Characterization of *Rhizoctonia* Populations Obtained from Sugarbeet Fields with Differing Soil Textures

L. J. Herr and D. L. Roberts

Professor and former research assistant, respectively, Department of Plant Pathology, Ohio Agricultural Research and Development Center, Wooster 44691 and The Ohio State University, Columbus 43210.

This research was partially supported by Ohio Sugar Beet Grower's Associations and Processors. In part based on a portion of a MS thesis submitted to The Ohio State University, Columbus by D. L. Roberts.

Approved for publication as Journal Series Article 140-79 of the Ohio Agricultural Research and Development Center, Wooster.

The authors thank James Holman, OARDC Statistician, for statistical advice and analyses.

Accepted for publication 15 November 1979.

ABSTRACT

HERR, L. J., and D. L. ROBERTS. 1980. Characterization of *Rhizoctonia* populations obtained from sugarbeet fields with differing soil textures. Phytopathology 70:476-480.

Representative *Rhizoctonia* cultures were selected from populations isolated from soil and from weed hosts in areas of apparently healthy and crown- and root-rot diseased beets. The four sugarbeet fields assayed during the summer and fall of 1976 differed in soil textures (sandy loam, fine sandy loam, silt loam, and silty clay loam). The selected isolates were distinguished by cultural characteristics, anastomosis grouping, numbers of nuclei, sugarbeet seedling pathogenicity assays, and older sugarbeet plant pathogenicity assays. Multinucleate and binucleate isolates could not be distinguished by cultural characteristics. Most multinucleate isolates were of two anastomosis groups, AG-2 and AG-4. In fine textured soils AG-2 isolates predominated, whereas in the coarser textured soils numbers of AG-2 and AG-4 isolates were nearly equal in one soil and AG-4 isolates predominated in the other. AG-4 isolates were significantly more virulent to sugarbeet seedlings than were AG-2 isolates; whereas AG-2 isolates were significantly more virulent than were AG-4 isolates on (6- to 8-wk-old) plants. Binucleate isolates were avirulent to weakly virulent on sugarbeets in both assays. Most binucleate isolates were obtained from weeds.

Recently, we reported on soil populations of *Rhizoctonia solani* obtained from areas of healthy and areas of crown rot- and root rot-diseased beets within four sugarbeet fields differing in soil texture (19). This paper is an extension of that earlier research. Its principal purpose is to characterize by biologically meaningful criteria the previously described *Rhizoctonia* populations (19). Isolates from weed hosts have been included because of their probable role in supporting and perpetuating *R. solani* populations in soil (1,3).

According to current concepts, *R. solani* Kühn (*Thanatephorus cucumeris* [Frank] Donk) consists of four hyphal anastomosis groups designated AG-1, AG-2, AG-3, and AG-4 (15,24). Japanese research workers (9,13) have described additional anastomosis groups, but these have not yet been reported outside of Japan. According to Sherwood (24), each anastomosis group is characterized by distinctive morphologic, pathologic, and physiologic norms, with some overlapping of isolates among groups. On sugarbeets (*Beta vulgaris* L.), *R. solani* may cause several different disease syndromes (ie, damping off, root rot, crown rot, or foliage blight) depending on the pathological, physiological, and morphological attributes of the particular isolates involved (8,10,23). In Ohio, crown and root rot of sugarbeets are the most important syndromes (5,6,19). Ruppel (20) reported that all isolates of *R. solani* obtained from sugarbeet seedlings, crowns, and foliage belonged in AG-4, whereas all root isolates were in AG-2. Moreover, crown and foliage isolates caused significantly more foliage blight than did root isolates, and root isolates caused more severe root rot than did foliar or crown isolates. Significant seedling damping-off was caused by all isolates of both AG-2 and AG-4. In addition, Ruppel (20) stated that the association between cultural characteristics, anastomosis groups, and pathological activity made it possible to distinguish foliar and crown isolates from root isolates by either cultural characteristics or anastomosis tests.

Criteria for distinguishing *R. solani* from other fungi closely resembling it (17) include the presence of a prominent, doliform septal pore apparatus (which distinguishes basidiomycetes from nonbasidiomycetes) and multinucleate vegetative hyphal cells (which aids in distinguishing multinucleate *R. solani* from binucleate *Rhizoctonia*-like fungi, which are similar in cultural appearance, mycelial morphology, and habitat) (17). Certain of these *Rhizoctonia*-like binucleate fungi are pathogenic on turfgrass and some additional plants (21).

