 and 2.1, respectively (Table 1, 1 to 5 scale). Individual cultures of AG 2-2 IIIB had a range of disease ratings from 1.8 to 4.1 (Table 1) and 14 cultures (58%) resulted in disease ratings >3-4 and >4-5 (Fig. 4B). Individual cultures of AG 2-2 IV resulted in a range of disease ratings from 1.2 to 3.0 (Table 1). Twelve cultures of AG 2-2 IV (50%) had very low disease ratings of 1-2; 9 cultures (37%) had ratings of >2-3; and only 2 cultures (8%) resulted in ratings >3-4 (Fig. 4B). Cultures of *R. solani* were isolated from 100% of plants in *Rhizoctonia*-infested soil and were not isolated in the non-inoculated control.

<u>Sweet corn</u>. *R. solani* AG 2-2 IIIB caused significantly more root rot than AG 2-2 IV (Table 1) and ratings averaged 2.7 and 2.3, respectively. Individual cultures of both ISGs, however, spanned the same range of root rot values from 1.2 to 4.3 (Table, 1 to 5 scale). Individual cultures of AG 2-2 IV tended to dominate the lower root rot rating categories (1-2 and >2-3) while AG 2-2 IIIB spanned all disease categories and 37, 17 33, and 13% of cultures fell into the 1-2, >2-3, >3-4, and >4-5 disease categories, respectively (Fig. 4C). Cultures of *R. solani* were isolated from roots of 17% of sweet corn seedlings in *Rhizoctonia*-infested soil and were not isolated in the control.

<u>Adult Plant Pathogenicity Tests:</u>

<u>Sugarbeet.</u> Both ISGs were equally aggressive in causing RCRR on adult plants at 12 days after inoculation (P = 0.1002, Table 1). Ratings for RCRR (0 to 7 scale) for AG 2-2 IV averaged 5.0 (ranged from 3.3 to 5.6) and for AG 2-2 IIIB averaged 4.9 (ranged from 3.8 to 5.9) (Table 1). All cultures resulted in disease ratings >3 and cultures of AG 2-2 IIIB predominated in the >3-4 and >4-5 categories, but AG 2-2 IV predominated in the >5- 6 category (Fig. 2B). The non-inoculated control was disease-free and averaged a rating of 0.3. Cultures of *R. solani* were isolated from 96% of roots in inoculated soil and were not isolated in the non-inoculated control.

<u>Pinto bean</u>. The AG 2-2 IIIB population was statistically more aggressive in causing stem and root rot than AG 2-2 IV, but average values were nearly identical for both ISG's (Table 1). The average disease rating for AG 2-2 IIIB was 3.1 (range from 2.3 to 3.8) and for AG 2-2 IV was 2.9 (range from 2.5 to 3.4). Individual cultures of AG 2-2 IV predominated the >2-3 disease category while cultures of AG 2-2 IIIB predominated the >3-4 category (Fig. 3D). No cultures of either ISG were in the 1-2 or >4-5 disease categories. Cultures of *R. solani* were isolated from 50% of basal stems in inoculated soil and were not isolated in the non-inoculated control.

