# Etiology

# Characteristics of Rhizoctonia Isolates Associated with Bottom Rot of Lettuce in Organic Soils in Ohio

#### Leonard J. Herr

Professor, Department of Plant Pathology, The Ohio State University, Ohio Agricultural Research and Development Center (OARDC), Wooster 44691.

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## ABSTRACT

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Rhizoctonia isolates obtained from lettuce with bottom rot symptoms—collected at two major Ohio lettuce production areas, Celeryville in 1989 and Hartville in 1990—were characterized according to anastomosis groups (AGs) and, when feasible, by intraspecific groups (ISGs). Isolates belonging in AG-2-1 were further subdivided into three new provisional ISGs (designated types a-c) on the basis of their cultural characteristics. Thirty-three of 51 Celeryville isolates and 28 of 59 Hartville isolates were characterized as AG-1-1B, the web blight ISG of AG-1. Additionally, AG-1-1C isolates were obtained from diseased lettuce at Celeryville (10 isolates) and Hartville (four isolates). Isolation of these ISGs of AG-1 represents a first report of the occurrence of any of the ISGs of AG-1 in Ohio. AG-2-1 (20 isolates) and AG-5 were obtained only from Hartville,

whereas AG-2-2 was isolated only from Celeryville. Other isolates of AG-4, binucleate *Rhizoctonia* spp., and *Laetisaria arvalis* occurred infrequently at both locations. Of the major AGs collected and evaluated in greenhouse tests, AG-1-1B isolates were collectively the most virulent, isolates of AG-1-1C were intermediate, and isolates of AG-2-1 were least virulent. Of three lettuce types used in greenhouse tests of the Celeryville isolate population, romaine was susceptible to more isolates than Boston, and red leaf was susceptible to the lowest number of isolates. Hartville isolates were tested for virulence only on romaine and were found to differ significantly in virulence capabilities among isolates of the population of *R. solani* collected.

Additional keywords: Lactuca sativa, Thanatephorus cucumeris.

A report of a leaf-rotting disease of greenhouse lettuce crops in Massachusetts in 1900 by Stone and Smith (31) appears to be the first account of *Rhizoctonia* causing disease on older lettuce (*Lactuca sativa* L.) plants. Dye (4) coined the name "bottom rot" for the destructive *Rhizoctonia* disease of lettuce occurring in New York organic field soils. Symptoms were described as "rusty lesions on midribs and a rotting away of leaf blades of lower leaves." In Florida, bottom rot occurred on escarole and endive (*Cichorium endivia* L.) as well as on lettuce, and the cause of the disease was attributed to *Corticium vagum* sensu Burt, pro parte = *Thanatephorus cucumeris* (A. B. Frank) Donk, anamorph *Rhizoctonia solani* Kühn (34). During the early 1930s, bottom rot disease losses of 30% annually were reported in lettuce crops in New York organic soils (32). Little evidence of plant resistance to *R. solani* was observed, but upright lettuce types, such as

romaine, had a tendency to escape disease because of greater air circulation and drying at the soil surface-lettuce interface compared with that of the spreading leaf lettuce types (32). However, susceptibility differences between two spreading lettuce types, Butterhead and White Boston, could not be explained on this basis. Furthermore, Townsend (32) noted the occurrence of different strains (races) among the R. solani cultures isolated from lettuce as exemplified by their virulence on lettuce, growth at different temperatures, and cultural characteristics. Except for the infrequent observation of the perfect state of R. microsclerotia J. Matz = T. cucumeris, anamorph R. solani AG-1-1B, these strains cannot be related to present day Rhizoctonia anastomosis groups (AGs) (19). The roles of soft rot bacteria and R. solani in lettuce bottom rot were investigated, and only R. solani could initiate the disease in unwounded plants. In advanced stages of bottom rot, soft rot bacteria occurred commonly and exacerbated rotting (23). In a study in the Netherlands (13) of glasshouse lettuce crops, 24 R. solani isolates obtained from diseased lettuce

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and 11 from glasshouse soils were classified into the four AGs of Parmeter et al (22). At most of the 17 glasshouses sampled, AG-1 was the most prevalent AG isolated from lettuce, whereas AG-2 and AG-4 isolates were found only occasionally. Soil isolates were mainly AG-4 and infrequently AG-3. The relative virulence of these *R. solani* isolates from different AGs was not ascertained (13).

