

Zonate Eyespot on Wild Rice Caused by *Drechslera gigantea*

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ABSTRACT

Kardin, M. K., Bowden, R. L., Percich, J. A., and Nickelson, L. J. 1982. Zonate eyespot on wild rice caused by *Drechslera gigantea*. Plant Disease 66:737-739.

Drechslera gigantea was identified as the causal agent of zonate eyespot of wild rice (*Zizania aquatica*) in Minnesota. The disease was observed in four growers' fields, two University of Minnesota Experiment Station fields, and two natural stands. In greenhouse tests, wild rice cultivars Netum, K-2, and Johnson were susceptible to *D. gigantea*. Host range studies indicated that smooth brome (*Bromus inermis*), quackgrass (*Agropyron repens*), and reed canarygrass (*Phalaris arundinacea*) can serve as alternate hosts of *D. gigantea*.

In 1979, an undescribed leaf spot disease of wild rice (*Zizania aquatica* L.) was observed in growers' fields in Aitkin and Clearbrook counties, MN, and at the University of Minnesota Agricultural Experiment Station at Rosemount. The disease resulted in only slight leaf damage on wild rice plants at the experiment station farm and growers' fields in Clearbrook County. However, the disease caused considerable leaf damage in one grower's fields in Aitkin County, even though it only occurred sporadically. The objectives of this study were to ascertain the pathogen, determine its host range, identify resistant cultivars, and determine its geographic distribution.

MATERIALS AND METHODS

Isolation, identification, and distribution of the pathogen. Conidia, when present on the lesions, were touched with a fine needle, spread onto thin, 2% water agar or potato-dextrose agar (Difco), and monoconidial isolates were obtained using a micromanipulator. If conidia were not available on the lesions, naturally infected leaves were cut into small pieces (0.25 cm²), surface sterilized with 70% ethanol for 10 seconds, drained, and placed on potato-dextrose or V-8 agar medium (200 ml of V-8 juice, 3 g of calcium carbonate, 20 g of Bacto agar, and 800 ml of distilled water). Single

conidia were obtained from the resulting colonies. Cultural characters and conidial measurement were determined using 15-day-old cultures grown on V-8 agar at 24 C under continuous darkness.

Fields were surveyed in August 1980 to study the geographic distribution of zonate eyespot and other diseases of wild rice in 13 commercial farms and six natural stands in Minnesota. Diseased leaves with zonate eyespot symptoms were collected and the pathogen isolated as described above.

Pathogenicity tests. Three isolates of the fungus *Drechslera gigantea* (Heald and Wolf) Ito (Dg 7903, Dg 7904, and Dg 7906) were selected for pathogenicity tests. A conidial suspension was prepared from 2-wk-old cultures grown at 24 C under continuous darkness. Two wild rice plants per pot of cultivar K-2 were grown in 16.5-cm-diameter pots and fertilized with 1.5 g of fertilizer (N-P-K, 16-16-16). The pots were placed in wooden boxes lined with plastic sheets and flooded. The water depth was maintained at 0.5 cm above the soil surface to facilitate rapid aerial leaf development and to prevent a floating

leaf stage.

Plants were inoculated with a conidial suspension (20 ml/pot) at the tillering stage using a DeVilbiss atomizer. The conidial concentration of isolates Dg 7903 and Dg 7906 were 4,000 and 1,200 conidia per milliliter, respectively. The inoculated plants were placed in a moist chamber at 20–35 C, 90–100% relative humidity, and a 12-hr photoperiod in the greenhouse for 10 days. Lesions were present by day 5, with sporulation beginning on day 7. The pathogen was subsequently reisolated from sporulating lesions.

Susceptibility of wild rice cultivars. Plants of cultivars Netum, K-2, and Johnson at seedling, tillering, and flowering stages were inoculated with a conidial suspension of isolate Dg 7904 (10 ml/pot; 1,250 conidia per milliliter). Each cultivar had five replicates in the three respective growth stages (one pot represented one replicate). There were five seedlings per pot for each cultivar, whereas only two plants per pot were grown for the tillering and the flowering stages, respectively. The inoculated plants were kept in the moist chamber at 25–35 C. The reaction of plants was evaluated after 10 days using a 0–4 scale: 0 = no visible symptoms; 1 = resistant (small lesions, diameter <1 mm); 2 = moderately resistant (lesion diameter >1 mm); 3 = susceptible (zonate lesion, 0.8–1 × 1.5–2 cm); and 4 = very susceptible (large, tan to brown lesions that expanded and killed the leaves rapidly).