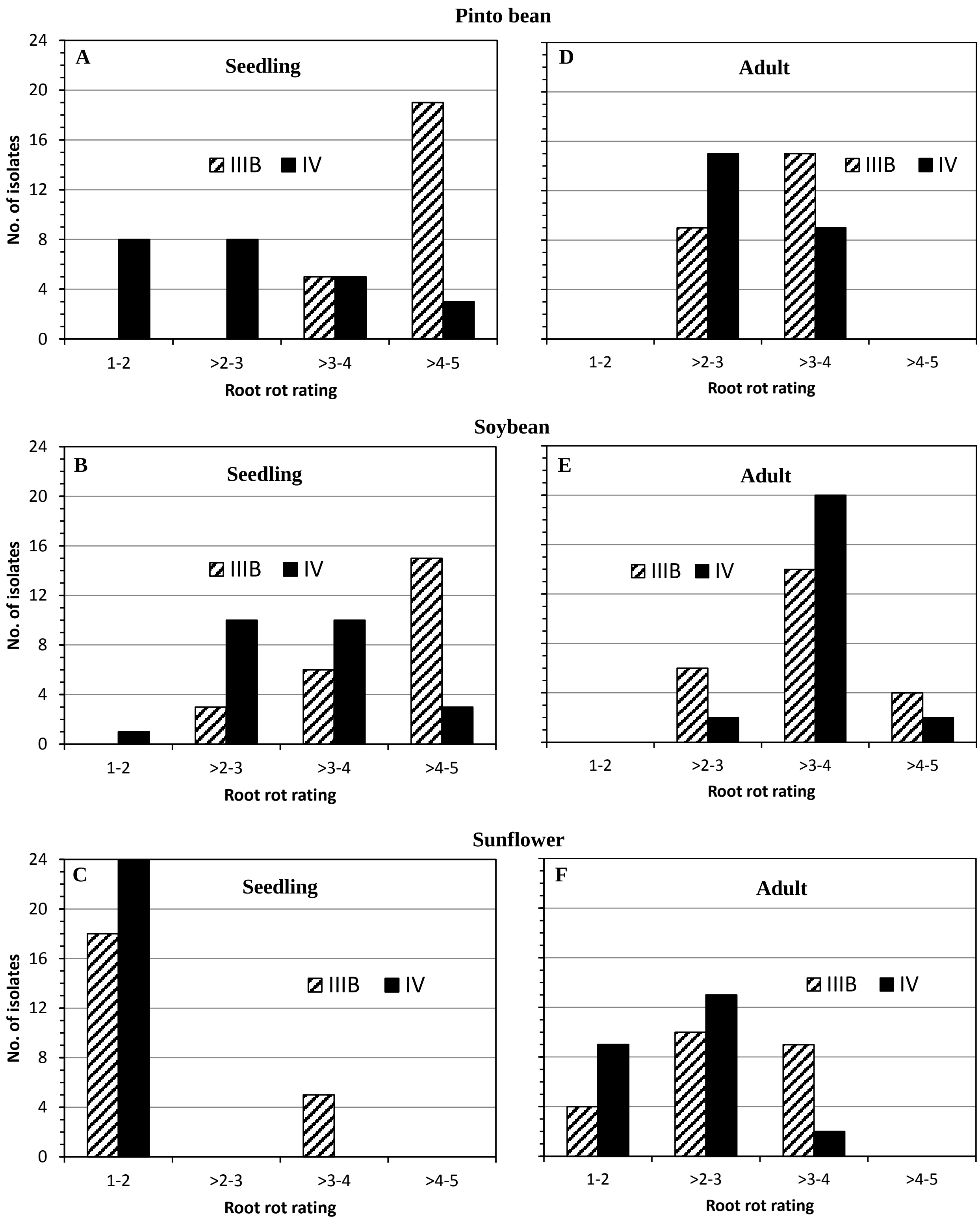

Fig. 3. Number of cultures of *Rhizoctonia solani* AG 2-2 IV (24 cultures*)* and AG 2-2 IIIB (24 cultures) in various root rot disease categories based on pathogenicity tests **Seedling trials** on **A)** pinto bean, **B)** soybean, and **C)** sunflower. **Adult plant trials** on **D)** pinto bean, **E**) soybean, and **F)** sunflower. A root rot rating of 0 denotes healthy roots; disease severity increases with increasing higher intervals and at 5, plants are dead or severely rotted. Data for each culture are averaged across two trials, four replicates per trial.