The principal objectives of this study were 1) to classify *R. solani* isolates, obtained from diseased lettuce grown at the two locations of Ohio lettuce production on organic soils, by AG and, when feasible, by intraspecific groups (ISGs); 2) to evaluate the relative virulence of the isolates within these AGs to lettuce in greenhouse tests; and 3) to ascertain the relative susceptibility of three different lettuce types to *R. solani* isolates from different AGs. A preliminary report on bottom rot of lettuce and *R. solani* isolates from the Celeryville area was published previously (9).

## MATERIALS AND METHODS

Field collections of lettuce. Samples were collected from the major areas of lettuce production in Ohio, which are near Celeryville (Huron County) in north central Ohio and Hartville (Stark County) in northeastern Ohio. Although separated by approximately 120 km, both areas have the same soil type (Rifle peat, an euic Typic Borohemists [1]). Romaine, Boston, red leaf, green leaf, and bibb lettuce, escarole, and (Hartville area only) endive with bottom rot symptoms were collected from growers' fields on 19 July and 14 August 1989 at Celeryville and on 21 June, 18 July, 26 July, and 16 August 1990 at Hartville. Disease incidence in 1990, a cooler and wetter season than normal, was lower than in 1989.

Isolation procedures. Diseased plants were stored at 4 C until processed, which in most cases was within 24 h of collection. Plants were washed to remove the soil, separated into leaves, stems, and roots, and cut into large (approximately 2 cm) pieces exhibiting symptoms. These pieces were placed in a stainless steel kitchen sieve inserted into a beaker and washed under cold running tap water for at least 10 min. A wetting agent, Tween 20 (Nutritional Biochemical Co., Cleveland, OH), was added initially to aid washing. After washing, plant material was blotted on paper towels, cut into smaller pieces, and plated (five pieces per plate) on 2% distilled water agar (WA) in petri plates. WA plates were enclosed in paper bags and incubated for 24-48 h at 24 C. Fungi that grew from plated pieces were examined at 100-200× magnification through the bottom of closed plates. Colonies typical of Rhizoctonia spp. were marked, and hyphal tips were transferred to WA plates and freed of contaminating microorganisms by a modification of Schmitthenner and Hilty's technique (27). The WA layer in plates was inverted with a spatula and cut (approximately 1 cm from edge) around the periphery of the plate to seal the layer to the plate bottom. When fungal growth was evident on the upper surface of the agar layer, a piece of mycelium was carefully removed without completely penetrating the agar layer and was transferred to potato-dextrose agar (PDA). After subsequent growth, to ensure freedom from contaminating bacteria, the isolates were transferred to PDA slants in test tubes and were periodically transferred (at 3-mo intervals) to new slants kept at room temperature.

Characterization of isolates. The number of nuclei in hyphal vegetative cells and the nature of the septal pore apparatus of isolates were ascertained by use of a rapid aniline blue staining technique or, if necessary, by a HCl-Giemsa method (7). Isolates that conformed to the criteria for *Rhizoctonia* spp. (18) were retained in culture and were further subdivided into *R. solani* isolates according to the criteria characteristic for *R. solani* (21). The clamp connection-bearing basidiomycete, *Laetisaria arvalis* Burdsall, also was occasionally isolated and retained in culture.

Anastomosis pairings for AG assignment were made in duplicate (with unresolved pairings repeated as necessary) on either WA-coated slides (10) or the WA plate method (5 or 7 ml of 1.7% WA in 90-mm-diameter plastic petri plates, modified from Ogoshi [17]) according to Ogoshi's criteria for anastomosis (19).

Anastomosis tester isolates included AG-1-1C, AG-2-2, AG-3, and AG-4 testers received from N. Anderson, Department of Plant Pathology, University of Minnesota, St. Paul; several AG-2-2 IV and AG-4 testers from our collection; AG-2-1, AG-2-2 IIIB, AG-6, AG-7, AG-8, and AG-BI from D. K. Bell and D. R. Sumner, Department of Plant Pathology, University of Georgia, Coastal Plain Agricultural Experiment Station, Tifton; AG-5 from G. S. Abawi, Cornell University, New York State Agricultural Experiment Station, Geneva; AG-2-1 and AG-9 from B. Nelson, Department of Plant Pathology, North Dakota State University, Fargo; AG-8 from L. L. Burpee, Department of Plant Pathology, University of Georgia, Georgia Agricultural Experiment Station, Griffin; and AG-2-1, AG-1-1A, AG-1-1A, AG-1-1B, AG-1-1B, and AG-2-2 IIIB (ATCC 62805, 66157, 66158, 66150, 66151, and 66153, respectively) from the American Type Culture Collection, Rockville, MD.