Our objectives were to characterize *Rhizoctonia* isolates obtained from soil and weeds by cultural characteristics, anastomosis groupings, numbers of nuclei, sugarbeet pathogenicity assays on seedlings and on older sugarbeet plants, and to relate these findings to the ecology and epidemiology of *R. solani* in the field.

MATERIALS AND METHODS

Sources and isolation of *Rhizoctonia* cultures. In July 1976, four sugarbeet fields affected with Rhizoctonia root- and crown-rot disease and differing in soil textures were selected. The soil series names, textural classes, mechanical analyses, and pHs were: Bellmore sandy loam (SL)–30.3% sand, 52.2% silt, 17.5% clay, pH 6.7; Digby fine sandy loam (FSL)–41.5% sand, 45.7% silt, 12.8% clay, pH 7.1; Toledo silt loam (SiL)–27.2% sand, 43.2% silt, 29.6% clay, pH 7.0; and Toledo silty clay loam (SiCL)–7.4% sand, 55.5% silt, 37.1% clay, pH 7.0. More complete analyses of soil chemical factors and cropping histories have been published elsewhere (19). Within each field, four areas of beets affected by root- and crown-rot disease (DA) and four areas of apparently healthy beets (AH) were chosen and plotted. Monthly soil samples from these areas were assayed for *R. solani* (19) by a garden beet seed colonization method. The soil sampling and assay procedures used were described previously (18). One hundred twenty-six cultures, presumed to be *R. solani*, were collected from sugarbeet field soils during the summer and fall of 1976 by hyphal tip isolation from representative colonies. In addition, 40 cultures were isolated from weeds, volunteer corn, and tomato plants in AH and DA of the fields during summer and fall of 1976. Small pieces of root and stem (usually bearing distinct lesions) were washed in a sieve nested in a beaker under cold, running tap water with several drops of Tween 20 (ICN, Cleveland, OH 44128) wetting agent added to improve washing, and then plated on 2% water agar (WA). Cultures were hyphal tip transferred and maintained on potato dextrose agar supplemented with 1 g of yeast extract (PDA-YE) per liter (20).

0031-949X/80/06047605/$03.00/0
©1980 The American Phytopathological Society

Cultural and anastomosis groupings. Before anastomosis groups were determined by actual pairing, the cultural characteristics of isolates grown for 2 wk on PDA-YE plates were ascertained. These cultural types were then tentatively assigned to specific anastomosis groups by Ruppel's method (20).

All cultures were paired with known anastomosis group (AG) test cultures (courtesy of Neil Anderson, Dept. of Plant Pathology, University of Minnesota, St. Paul) for actual hyphal anastomosis studies. Pairings were made on sterile glass slides coated with 2% WA and placed on WA plates to maintain moisture. A rectangular mycelial fragment (~1.5 cm × 0.5 cm) of an unknown isolate was taken from a rapidly growing colony on PDA-YE and placed parallel to and 2–3 cm from a similar agar piece of a known AG culture on an agar-coated slide. After the intermingling of hyphae (3–5 days of growth at 24 C), slides were stained with cotton blue in dilute lactophenol and microscopically examined for hyphal anastomoses (15).

Septal pores and numbers of nuclei. Characteristics of the septal pore apparatus and numbers of nuclei in vegetative cells of all cultures were ascertained by staining and microscopic examination. Difco potato dextrose agar (Difco Laboratories, Detroit, MI 48232) cultures were stained by two modified rapid staining procedures with 0.5% aniline blue (25) and 0.05% trypan blue (21), in addition to the HCl-Giemsa nuclear stain for fungi (7).

Pathogenicity and virulence. All cultures were tested for pathogenicity and relative virulence on sugarbeet seedlings by an adaptation of Schmitthenner and Hilty's (22) inoculum layer technique. The soil mixture used was an autoclaved mixture of soil, peat, and perlite (1:1:1). Styrofoam cups (Dart® 16M20, Dart Container Corp., Mason, MI 48854) of 535-ml capacity were perforated with three bottom holes for drainage and used as containers. Four hundred cubic centimeters of soil mixture were put into each cup, leveled, and a completely colonized 2% WA agar layer of inoculum from a petri dish culture was added. The inoculum layer was covered with 50 cc of the soil mixture, then seeded with 25 sugarbeet seeds (cultivar US H20) and covered with a final 50 cc of the soil mixture. A randomized complete block design was used with four replicates of each isolate. Seedling assays were made in a greenhouse maintained at about 26 C. Numbers of seedlings surviving were recorded after 3 wk.