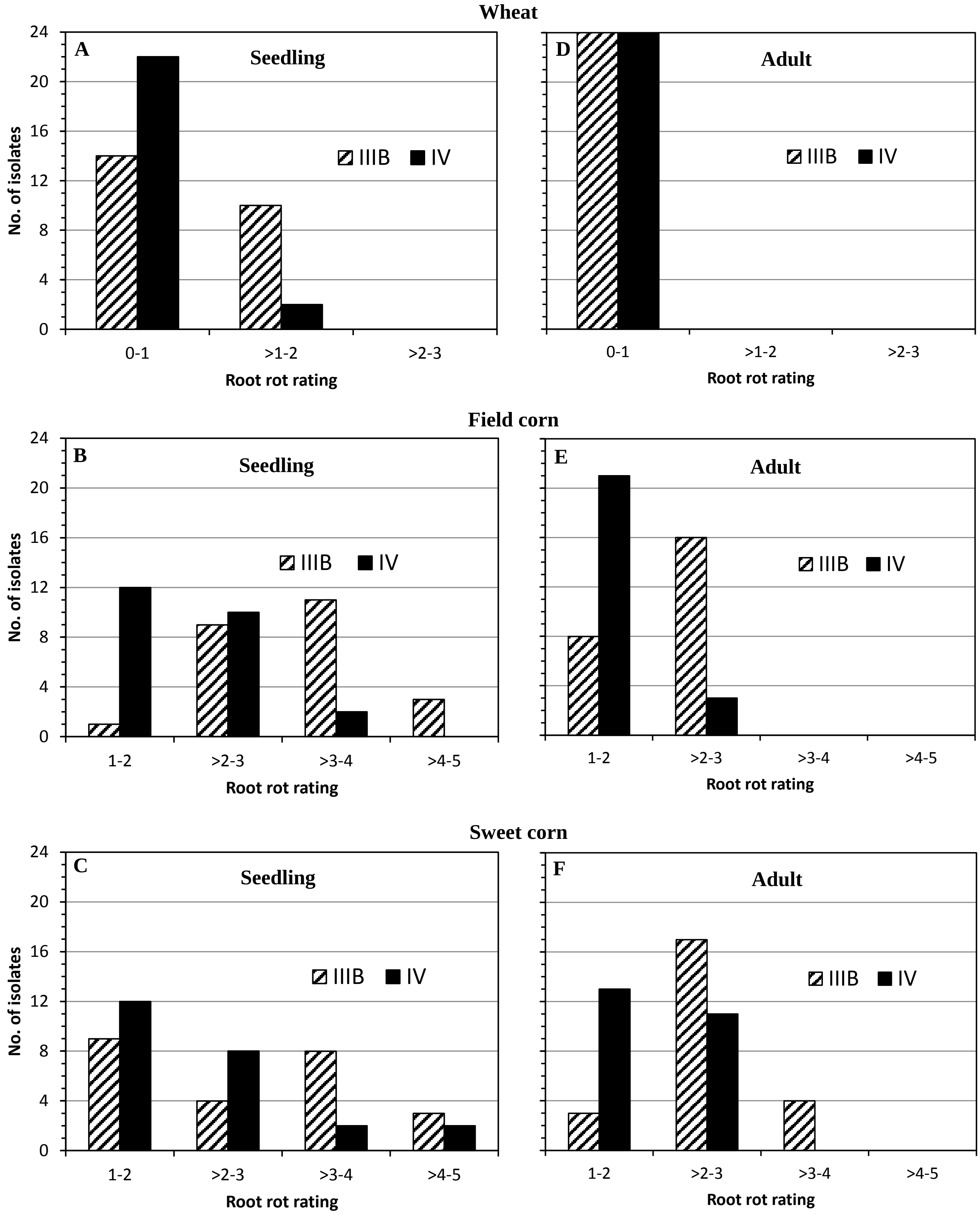

Fig. 4. Number of cultures of *Rhizoctonia solani* AG 2-2 IV (24 cultures) and AG 2-2 IIIB (24 cultures) in various root rot disease categories based on pathogenicity tests. **Seedling trials** on **A)** wheat, **B)** field corn, and **C)** sweet corn. **Adult plant trials** on **D)** wheat, **E)** field corn, and **F)** sweet corn. A root rot rating of 0 denotes healthy roots; disease severity increases with increasing higher intervals and at 3 (for wheat) or 5 (for field and sweet corn), plants are dead or roots severely rotted. Data for each culture are averaged across two trials, four replicates per trial.

Soybean. *R. solani* AG 2-2 IV and AG 2-2 IIIB were equally aggressive in causing stem and root rot and averaged the same disease rating of 3.5 (Table 1, 1 to 5 scale). Cultures within AG 2-2 IV ranged in disease ratings from 3 to 4.3 and within AG 2-2 IIIB ranged from 2.3 to 4.1 (Table 1). The >3-4 disease category accounted for most of the cultures, including 83% of AG 2-2 IV and 58% of AG 2-2 IIIB (Fig. 3E). There were more cultures of AG 2-2 IIIB than AG 2-2 IV in the >2-3 and 4-5 categories (Fig. 3E). *R. solani* was isolated from 56% of basal stems in *Rhizoctonia*-inoculated soil and was not isolated in the non-inoculated control.

Sunflower. Overall, *R. solani* AG 2-2 IIIB was significantly more aggressive than AG 2-2 IV, averaging disease ratings of 2.8 (ranged from 1 to 3.6) and 2.3 (ranged from 1.5 to 3.4), respectively (Table 1, 1 to 5 scale). Within AG 2-2 IV, 38, 54, and 8% of cultures fell in the 1-2, >2-3, and >3-4 disease categories, respectively (Fig. 3F). By comparison, 17, 43, and 39% of AG 2-2 IIIB cultures fell into the 1-2, >2-3, and >3-4 disease categories, respectively (Fig. 3F). Cultures of *R. solani* were isolated from 67% of basal stems in inoculated soil and were not isolated in the control.

Wheat. Disease ratings for AG 2-2 IV and AG 2-2 IIIB were extremely low and averaged 0.3 and 0.1, respectively (Table 1, scale 0 to 3). All cultures of both ISGs fell into the 0-1 disease category (Fig. 4D). In this case, statistical significance was inconsequential because the cultures caused negligible or no disease. Cultures of *R. solani* were isolated from 80% of subcrown internodes in inoculated soil and were not isolated in the non-inoculated control.