These AG tester isolates were organized into "pathogenic" AGs, i.e., typically pathogenic on one or more green plant hosts, although individual isolates within an AG may be avirulent, and "saprophytic" AGs, i.e., AG-6, AG-7, and AG-BI, reported only as soil saprophytes (19). The pathogenic AGs include AG-1-1A, AG-1-1B, AG-1-1C, AG-2-1, AG-2-2 (including AG-2-2 IIIB and AG-2-2 IV), AG-3, AG-4 (sometimes separated into AG-4 HG-I and AG-4 HG-II), AG-5, AG-8, and AG-9. Of these 12 pathogenic AG, AG-8, found on wheat roots in Australia (16,25) and the Pacific Northwest (20), and AG-9, pathogenic on potato in Alaska (3), have not yet been reported in the midwestern United States. Therefore, the principal AG tester isolates used in this study were in the pathogenic AG-1 through AG-5.

A systematic approach was used to assign the relatively large numbers of collected lettuce isolates into AGs and, when feasible, to ISGs, as delineated by Ogoshi (19). First, all lettuce isolates and the pathogenic testers AG-1 through AG-5 were transferred to PDA and to PDA + 1.0 g of yeast extract per liter (26) media in petri plates. Over several weeks of incubation at 24 C (enclosed in paper bags), they were tentatively assigned into culturally similar groups on the basis of colony characteristics. The isolates from lettuce were then paired with culturally similar AG tester isolates and related ISGs. The ISGs assigned were distinguished on the basis of cultural characteristics (5,12,19,28,35) of the three ISGs included in AG-1 (which all readily anastomose with one another) after incubation for approximately 14 days at 24 C on PDA and WA plates and on the basis of frequency of anastomosis for ISGs within AG-2, i.e., AG-2-1 and AG-2-2 (18). No attempt was made to further separate AG-2-2 into AG-2-2 IIIB and AG-2-2 IV (19), nor was separation of AG-4 into AG-4 HG-I and AG-4 HG-II (14) attempted.

Greenhouse virulence tests. Black polystyrene plug flats (28 cm in width × 55 cm in outside diameter length; 288 plug holes squared and tapered in shape, 2.0 cm width × 3.2 cm depth; TLC Polyform, Inc., Plymouth, MN) were filled with Metro Mix 350 plug mix (W. R. Grace & Co., Cambridge, MA) and seeded with pelleted lettuce seed. In the 1989 Celeryville isolate tests, three lettuce types were used: romaine cv. Tall Guzmaine MF, Boston cv. Oresto, and red leaf cv. Sierra. In the 1990 Hartville isolate test, only romaine cv. Tall Guzmaine MF was used. The seeded plug flats were placed in a growth chamber with 14 h of light (150  $\mu$ mol·m<sup>-2</sup>·s<sup>-1</sup>) alternated with 10 h of dark (21  $\pm$ 1 C) for germination and early growth. After approximately 2 wk, the flats were transferred to a greenhouse bench, and seedlings were transplanted for testing approximately 7 days later. Styrofoam cups (16-oz [473 ml], squat cups, model 16MJ20, Dart Container Corp., Mason, MI) with four holes (5 mm in diameter) punched into each bottom for drainage were filled with a greenhouse potting mix consisting of Rifle peat, Wooster silty loam (fine-loamy, mixed, mesic, Typic Fragiudalfs), and sphagnum peat moss (v/v) in a 5:15:1 ratio, with addition of 142 g of 9-45-15 fertilizer and 341 g of lime per 0.21 m3 of potting soil mix. One lettuce plug plant was transplanted into the center of the potting mix in each styrofoam cup, placed on greenhouse benches, and maintained at 26  $\pm$  3 C (day) and 20  $\pm$  2 C (night). Supplemental fluorescent lighting (16 \(\mu\text{mol·m}^{-2}\cdot s^{-1}\)\) was provided daily for 14 h. All lettuce plants, including plug flat seedlings in the greenhouse, were fertilized weekly with a liquid fertilizer solution (2.15 g of 20-20-20 per liter of water).