The pathogenicity and relative virulence of 32 selected *Rhizoctonia* isolates to older (6- to 8-wk-old) plants also were investigated. Each replicate consisted of seven 15-cm-diameter pots of autoclaved soil mixture each with a single sugarbeet plant (cultivar US H20) per pot. In a series of tests, three replicates per isolate were arranged in randomized block design on greenhouse benches (temperature ~26 C, with ~8,500 lx supplemental lighting 6 hr daily). Uninoculated controls and reference isolates were included in individual tests.

Inoculum for these tests was increased in stationary cultures in 25 ml of Czapek's Dox broth, plus 0.1% yeast extract, in 250-ml flasks, grown for 1 wk at 24 C. Harvested mycelium was washed with tap water, drained, weighed, and then comminuted (two 15 sec intervals) in 100 ml of tap water in a blender. Final inoculum concentrations were adjusted to concentrations of 0.1 g (summer tests) or 0.2 g (winter tests) of mycelium (fresh wt) per milliliter of water.

Soil in the pots was infested by placing 5 ml of mycelial suspension into each of three holes (approximately 1 cm in diameter × 2.5 cm deep) made in the soil of each pot approximately 2.5 cm from the beet root. An additional 200 cc of autoclaved soil mixture was added to each pot. After 3–4 wk of incubation, the beets were removed from the pots, washed, and rated on a 0–5 disease rating scale (0 = healthy, 5 = dead).

RESULTS

Cultural types and anastomosis groupings. Isolates with appressed, tan-white or tan to silver-gray-brown mycelium and, generally, inconspicuous sclerotia after 2 wk of growth on PDA-YE were designated as cultural type 4 (CT-4), and probable AG-4 types. These CT-4 isolates were distinguished from isolates with abundant light-brown to dark-brown aerial mycelium and, usually, abundant large brown sclerotia (2,20,24), which were designated CT-2 and probable AG-2. Comparisons of these culturally determined probable anastomosis groups with actual anastomosis groups ascertained by pairings with known isolates and determinations of numbers of nuclei indicated that, when classification was based on cultural characteristics alone, 14% of the isolates were incorrectly assigned. Of all isolates, 10% were binucleate (nine CT-2 and eight CT-4). Thus, binucleate *Rhizoctonia*-like isolates can appear to be either CT-2 or CT-4, which emphasizes the importance of nuclear staining. In the case of multinucleate isolates, one CT-4 isolate was actually an AG-2; whereas, six CT-2 were found to be AG-4. Further, six multinucleate isolates failed to anastomose with known AG isolates (five CT-2 and one CT-4) and could only be classified culturally.

The multinucleate isolates obtained from soil and weeds, which anastomosed with known AG reference cultures, fell into either of two groups, AG-2 or AG-4. In nearly all cases, unknown isolates belonging in AG-4 anastomosed readily with the standard AG-4 reference culture, which facilitated their assignment to the proper anastomosis group. The unknowns found to belong in AG-2 were more difficult to distinguish, because only some of the isolates anastomosed readily with the standard AG-2 reference culture. For our purposes, those isolates were designated as primary isolates. We then paired these primary isolates with other unknowns and classified those that anastomosed with the primary isolates as secondary isolates. By using three known isolates (ie, the standard AG-2, a primary isolate, and a secondary isolate) most unknowns were assignable to the proper anastomosis group.

Of 166 cultures presumed to be *R. solani,* 23 did not anastomose with any of the four known AG reference cultures or the primary and secondary AG-2 cultures. Six of the 23 were multinucleate fungi that fitted the criteria of Parmeter and Whitney (16) for *R. solani.* The remaining 17 were binucleate fungi (not *R. solani* [16]), which did not anastomose with known *R. solani* AG cultures, although culturally they were virtually indistinguishable from *R. solani* (17). Ten of the 17 binucleate isolates were obtained from weed hosts (25% of all weed isolates were binucleate). Most (60%) of these binucleate isolates were from either giant foxtail (*Setaria faberi* Herrm.) or yellow foxtail (*Setaria lutescens* [Weigel] Hubb.), and all the isolates obtained from both foxtail grasses were binucleate.

Distribution of anastomosis groups. Within fields there were no discernible differences in the distribution of specific anastomosis groups between AH and DA; however, major differences were found in the occurrence and distribution of specific anastomosis groups among fields having different soil textures (Fig. 1). *R. solani* populations in the fine-textured SiL and SiCL soils consisted predominately of AG-2 isolates; whereas, in the coarser-textured soils, SL had nearly equal percentages of AG-2 and AG-4 isolates, and FSL populations consisted primarily of AG-4 isolates. In one of the fields (SiL) only AG-2 was found. In the remaining three fields both AG-2 and AG-4 were found, and the differences in AG distribution among these fields were due to differences in frequency of isolation of these two specific anastomosis groups.