Host range. Fifteen species of crop plants and grasses were grown in plastic pots (two pots per species) in the greenhouse. The plants were inoculated

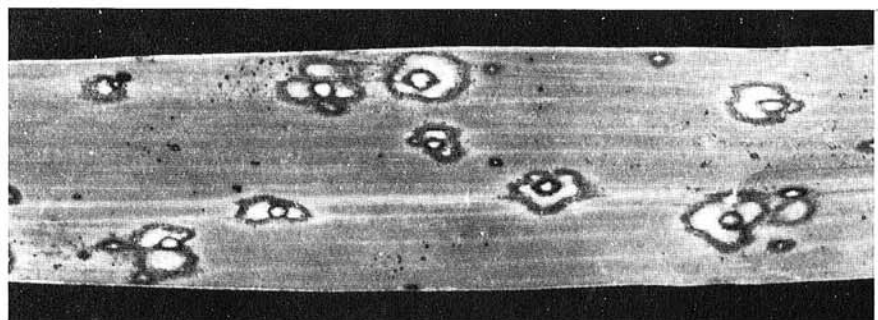


Fig. 1. Zonate eyespot symptoms on wild rice (*Zizania aquatica*) caused by *Drechslera gigantea* (×0.8).

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with 500 ml of a conidial suspension (3,500 conidia per milliliter) and then were kept in the moist chamber for 10 days. After removing the plants from the moist chamber, symptoms were described and fungal sporulation on infected leaves was observed using a dissecting microscope.

RESULTS

Symptoms and pathogen description.

The early symptoms on wild rice leaves were small, water-soaked, grayish green lesions, approximately 1 mm in diameter, with a well-defined brown margin. As the lesions enlarged, they became tan in the center with secondary lesions forming around the primary lesions, producing typical zonate eyespot symptoms (Fig. 1). Lesions were 0.8×1.5 cm, although some lesions were as large as 1×2 cm. Under humid conditions, white, prostrate mycelial strands growing centrifugally from the margin of the lesions could also be observed. Conidiophores and macroconidia could be found on both surfaces of the infected leaves.

The fungus was identified as *D. gigantea* (8,11). On V-8 medium, the fungus produced dark gray to blackish green colonies and sporulated moderately. Both microconidia and macroconidia were produced in this medium, but only macroconidia were observed on infected leaves. The macroconidia were cylindrical, hyaline to light brown, $173.4-479.4 \times 11.2-20.4$ μm (average of $319.7 \pm 65.7 \times 15.5 \pm 2.7$ μm) with 5-16 septa (mostly 6-9 septa). Both basal and apical ends were subspherical. Most germ tubes grew from basal and apical cells, although germination from intercalary cells was also common. Up to four germ tubes were often found emerging from apical or basal cells. Secondary or repetitive microconidia were hyaline to olive with one to three cells, ellipsoid, $9.5-23.8 \times 3.6-7.1$ μm (average of $13.9 \pm 3.3 \times 4.8 \pm 0.5$ μm) in size and produced in branched chains (*Cladosporium*-like).

The microconidia were borne on conidiophores from germ tubes of macroconidia or from hyphae. The conidiophores were dark brown, $153-469.2 \times 8.3-14.3$ μm (average of $256.3 \pm 58.3 \times 9.7 \pm 0.8$ μm) with two to 14 septa. The tips of conidiophores were slightly swollen and lighter in color.

Pathogenicity tests and reaction of wild rice cultivars. The pathogenicity of *D. gigantea* to wild rice plants was determined following Koch's postulates. Isolates Dg 7903, Dg 7904, and Dg 7906 produced typical zonate eyespot symptoms on cultivars Netum, K-2, and Johnson at each growth stage. Large tan to brown lesions that expanded rapidly, coalesced, and ultimately covered the whole length of the infected leaves developed on cultivar Johnson. This symptom was consistently observed at seedling, tillering, and flowering stages on approximately 20% of the population of cultivar Johnson.

Distribution and economic importance.

In summer 1979, zonate eyespot was found at the agricultural experiment station at Rosemount and at two growers' farms. The disease caused severe foliar damage on wild rice plants at one grower's farm in Aitkin County, but it occurred sporadically and was not economically important. In 1980, the disease was also recorded at the University of Minnesota Experiment Station at Excelsior, MN, and at three additional growers' farms, but it was only found on two plants in natural stands. In all cases the disease occurred only sporadically. The disease was also found in one grower's fallow paddy on volunteer wild rice plants.