One to 5 days after seedlings were transplanted, the soil in the styrofoam containers was infested with appropriate isolates (four replicate cups [plants] per isolate). For soil infestation, two 9-mm-diameter disks cut with a cork borer from 7-day-old isolate cultures on PDA were buried in slits, made in the potting soil with a spatula on opposite sides and immediately adjacent to the lettuce plant plug, to a depth of approximately 2 cm. The lettuce plants were rated for disease 7-10 days after infestation of the soil on a scale where 1 = healthy, 2 = diseased, and 3 = dead. Tests for each set of isolates, 1989 Celeryville and 1990 Hartville, were conducted twice.

Disease rating (DR) results were analyzed by analysis of variance as a complete randomization design, and confidence intervals were determined by Fisher's least significance test at the 5% level.

### RESULTS

Isolate pairings based on similar cultural characteristics yielded mostly positive anastomoses, providing evidence that many isolates were correctly classified by cultural characteristics. However, some isolates were incorrectly assigned or unassignable to specific AGs on the basis of cultural characteristics; thus, they were classified correctly only after anastomosis with known tester isolates. A group of 20 isolates from the 1990 Hartville set anastomosed with one another but failed to do so with our standard AG-1 through AG-5 tester isolates or our less-used AG-6, AG-7, AG-BI, AG-8, and AG-9 anastomosis tester isolates. Microscopic reexamination of the anastomosis tester isolates revealed that the single AG-2-1 tester then available deviated from R. solani descriptions, primarily in that young hyphae at colony margins branched excessively and noncharacteristically and were much smaller in diameter than the runner hyphae. Two additional AG-2-1 tester isolates were obtained that anastomosed with the 20 Hartville lettuce isolates and with one another but did not anastomose with the original deviate AG-2-1 tester. These 20 previously unclassified isolates were placed in AG-2-1, although two distinct cultural types, designated a and b, occurred within this group. Type a colonies had distinctive, angular, monilioid cells and were reddish brown in color; colonies usually exhibited concentric crusty rings of dense monilioid cells (sclerotia) interspersed with reddish appressed hyphae. Type b colonies were brown, with a darker brown circular central area (approximately 2-3 cm in diameter) of sclerotia. Some larger (2-10 mm) light brown sclerotia, often with droplets of liquid exudate, usually occurred on the outer periphery of the central sclerotial area. Both types differed culturally from either of the two newly acquired AG-2-1 tester isolates. The two tester isolates differed from one another in minor ways: one (ATCC 62805) had few to no sclerotia and light to medium brown appressed mycelium, the other tester had light brown mycelium (very similar in color to ATCC 62805) with limited amounts of aerial hyphae. Some areas of the colony were darker brown, and scattered small (approximately 1.0 mm in diameter) sclerotia were present in limited areas of the plate. These two testers were designated as type c and generally conformed to the cultural descriptions of AG-2-1 (6,24,25). Despite the markedly different cultural characteristics of the Hartville type a and type b isolates, they definitely anastomosed, although sparingly with one another in all pairings. Type a anastomosed readily with the two AG-2-1 type c testers, whereas type b anastomosed sparingly with them. All of these cultural types were therefore tentatively classified as provisional ISGs within AG-2-1 pending further study.

Thirty-three of 51 Celeryville isolates and 28 of 59 Hartville isolates were AG-1-1B (Figs. 1 and 2). Aside from these predominating AG-1-1B isolates, the isolate collections from the two areas differed in either numbers or types of given AG. Specifically, AG-2-1 (20 isolates) and AG-5 (one isolate) were found at Hartville but not at Celeryville, whereas AG-2-2 (two isolates) were found

only at Celeryville. Isolates of AG-1-1C and AG-4 were recovered more frequently from Celeryville than from Hartville (Figs. 1 and 2). Binucleate *Rhizoctonia* spp. and *L. arvalis* isolates were isolated infrequently from both locations.