Pathogenicity and virulence. Significant differences were found in the relative virulence of the 166 *Rhizoctonia* isolates to sugarbeet seedlings. The isolates ranged from avirulent (all 25 seedlings surviving) to highly virulent (no surviving seedlings). (Mean = 10.70, $LSD_{(P=0.05)}$ = 5.04). The data were organized into five virulence rating groups (0, I, II, III, and IV; representing 21–25, 16–20, 11–15, 6–10, and 0–5 surviving seedlings, respectively), and the percentages of isolates in each virulence rating group were recorded for specific categories of isolates (Table 1). Within fields, isolates from healthy areas (AH) and diseased areas (DA) generally were quite similar in relative virulence, whereas among fields marked differences in relative virulence were evident. The FSL soil had the highest percentage of group IV (highly virulent) isolates, SiL had none, and the two remaining fields (SiCL and SL) had intermediate percentages of highly virulent isolates. These results were related to the distribution of anastomosis groups among fields (Fig. 1). Thus,

a greater percentage of AG-4 isolates were highly virulent to sugarbeet seedlings than were present among AG-2 isolates. Binucleate isolates essentially were nonpathogenic to sugarbeet seedlings. Weed isolates, which included a large portion of binucleate isolates, tended to be less virulent on sugarbeet seedlings than were isolates from soil.

The relative virulence of 32 selected *Rhizoctonia* isolates on older plants gave quite different results from those obtained with the same 32 isolates in seedling tests (Table 2). In Table 2, the isolates were ranked according to relative virulence and, in both assays, the data spanned a continuum ranging from avirulent to highly virulent. Isolates within anastomosis groups mostly were grouped together rather than being randomly dispersed throughout the entire virulence range in both assays. The collective means data indicated that AG-4 isolates were significantly more virulent to sugarbeet seedlings than were AG-2 or binucleate isolates, which did not differ significantly. In contrast, AG-2 isolates were significantly more virulent than either AG-4 or binucleate isolates, and AG-4 isolates were significantly more virulent than were binucleate isolates in assays on older plants. Binucleate isolates were essentially avirulent on older sugarbeet plants. A correlation coefficient for comparison of all 32 isolates (regardless of grouping) in seedling asssays versus older plant assays was nonsignificant ($r = 0.15$), indicating no relationship between virulence on seedlings and virulence on older plants.

DISCUSSION

Our inability to distinguish between binucleate isolates (definitely not *R. solani*) and multinucleate isolates (probably *R. solani*) by cultural appearance was a serious shortcoming of the cultural typing method. In no case could we with certainty differentiate between binucleate and multinucleate isolates without nuclear staining. This agrees with the conclusion of Parmeter et al (17) that binucleate and multinucleate *Rhizoctonia*-like isolates cannot be separated by cultural appearance. Moreover, the misidentification of the anastomosis groupings of several multinucleate isolates by cultural methods, resulting from the overlapping of cultural characteristics among groups (24), demonstrated the need for anastomosis pairings. Because collections of unknown *Rhizoctonia* -like isolates may well include binucleate isolates and culturally overlapping multinucleate isolates, we believe grouping of isolates by cultural characteristics should be limited to initial screening, preparatory to more precise characterization by determinations of number of nuclei and anastomosis pairings.

The lack of cultural and morphological distinctions between binucleate and multinucleate *Rhizoctonia*-like fungi may affect the accuracy of inoculum density assays based on recognition of *R. solani* colonies by appearance and morphology in isolation plates. The relative proportions of binucleate and multinucleate propagules in the populations being assayed also should be ascertained. Correction for the binucleate proportions of populations can then be made, if deemed necessary.