Field corn. Cultures of AG 2-2 IIIB were significantly more aggressive than AG 2-2 IV and averaged disease ratings of 2.1 and 1.5, respectively (Table). Individual cultures of AG 2-2 III ranged in disease ratings from 1.3 to 2.6 (Table 1) and most isolates (67%) were in the >2-3 category (Fig. 4E). Cultures of AG 2-2 IV ranged in disease ratings from 1.1 to 2.6 (Table 1) and most cultures (87%) were in the 1-2 disease category (Fig. 4E). Cultures of *R. solani* were isolated from roots of 94% of adult corn plants in inoculated soil and were not isolated in the control.

Sweet corn. This crop was slightly more susceptible to root rot than field corn (Table 1) and cultures of AG 2-2 IIIB were significantly more aggressive than AG 2-2 IV and averaged disease ratings of 2.6 and 2.1, respectively (Table 1). Individual cultures of AG 2-2 IIIB ranged in disease ratings from 1.9 to 3.4 (Table 1) and most (71%) fell into the >2-3 category (Fig. 4F). Individual cultures of AG 2-2 IV ranged in disease ratings from 1.6 to 2.5 (Table 1) and 54% were in the low disease category of 1-2 and 46% were in the >2-3 category (Fig. 4F). Cultures of *R. solani* were isolated from 56% of roots from adult sweet corn plants in the inoculated soil and were not isolated in the control.

DISCUSSION

The data reported here are the first to document uniform aggressiveness of cultures of *R. solani* AG 2-2 IV and 2-2 IIIB in causing severe RCRR on adult sugarbeet roots. This is in contrast to Panella (6), who determined *R. solani* AG 2-2 IIIB was more aggressive than AG 2-2 IV in causing RCRR on sugarbeet. We isolated all cultures from sugarbeet roots/crowns with RCRR and both ISGs produced identical symptoms. In 1987, Ogoshi (5) reported AG 2-2 IV caused RCRR of sugarbeet while AG 2-2 IIIB caused root diseases on a wide range of other crops including beans. Occurrence of both AG 2-2 IV and AG 2-2 IIIB on sugarbeet in the United States (1,6,9) and AG 2-2 IIIB in Europe (2,4) indicates that AG 2-2 IIIB is becoming a predominate pathogen of sugarbeet. This trend may reflect more intensive surveying and understanding of RCRR in the last two decades, but also suggests possible shifts in ISGs associated with long-term cultivation of sugarbeet and susceptible rotation crops. Both ISGs are pathogenic on adult sugarbeet roots, so this may account for increases in prevalence of RCRR worldwide.

Rotation crops varied in susceptibility to *R. solani* AG 2-2 but bean crops (on seedlings and adult plants) were most susceptible. Seedlings of other rotation crops in descending order of susceptibility were: corn (field and sweet corn), sunflowers, and wheat. For adult plants, bean crops were most susceptible, followed in descending order of susceptibility by sunflower, corn (sweet and field), and wheat. Overall, *R. solani* AG 2-2 IIIB was more aggressive than AG 2-2 IV on seedlings of sugarbeet and all rotation crops. When looking at aggressiveness of individual cultures, however, there were some cultures of AG 2-2 IV that caused more disease on seedlings and adult plants than AG 2-2 IIIB. And conversely, some cultures of AG 2-2 IIIB caused lower disease ratings than AG 2-2 IV. Thus, isolation of AG 2-2 IIIB from sugarbeet does not necessarily mean the culture will be highly pathogenic to rotation crops, although it may be. The least susceptible/non-host rotation crop was wheat, especially adult plants.

Cultures of *R. solani* were isolated from superficial, external tissues of wheat epidermis at nearly 100%, but this delicate tissue decomposes quickly in soil, and then *R. solani* dies.

In general, disease ratings tended to be lower on adult plants than seedlings of some rotation crops (pinto bean, soybean, sunflower, field corn, wheat). This may be accounted for by the greater susceptibility of seedlings to root infection because of immature tissue. As plants age, roots develop more resistance to root pathogens. Seedlings and adult roots of sweet corn were about equal in susceptibility to *R. solani*. Ideally, varieties of each rotation crop should be screened for susceptibility to *R. solani* AG 2-2 IV and AG 2-2 IIIB because there likely is variability in susceptibility to the pathogen.

Overall, planting back-to-back crops susceptible to *R. solani* AG 2-2 is ill-advised and should be avoided. Hard red spring wheat is an important crop in the sugarbeet rotation because it essentially is a non-host to *R. solani* AG 2-2.

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