Results of the two greenhouse tests of virulence with isolates collected at Celeryville on red leaf, Boston, and romaine lettuce were combined for analysis (homogeneous variances) and presented as ranked arrays of constituent AGs of isolate DRs on cultivar in Figure 1. The interaction of isolates X lettuce cultivars was significant (P < 0.0001, LSD<sub>0.05</sub> = 0.63). To distinguish levels of virulence among isolates, those isolates with DR values of 2.0 or greater were arbitrarily considered highly virulent. Accordingly, red leaf cv. Sierra (Fig. 1A) was highly susceptible to 22 isolates (the fewest isolates), Boston cv. Oresto (Fig. 1B) was highly susceptible to 35 isolates (an intermediate number of isolates), and romaine cv. Tall Guzmaine MF (Fig. 1C) was highly susceptible to 42 isolates (the greatest number of highly virulent isolates within the Celeryville population of R. solani isolates). Thus, the relative susceptibilities of the three lettuce types to the population of R. solani isolates differed, but the "absolute" susceptibility to certain isolates did not, i.e., some (variable number) isolates were maximumly virulent (DR = 3.0,

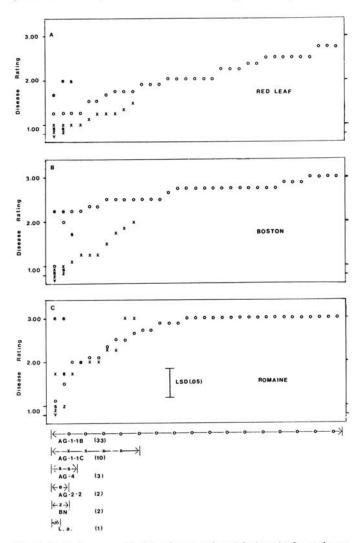


Fig. 1. Ranked arrays of isolate disease ratings (virulence) of constituent anastomosis groups on cultivars of a population of *Rhizoctonia solani* isolates obtained from lettuce plants exhibiting bottom rot disease symptoms in organic soil fields at Celeryville, OH, in 1989. A, Red leaf cv. Sierra; B, Boston cv. Oresto; C, romaine cv. Tall Guzmaine MF. Disease was rated on a scale where 1 = healthy, 2 = diseased, and 3 = dead plants. Anastomosis groups (AG) and intraspecific groups are identified below graphs. BN = binucleate *Rhizoctonia* spp., L. a. = *Laetisaria arvalis*. Graph scale (disease rating = 1.0) expanded to accommodate isolate symbols.

dead plant) on all three lettuce types. Of the AGs, AG-1-1B was collectively the most virulent and most frequently isolated of the isolates. With the exception of two of the isolates on romaine lettuce (Fig. 1C), all AG-1-1C isolates ranked below the most virulent isolates of AG-1-1B in virulence (Fig. 1). Of the other AGs isolated infrequently, AG-2-2 exhibited high virulence and AG-4 variable virulence on romaine. The virulence of both AGs was modified by the relative susceptibility of the lettuce types to them (Fig. 1). Binucleate Rhizoctonia spp. and L. arvalis isolates were nonpathogenic on all three lettuce types. A range of virulence, ranging on romaine (Fig. 1C) from low or avirulent to maximum (DR = 3.0), was evident among isolates within those AGs (AG-1-1B and AG-1-1C) that had sufficient numbers of isolates for such evaluations to be made.

The Hartville 1990 collection of isolates was tested for virulence only on romaine cv. Tall Guzmaine MF type lettuce. The results of two tests (homogeneous variances) were combined for analysis. Isolates differed from one another in virulence (P = 0.0001, $LSD_{0.05} = 1.03$ ) (Fig. 2). Again, isolates within AG-1-1B were the most numerous of the groups and included the most virulent isolates. The AG-1-1B isolates (Fig. 2) ranged from a low order of virulence (DR = 1.50) to maximum virulence (DR = 3.0, i.e., plants dead). A major difference between the Celeryville and Hartville collections of isolates was the occurrence of AG-2-1 (20 isolates) in the Hartville collection versus absence of this ISG in the Celeryville collection. Most AG-2-1 isolates were weakly virulent, and half were avirulent. The four AG-1-1C isolates were moderately virulent, whereas the infrequently isolated AG-4, AG-5, and L. arvalis (one isolate each) and the four binucleate Rhizoctonia spp. of the Hartville isolates were avirulent (Fig. 2).