In our study, only 5.5% of the isolates obtained from soil by the beet seed colonization method were binucleate. More were from AH than from DA. The majority of binucleate isolates were from weeds and most of these were from two species of foxtail grass that yielded only binucleate isolates. The finding that weeds may exhibit selectivity towards binucleate or multinucleate fungi emphasizes the importance of determining numbers of nuclei in isolates obtained from weeds in studies of the role of weeds in the ecology and epidemiology of *R. solani.*

All except six of our multinucleate isolates anastomosed with known AG reference cultures of *R. solani.* According to Parmeter et al (17), successful anastomosis strongly indicates that the paired isolates are closely related; however, failure to anastomose, of itself, does not necessarily indicate a lack of relationship. The six nonanastomosing multinucleate isolates appeared to be *R. solani,* both morphologically and culturally. Because five of these six isolates were CT-2 and some AG-2 isolates are known to anastomose with difficulty (15), they were paired with several additional primary and secondary AG-2 cultures without succcess. Previously, Parmeter et al (15) found some multinucleate *Rhizoctonia*-like isolates that failed to anastomose. They suggested that some isolates may be unable to anastomose, anastomosis may be extremely rare in some isolates, additional anastomosis groups may exist, or these isolates may be other species with mycelial characteristics similar to *R. solani.* Without formation of the basidial stage, we had insufficient evidence to identify our nonanastomosing multinucleate isolates other than by cultural types.

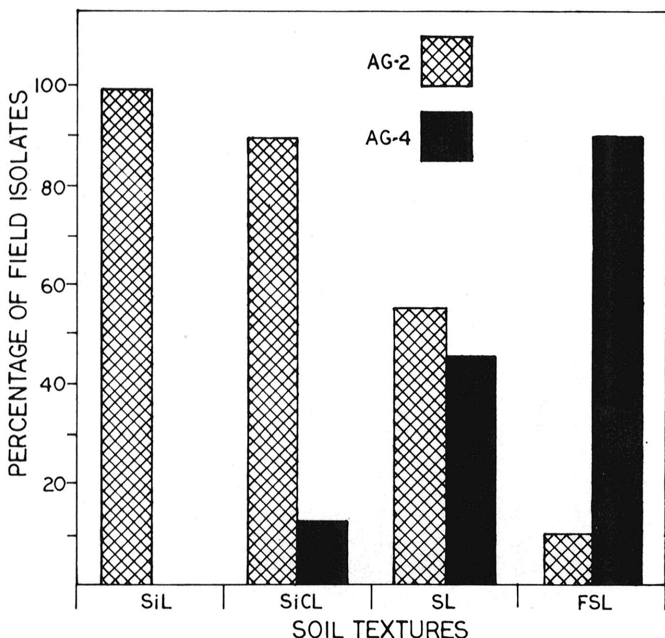

Fig. 1. Percentages of *Rhizoctonia solani* isolates in anastomosis groups in AG-2 and AG-4 obtained from four fields with differing soil textures. (SiL = silt loam, SiCL = silty clay loam, SL = sandy loam, and FSL = fine sandy loam.)

TABLE 1. Relative virulence of *Rhizoctonia* isolates from soil and weeds to sugarbeet seedlings according to specified categories

Soil texture and disease incidence categories	Number of isolates	Percentage of isolates with virulence rating[a] 0	I	II	III	IV
Silty clay loam (SiCL):						
Healthy areas	15	27	0	27	27	20
Diseased areas	19	21	26	32	16	5
Silt loam (SiL):						
Healthy areas	6	33	33	17	17	0
Diseased areas	17	12	53	18	18	0
Fine sandy loam (FSL):						
Healthy areas	29	3	0	3	21	72
Diseased areas	15	7	0	0	20	73
Sandy loam (SL):						
Healthy areas	11	27	18	0	27	27
Diseased areas	14	21	7	29	21	21
AG-2	66	21	26	25	21	8
AG-4	77	7	0	3	25	69
Binucleate isolates	17	93	7	0	0	0
Multinucleate (not grouped)	6	67	17	17	0	0
Weed isolates	40	36	5	5	23	31
Spoil isolates	126	13	13	17	16	40
Total isolates	166	21	12	13	20	34

[a]Virulence ratings of 0, I, II, III, and IV indicate 21–25, 16–20, 11–15, 6–10, and 0–5 surviving seedlings, respectively.

The apparent influence of soil textures on the distribution of anastomosis groups AG-2 and AG-4 (Fig. 1) indicated that there is ecological specialization of these two anastomosis groups. The existence of such ecological specialization of anastomosis groups previously was suggested (12,15,24). The fine textured, poorly drained soils probably are less well aerated and have higher CO_2 concentrations than do the coarser textured, better drained soils. Durbin (4) found that root rotting isolates possessed greater tolerance of high CO_2 concentrations than did foliage and crown isolates. This finding was confirmed by Watanabe and Matsuda (26). According to Ruppel (20), *R. solani* isolates from sugarbeet foliage and crowns all were classified in AG-4, whereas root isolates were AG-2's. Thus, the greater tolerance of high CO_2 concentrations by AG-2 (root isolates) may well explain their predominance in the fine textured soils (Fig. 1). Conversely, the increased percentages of AG-4 (foliage and crown isolates) in the coarser textured soils may be related to a competitive advantage based on their growth rates being higher than those of AG-2 isolates (4,20,24) under nonlimiting CO_2 concentrations.