Host range of *D. gigantea*. Among 15 species of plants tested in the greenhouse, lesions with secondary symptom development and fungal sporulation were observed on only four species (Table 1): wild rice, smooth brome, quackgrass, and reed canarygrass. Other plants, such as rice, wheat, barley, corn, sorghum,

barnyard grass, Manchurian wild rice, southern wild rice, and foxtail, developed only small (no more than 1 mm in diameter), nonsporulating lesions (Table 1).

DISCUSSION

D. gigantea has previously been reported mainly from the southern United States (2,6,7), where it caused zonate eyespot symptoms on many grasses. The fungus has also been reported attacking bananas (*Musa* sp.) and other hosts in Jamaica (9,10) and on rice in Colombia, Panama, and Peru (1). The occurrence of *D. gigantea* on various grasses including barnyard grass, brome smooth, quackgrass, and reed canarygrass in the northern United States has been reported by Drechsler (3-5). In Minnesota, the fungus was found only on velvet bent (*Agrostis canina* L.) but not on other grass hosts (5). Therefore, we believe that this is the first report of *D. gigantea* attacking wild rice. Additional hosts of *D. gigantea* in Minnesota were also found in 1981. The fungus was found on reed canarygrass and occasionally on smooth brome and quackgrass in five counties in southern Minnesota (*unpublished data*).

The failure of *D. gigantea* from wild rice to produce well-developed lesions and conidia on rice and barnyard grass might be attributed to genetic variations in either the pathogen or host, or both (?). Ahn (1) pointed out that rice cultivars varied in their susceptibility to *D. gigantea*.

A small proportion (about 20%) of cultivar Johnson plants showed large tan to brown lesions that expanded, and the infected leaves were killed rapidly. This type of reaction has also been observed on the cultivar Johnson plants infected with *D. oryzae* (B. de Haan) Subram. & Jain in our experiments in the greenhouse (*unpublished data*). Preliminary experiments indicated that plants showing tan to brown lesions due to *D. gigantea* also showed the same symptom when inoculated with *D. oryzae*. This symptom was observed only on the cultivar Johnson but not on Netum or K-2. The cultivar Johnson may consist of two groups of plants that differed genetically in their reaction to both *D. gigantea* and *D. oryzae*.

The reaction of wild rice plants to *D. gigantea* at the seedling stage was consistent with the reaction of plants at tillering and flowering stages. Therefore, it will be possible to do a preliminary screening for resistance to *D. gigantea* in the greenhouse using seedlings of wild rice plants. Seedlings were also used to evaluate the resistance of rice cultivars or lines to *D. gigantea* by Ahn (1).

D. gigantea infection of wild rice affecting at least 5% of the total leaf area has only been associated with cultivated paddies having dense plant stands (10-14

Table 1. Host range of *Drechslera gigantea*

Scientific name of host	Common name of host	Cultivar	Host reaction ^a
<i>Triticum aestivum</i> L.	Wheat	Angus	R
<i>Avena sativa</i> L.	Oat	Lodi	0
<i>Hordeum vulgare</i> (L.) amend. Lam.	Barley	Morex	R
<i>Zea mays</i> L.	Corn	Golden Cross Bantam	R
<i>Sorghum bicolor</i> (L.) Moench	Sorghum	Bugoff	R
<i>Oryza sativa</i> L.	Rice	Starbonnet	R
<i>Zizania aquatica</i> L.	Wild rice	Netum	S
<i>Z. latifolia</i> (L.) Stapf.	Manchurian wild rice		R
<i>Zizaniopsis miliacea</i> (Michx.) Doll & Aschers	Southern wild rice		R
<i>Phalaris arundinacea</i> L.	Reed canarygrass	Rise	S
<i>Setaria glauca</i> (L.) Beauv.	Yellow foxtail		R
<i>Echinochloa crus-galli</i> (L.) Beauv.	Barnyard grass		R
<i>Agropyron repens</i> (L.) Beauv.	Quackgrass		S
<i>Bromus inermis</i> Leyss	Smooth brome		S
<i>Digitaria sanguinalis</i> (L.) Scop.	Large crabgrass		R

^a0 = No infection; R = resistant, lesions less than 1 mm; S = susceptible, typical eyespot lesions of *D. gigantea*.

plants per 30 cm²), emergent debris from previous cropping, 72 hr of continuous 90–100% relative humidity, and temperatures of at least 30 C.

It is not known whether the fungus overwintered on wild rice plants as dormant mycelia, as is reported on many grasses (4). However, we believe that volunteer wild rice plants may perpetuate the fungus when the paddy is fallowed.

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