## DISCUSSION

The predominance of R. solani isolates of AG-1-1B, web blight type of AG-1 (19,28), in the isolate collections from diseased lettuce both at Celeryville and Hartville was not anticipated because none of the three ISGs of AG-1 had been reported in previous studies of crops grown in the field on mineral soils of Ohio (8,10,15) or from greenhouse bedding plants or soils (30). Additionally, the AG-1-1B isolates usually have been reported from southern

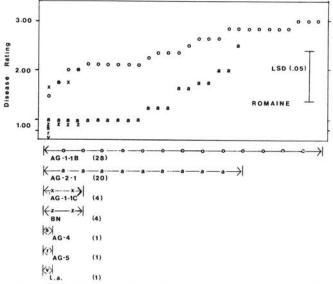


Fig. 2. Ranked array of isolate disease ratings (virulence) of constitutent anastomosis groups on romaine lettuce of a population of *Rhizoctonia solani* isolates obtained from lettuce plants exhibiting bottom rot disease symptoms in organic soil fields at Hartville, OH, in 1990. Disease was rated on a scale where 1 = healthy, 2 = diseased, and 3 = dead plants. Anastomosis groups (AGs) and intraspecific groups are identified below graph. BN = binucleate *Rhizoctonia* spp., L. a. = *Laetisaria arvalis*. Graph scale (disease rating = 1.0) expanded to accommodate isolate symbols.

states of the United States rather than in the north, although AG-1-1B isolates occurred on turf in Pennsylvania (2,33). Furthermore, AG-1-1B isolates occur in all parts of Japan, including Hokkaido, the northernmost Japanese island (19). In New York R. microsclerotia (=R. solani AG-1-1B by current classification) occurred in hymenial form on lettuce on three occasions over a period of several years (32). In the Netherlands, Kooistra (13) reported AG-1 to be the most commonly isolated AG of the four recognized by Parmeter et al (22). Unfortunately, his AG-1 isolates were not further subdivided into ISGs (19), so they could have been any one or more of the ISGs of AG-1. AG-1-1B may produce microsclerotia on plant hosts but forms large sclerotia on nutrient media (5,12,28,35). Use of 2% WA for microsclerotial production in culture (35) proved most useful for separating isolates of AG-1-1B from AG-1-1A isolates, along with cultural comparisons of tester AG-1-1A and AG-1-1B isolates with lettuce isolates on PDA. In contrast, the AG-1-1C isolates, conforming to Sherwood's (28) type 3 ("salt and pepper" colony type) of AG-1, can be readily distinguished from AG-1-1A (sasakii type) and AG-1-1B (web blight type) isolates. Neither AG-1-1B nor AG-1-1C types of AG-1, isolated at both locations, or AG-5 from Hartville had previously been reported as occurring in Ohio.

A unique finding was the occurrence of two cultural types within the AG-2-1 isolates obtained from diseased lettuce at Hartville. Neither of these cultural types, which were designated a and b, was similar culturally to two newly acquired AG-2-1 tester isolates (subsequently designated as a third type, type c). These type c isolates corresponded to generally accepted descriptions of AG-2-1 (6,24,25). However, Sneh et al (29) characterized AG-2-1 as "comprised of slow growing isolates . . . forming reddish sclerotia in rings . . . " Type a of AG-2-1, isolated from Hartville, corresponded in most respects to the limited (29) description of Sneh et al. If type a is indeed similar to the AG-2-1 described by Sneh et al (29), then types a and c both have been described previously but not recognized as distinct cultural types of AG-2-1. The Hartville AG-2-1 isolates designated as type b have no described equivalent in the literature examined. These isolates were somewhat similar culturally to AG-1-1A and AG-1-1B but did not anastomose with ISGs of AG-1 in any combinations. They anastomosed sparingly in every pairing with the AG-2-1 type c (tester isolates) and type a isolates from Hartville, as well as with an AG-2-1 type a isolate obtained from canola in Indiana (11). Further, these type b isolates anastomosed readily with one another. None of these AG-2-1 types a-c was observed to anastomose when paired with tester isolates of AG-2-2 IIIB or AG-2-2 IV. Lastly, both AG-BI and AG-8 are bridging-type AGs (19,25) capable of anastomosing readily with one another and rarely with other isolates, including AG-2-1 and AG-2-2 IIIB. Neither of these tester isolates anastomosed with isolates of AG-2-1 type b from lettuce. Tentatively, these AG-2-1 cultural types a-c are designated as provisional ISGs pending further study. If these provisional ISGs can be demonstrated to differ pathogenically, ecologically, and/or geographically, that is, in characters defining recognized ISGs, the "provisional" term would be dropped in favor of simply ISG designations.