Naiki and Kanoh (11) reported that *R. solani* was less frequently isolated from diseased spinach plants growing in clay soil than from plants growing in clay loam or loam soil. These results are compatible with the soil population results obtained by Herr (6) in artificially infested silty clay soil (low inoculum densities) compared with the soil populations obtained by Roberts and Herr (19) in naturally infested silt loam and silty clay loam soils (high inoculum densities). Further, Naiki and Kanoh (11) isolated higher percentages of AG-2, type 2 (Ogoshi's [13] designation of *R. solani* strains that cause crown rot of sugarbeet) from spinach plants in clay loam soil than from those in loam soil. Plants from the latter soil yielded nearly 100% AG-4 isolates. Thus, although the methods of isolation differed (plant isolations versus soil isolations), a considerable degree of correspondence in results existed between our results and those of Naiki and Kanoh (11) in distribution of anastomosis groups in soils of different textures.

From an epidemiological standpoint, the pathogenicity of *Rhizoctonia* isolates from soil and weeds to sugarbeets must be ascertained by appropriate assays. We found that soil and weed isolates ranged from avirulent to highly virulent to both sugarbeet seedlings and older plants (Tables 1 and 2). Previously, Papavizas and Davey (14) reported that *R. solani* cultures from several soils ranged from avirulent to highly virulent on a number of hosts, including sugarbeet seedlings, and Daniels (3) found that *R. solani* isolates from weeds ranged from avirulent to highly virulent on seedlings of several crops. Moreover, Daniels (3) postulated that the typical pattern of scattered patches of diseased plants in the field may arise from the presence of highly virulent strains of *R. solani* within the diseased plant areas that are absent in the surrounding areas containing apparently healthy plants. Our seedling assay results indicated that, within fields, isolates from AH and DA generally were similar in relative virulence (Table 1), which does not appear to support Daniels' (3) hypothesis. However, one also should consider the distribution of isolates highly virulent to older plants (ie, those involved in the root and crown rot syndrome) in AH and DA before rejecting her hypothesis. In this general context, Roberts and Herr (19) suggested that factors other than initial *R. solani* inoculum density per se cause plants in some areas within sugarbeet fields to become diseased, while those in other areas remain healthy. This report should give further impetus for a more complete study of Daniels' hypothesis (3), and other possible hypotheses regarding the origin of diseased patches within fields.

Among fields there were marked differences in the relative virulence of isolates to sugarbeet seedlings (Table 1) which were clearly related to the distribution of anastomosis groups (Fig. 1). More AG-4 isolates (Table 1) were highly virulent (69 in category IV) to sugarbeet seedlings than were isolates in AG-2 (eight in category IV). Thus, in agreement with Sherwood (24), the norms for virulence of AG-4 and AG-2 were distinct, even though overlapping occurred with certain isolates. This indicated that a certain minimum number of isolates is required to adequately characterize an anastomosis group. Comparison of seedling pathogenicity results in Table 1 versus Table 2 confirms this contention. In Table 2, with only 32 isolates, the means of all AG-2 and binucleate isolates do not differ, whereas, in Table 1, the differences between AG-2 and binucleate isolates are quite evident from consideration of the percentages of isolates in the five virulence rating categories. The conclusion that the AG-4 isolates were the most virulent to sugarbeet seedlings is evident in either table, however. Others (11,24) have noted the high virulence of AG-4 isolates to seedlings and the frequent association of AG-4 isolates with seedling diseases.

In contrast to the seedling pathogenicity results, AG-2 isolates were significantly more virulent than AG-4 or binucleate isolates in assays on older plants (Table 2). Again, the norms of virulence for AG-2 or AG-4 were distinct, but disease ratings of some isolates of each group overlapped. Occurrence of AG-4 isolates with higher than average virulence on older plants in the well-aerated FSL field may explain the observed occurrence of root and crown disease (similar to that of the other fields), although the percentage of AG-2 isolates was low. The SL field, with nearly equal percentages of AG-2 and AG-4 isolates, represented a similar but less extreme case. Apparently, in well-aerated soils, some AG-4 isolates may cause extensive crown- and root-rot disease losses in sugarbeets.