The populations of *R. solani* isolated from diseased lettuces from each of the two lettuce production sites consisted of a diversity of different AGs, differing in frequencies of isolation (numbers of isolates) within AGs and possessing variable virulence capabilities among and within isolates of the AGs. Also, differences were evident in the Celeryville population in the susceptibility of lettuce cultivars to the isolates within this population of *R. solani* as measured by the number of isolates rated highly virulent on each of the three cultivars tested in greenhouse studies.

Virulence capabilities of *R. solani* isolates within the Celeryville and Hartville populations (on romaine lettuce) ranged from avirulent to maximumly virulent. Other soil populations of *R. solani* (consisting of AG-2-2 and AG-4 isolates) exhibited a range of virulence of isolates on sugar beet (10), which may indicate such variations in virulence commonly occur in field populations of *R. solani*. Hypotheses and speculations that may account for the obvious differences in the relative abundance of different AGs

within the R. solani populations will be discussed in a different context elsewhere (L. J. Herr, unpublished).

The differences in relative susceptibility of the three cultivars were not due to the complete absence of any isolates within the R. solani population (Celeryville) that were capable of killing plants of any of the cultivars (a variable number of isolates did so). Instead they were due to differences in the numbers of isolates within the population tested that were capable of being highly virulent on individual cultivars. Conceivably the differences in the susceptibility of cultivars could be due to quantitative resistance factors (such as inhibitory substance content) among cultivars and to differences in tolerance to such postulated inhibitors among the component isolates within populations of R. solani. The implications of such a theory with respect to possible differences in disease losses of cultivars and possible selective effects on isolates within populations of R. solani in the field are worthy of consideration.

## LITERATURE CITED

- Anonymous. 1975. Soil taxonomy: A basic system of soil classification for making and interpreting soil surveys. USDA Soil Conserv. Serv. Agric. Handbook 436. U.S. Government Printing Office, Washington, DC.
- Anonymous. 1990. Rhizoctonia strains available from ATCC. American Type Culture Collection, Rockville, MD.
- Carling, D. E., Leiner, R. H., and Kebler, K. M. 1987. Characterization of a new anastomosis group (AG-9) of *Rhizoctonia solani*. Phytopathology 77:1609-1612.
- Dye, H. W. 1922. The bottom-rot disease of western New York lettuce. (Abstr.) Phytopathology 12:48.
- Exner, B. 1953. Comparative studies of four *Rhizoctonias* occurring in Louisiana. Mycologia 45:698-719.
- Grisham, M. P., and Anderson, N. A. 1983. Pathogenicity and host specificity of *Rhizoctonia solani* isolated from carrots. Phytopathology 73:1564-1569.
- Herr, L. J. 1979. Practical nuclear staining procedures for Rhizoctonia-like fungi. Phytopathology 69:958-961.
- 8. Herr, L. J. 1982. Characteristics of hymenial isolates of *Thanatephorus* cucumeris on sugar beets in Ohio. Plant Dis. 66:246-249.
- Herr, L. J. 1991. Anastomosis groupings and virulence of Rhizoctonia solani isolates from muck-grown lettuce with bottom rot. (Abstr.) Ohio. J. Sci. 91:27.
- Herr, L. J., and Roberts, D. L. 1980. Characterization of *Rhizoctonia* populations obtained from sugarbeet fields with differing soil textures. Phytopathology 70:476-480.
- Huber, D. M., Herr, L. J., Christmas, E. P., and Roseman, T. S. 1991. Crown rot, a serious disease of canola in the midwest. (Abstr.) Phytopathology 81:1186.
- Jones, R. K., and Belmar, S. B. 1989. Characterization and pathogenicity of *Rhizoctonia* spp. isolated from rice, soybean, and other crops grown in rotation with rice in Texas. Plant Dis. 73:1004-1010.
- Kooistra, T. 1983. Rhizoctonia solani as a component in the bottom rot complex of glasshouse lettuce. Mededel. Plantenziektenkundige Dienst. 160.
- Kuninaga, S., and Yokosawa, R. 1984. DNA base sequence homology in *Rhizoctonia solani* Kühn. III. Genetic relatedness within AG-4. Ann. Phytopathol. Soc. Jpn. 50:322-330.