The binucleate isolates were essentially nonpathogenic on older plants. Since binucleate isolates also were avirulent to weakly

TABLE 2. Comparison of the relative virulence rankings of designated *Rhizoctonia* isolates from soil and weeds to sugarbeet seedlings and to older (6–8 wk) sugarbeet plants and of the collective means of binucleate isolates (BN), anastomosis group 2 (AG-2) and anastomosis group 4 (AG-4) isolates

		Seedling assay[y]			Older plant assay[y]	
Source[x]	Isolate	Group	Surviving seedlings[z]	Isolate	Group	Disease rating[z]
S	1	BN	23.0 a	2	AG-2	4.5 a
S	2	AG-2	23.0 a	4	AG-2	4.4 a
W	3	BN	22.5 a	6	AG-2	4.4 a
S	4	AG-2	20.3 ab	13	AG-2	4.1 ab
S	5	AG-2	20.0 abc	30	AG-4	3.3 bc
S	6	AG-2	17.5 bcd	19	AG-2	2.9 cd
S	7	AG-2	17.3 bcd	14	AG-2	2.7 cde
S	8	AG-2	16.5 bcde	29	AG-4	2.4 cdef
S	9	AG-2	15.3 cde	27	AG-4	2.1 def
S	10	BN	13.3 def	5	AG-2	2.1 def
S	11	AG-2	12.8 def	9	AG-2	1.9 efg
S	12	CT-2	12.8 def	24	AG-4	1.8 efgh
S	13	AG-2	12.0 efg	28	AG-4	1.7 efgh
S	14	AG-2	10.3 fg	17	AG-4	1.6 fghi
S	15	AG-4	10.0 fgh	7	AG-2	1.6 fghi
S	16	AG-2	10.0 fgh	25	AG-4	1.5 fghi
W	17	AG-4	8.8 fghi	26	AG-4	1.5 fghi
S	18	AG-4	8.5 fghi	16	AG-2	1.5 fghi
S	19	AG-2	7.5 ghil	11	AG-2	1.0 ghil
W	20	AG-4	7.0 ghilm	31	AG-4	0.8 hil
S	21	AG-4	5.0 hilmn	23	AG-4	0.8 hil
W	22	AG-4	4.3 ilmn	32	AG-4	0.6 il
S	23	AG-4	3.5 lmn	22	AG-4	0.6 il
W	24	AG-4	2.5 mn	21	AG-4	0.4 l
S	25	AG-4	1.5 n	12	CT-2	0.3 l
S	26	AG-4	1.3 n	20	AG-4	0.3 l
W	27	AG-4	0.5 n	1	BN	0.2 l
S	28	AG-4	0.3 n	3	BN	0.2 l
S	29	AG-4	0 n	8	AG-2	0.2 l
S	30	AG-4	0 n	15	AG-4	0.2 l
S	31	AG-4	0 n	18	AG-4	0.1 l
S	32	AG-4	0 n	10	BN	0 l
		Mean all BN 17.9 a			Mean all AG-2 2.6 a	
		Mean all AG-2 15.2 a			Mean all AG-4 1.2 b	
		Mean all AG-4 3.3 b			Mean all BN 0.2 c	

[x] Source: S = isolated from soil; W = isolated from weed host.

[y] Seedling assay results based on number of surviving seedlings of 25 seed planted, virulence on older plants rated on a 0 to 5 scale (0 = healthy; 5 = dead).

[z] Means followed by the same letter are not significantly different ($P = 0.05$, according to Duncan's multiple range test).

virulent on seedlings, it appears they occurred only incidentally in sugarbeet fields, probably in association with weed hosts.

LITERATURE CITED

1. BOOSALIS, M. G., and A. L. SCHAREN. 1960. The susceptibility of pigweed to *Rhizoctonia solani* in irrigated fields of western Nebraska. Plant Dis. Rep. 44:815-818.
2. BUTLER, E. E., and C. E. BRACKER. 1970. Morphology and cytology of *Rhizoctonia solani*: Pages 32–51 in: J. R. Parmeter, Jr., ed. *Rhizoctonia solani:* Biology and Pathology. University of California Press, Berkeley, Los Angeles, and London. 255 pp.
3. DANIELS, J. 1963. Saprophytic and parasitic activities of some isolates of *Corticium solani.* Trans. Br. Mycol. Soc. 46:485-502.
4. DURBIN, R. D. 1959. Factors affecting the vertical distribution of *Rhizoctonia solani* with special reference to CO_2 concentration. Am. J. Bot. 46:22-25.
5. HERR, L. J. 1970. Resistant sugar beets show promise in Ohio. Ohio Rep. Res. Devel. 55:50-51.
6. HERR, L. J. 1976. In field survival of *Rhizoctonia solani* in soil and in diseased sugarbeets. Can. J. Microbiol. 22:983-988.
7. HERR, L. J. 1979. Practical nuclear staining procedures for Rhizoctonia-like fungi. Phytopathology 69:958-961.
8. KOTILA, J. E. 1947. Rhizoctonia foliage blight of sugar beets. J. Agric. Res. 74:289-314.
9. KUNINAGA, S., R. YOKOSAWA, and A. OGOSHI. 1978. Anastomosis grouping of *Rhizoctonia solani* Kühn isolated from non-cultivated soils. (Japanese, with English abstract.) Ann. Phytopathol. Soc. Jpn. 44:591-598.
10. LeCLERG, E. L. 1939. Studies on dry rot canker of sugar beets. Phytopathology 29:793-800.
11. NAIKI, T., and M. KANOH. 1978. Grouping of *Rhizoctonia solani* Kühn causing root diseases of spinach in plastic house cropping. Ann. Phytopathol. Soc. Jpn. 44:554-560.
12. NAIKI, T., and T. UI. 1978. Ecological and morphological characteristics of the sclerotia of *Rhizoctonia solani* Kühn produced in soil. Soil Biol. Biochem. 10:471-478.
13. OGOSHI, A. 1976. Studies on the grouping *Rhizoctonia solani* Kühn with hyphal anastomosis and on the perfect stage of groups. (Japanese, with English summary.) Bull. Nat. Inst. Agric. Sci., Ser. C., (Plant Pathol. and Entomol.) 30:1-63.
14. PAPAVIZAS, G. C., and C. B. DAVEY. 1962. Isolation and pathogenicity of *Rhizoctonia* saprophytically existing in soil. Phytopathology 52:834-840.
15. PARMETER, J. R., Jr., R. T. SHERWOOD, and W. D. PLATT. 1969. Anastomosis grouping among isolates of *Thanatephorus cucumeris.* Phytopathology 59:1270-1278.
16. PARMETER, J. R., Jr., and H. S. WHITNEY. 1970. Taxonomy and nomenclature of the imperfect state. Pages 7–19 in: J. R. Parmeter, Jr., ed. *Rhizoctonia solani*: Biology and Pathology. University of California Press, Berkeley, Los Angeles, and London. 255 pp.
17. PARMETER, J. R., Jr., H. S. WHITNEY, and W. D. PLATT. 1967. Affinities of some *Rhizoctonia* species that resemble mycelium of *Thanatephorous cucumeris.* Phytopathology 57:218-223.
18. ROBERTS, D. L., and L. J. HERR. 1979. Superiority of a soil debris isolation method over a beet seed colonization method for assay of *Rhizoctonia solani* at high soil inoculum densities. Can. J. Microbiol. 25:110-113.
19. ROBERTS, D. L., and L. J. HERR. 1979. Soil populations of *Rhizoctonia solani* from areas of healthy and diseased beets within four sugarbeet fields differing in soil texture. Can. J. Microbiol. 25:902-910.
20. RUPPEL, E. G. 1972. Correlation of cultural characters and source of isolates with pathogenicity of *Rhizoctonia solani* from sugar beet. Phytopathology 62:202-205.
21. SANDERS, P. L., L. L. BURPEE, and H. COLE, Jr. 1978. Preliminary studies on binucleate turfgrass pathogens that resemble *Rhizoctonia solani.* Phytopathology 68:145-148.
22. SCHMITTHENNER, A. F., and J. W. HILTY. 1962. A method for studying postemergence seedling root rot. Phytopathology 52:177-179.
23. SCHUSTER, M. L., S. G. JENSEN, and R. M. SAYRE. 1958. Toothpick method of inoculating sugar beets for determining pathogenicity of *Rhizoctonia solani.* J. Am. Soc. Sugar Beet Technol. 10:142-149.
24. SHERWOOD, R. T. 1969. Morphology and physiology in four anastomosis groups of *Thanatephorus cucumeris.* Phytopathology 59:1924-1929.
25. TU, C. C., and J. W. KIMBROUGH. 1973. A rapid staining technique for *Rhizoctonia solani* and related fungi. Mycologia 65:941-944.
26. WATANABE, B., and A. MATSUDA. 1966. Studies on the grouping of *Rhizoctonia solani* Kühn pathogenic to upland crops. (Japanese, with English summary.) Ibaraki Agric. Exp. Stn. (Japan) Bull. 7. 131 pp.