- Muyolo, G., Lipps, P. E., and Herr, L. J. 1991. Variability among isolates of *Rhizoctonia solani* associated with dry bean and soybean in Ohio, USA and Zaire, Africa. (Abstr.) Phytopathology 81:1180.
- Neate, S. M., and Warcup, J. H. 1985. Anastomosis grouping of some isolates of *Thanatephorus cucumeris* from agricultural soils in South Australia. Trans. Br. Mycol. Soc. 85:615-620.
- Ogoshi, A. 1972. Grouping of *Rhizoctonia solani* Kühn with hyphal anastomosis. Ann. Phytopathol. Soc. Jpn. 38:117-122.
- Ogoshi, A. 1975. Grouping of Rhizoctonia solani Kühn and their perfect stages. Rev. Plant Prot. Res. 8:93-103.
- Ogoshi, A. 1985. Anastomosis and intraspecific groups of Rhizoctonia solani and binucleate Rhizoctonia. Fitopatol. Bras. 10:371-390.
- Ogoshi, A., Cook, R. J., and Bassett, E. N. 1990. Rhizoctonia species and anastomosis groups causing root rot of wheat and barley in the Pacific Northwest. Phytopathology 80:784-788.
- Parmeter, J. R., Jr. 1970. Taxonomy and nomenclature of the imperfect state. Pages 7-19 in: *Rhizoctonia solani*: Biology and Pathology. J. R. Parmeter, Jr., ed. University of California Press, Berkeley.
- Parmeter, J. R., Jr., Sherwood, R. T., and Platt, W. D. 1969.
   Anastomosis grouping among isolates of *Thanatephorus cucumeris*. Phytopathology 59:1270-1278.
- Pieczarka, D. J., and Lorbeer, J. W. 1975. Microorganisms associated with bottom rot of lettuce grown on organic soil in New York State. Phytopathology 65:16-21.
- Roberts, F. A., and Sivasithamparam, K. 1986. Identity and pathogenicity of *Rhizoctonia* spp. associated with bare patch disease of cereals at a field site in Western Australia. Neth J. Plant Pathol. 92:185-195.
- Rovira, A. D., Ogoshi, A., and McDonald, H. J. 1986. Characterization of isolates of *Rhizoctonia solani* from cereal roots in South Australia and New South Wales. Phytopathology 76:1245-1248.
- Ruppel, E. G. 1972. Correlation of cultural characters and source of isolates with pathogenicity of *Rhizoctonia solani* from sugar beet. Phytopathology 62:202-205.
- Schmitthenner, A. F., and Hilty, J. W. 1962. A modified dilution technique for obtaining single-spore isolates of fungi from contaminated material. Phytopathology 52:582-583.
- Sherwood, R. T. 1969. Morphology and physiology in four anastomosis groups of *Thanatephorus cucumeris*. Phytopathology 59:1924-1929.
- Sneh, B., Burpee, L., and Ogoshi, A. 1991. Identification of Rhizoctonia species. American Phytopathological Society, St. Paul, MN.
- Stephens, C. T., Herr, L. J., Schmitthenner, A. F., and Powell, C. C. 1982. Characterization of *Rhizoctonia* isolates associated with damping-off of bedding plants. Plant Dis. 66:246-249.
- Stone, G. E., and Smith, R. E. 1900. The rotting of greenhouse lettuce. Mass. Agric. Exp. Stn. Bull. 69.
- Townsend, G. R. 1934. Bottom rot of lettuce. N.Y. (Cornell) Agric. Exp. Stn. Mem. 158.
- Vilgalys, R. 1988. Genetic relatedness among anastomosis groups in Rhizoctonia as measured by DNA/DNA hybridization. Phytopathology 78:698-702.
- Weber, G. F., and Foster, A. C. 1928. Diseases of lettuce, romaine, escarole, and endive. Fla. Agric. Exp. Stn. Bull. 195:299-333.
- Yang, X. B., Snow, J. P., and Berggren, G. T. 1989. Morphogenesis
  of microsclerotia and sasakii-type sclerotia in *Rhizoctonia solani*,
  anastomosis group I, intraspecific groups IA and IB. Mycol. Res.
  93:429-434.