Scab of Cultivated Wild Rice in Minnesota Caused by Fusarium spp.

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ABSTRACT

Nyvall, R. F., Mirocha, C. J., Porter, R. A., and Percich, J. A. 1994. Scab of cultivated wild rice in Minnesota caused by Fusarium spp.

Fusarium spp. were frequently isolated from wild rice (Zizania palustris) seed grown both in wild rice paddies and in lakes. No Fusaria were isolated from wild rice seed that was stored in water immediatedly after harvest but were frequently isolated from wild rice seed dried to 20-21 % moisture then stored either at -20 or 4 C. The most common fungus isolated was F. graminearum. Other Fusaria isolated were F. anthrophilum, F. culmorum, F. moniliforme, F. sporotichioides, and F. subglutinans. Nivalenol and dioxynivalenol were produced by F. graminearum cultures but was not isolated from the seed sample the cultures were isolated from.

Cultivated wild rice (Zizania palustris L.) is grown on approximately 6,882 ha in Minnesota. Diseases have been an important factor in the cultivation of wild rice since 1961 (2). However, most research efforts have concentrated on fungal brown spot which is considered to be the most severe disease of cultivated wild rice in Minnesota.

Scab caused by Fusarium spp. is a widespread disease that affects most small grains; however, it has not been previously reported on cultivated wild rice (6). During 1993, a severe epiphytotic of scab caused extensive damage to the wheat crop throughout northern Minnesota. A sample of wild rice seed that was stored dry (20-21 % seed moisture) for several days on a laboratory bench after harvest was noticed to have pinkish to red mycelium growing over it. Most wild rice seed is immediately processed or stored by immersion in water after harvest. Other samples of seed that had been frozen immediately after harvest were examined and found to contain bleached or otherwise discolored seed. Fusarium spp. were isolated from both samples.

Fusarium spp. have not previously been reported to be isolated from cultivated wild rice seed. A Fusarium sp. had been reported isolated from the roots of cultivated wild rice grown in Minnesota in 1970 (4). However, this information lacked data and was never published. Further investigations were conducted to determine the incidence and identity of seedborne Fusaria in cultivated and lake wild rice, effect of storage conditions, and production of toxins.

MATERIALS AND METHODS

To determine the incidence of scab, samples of seed grown in 1993 were obtained from the following conditions: a) seed dried to 20-21% seed moisture for 2-3 days at 21-24 C then stored at 4 C, b) seed dried to 20-21 % moisture for 2-3 days at 21-24 C then stored at -20 C, c) seed immersed in water after harvest and stored at 4 C, and d) seed processed immediately after harvest. Processed seed was obtained from a grocery store. Processing involves placing the caryopsis at about 94 C for approximately 1.5 hr, the temperature

then rises to 117-120 C for .5 hr when the heating stops. Seed samples were obtained from both cultivated and lake-grown wild rice; however, it was not possible to obtain an equal number of samples from each source.

Isolation of fungi was done from 300 whole seeds, 100 palea and lemma, and 100 caryopsis per sample. Whole seeds were surface treated by washing in running tap water (7 C, nontreated with chlorine) for 2 hr. Seeds were then placed in a 1:1 solution (v/v) of 1% sodium hypochlorite (NaOCl) and 75% ethanol (ETOH) for 3 min, rinsed in sterile disilled water and placed on potato dextrose agar adjusted to pH 4.5 with 50% lactic acid.

Isolation from palea and lemma, and caryopsis was done by washing whole seeds in running tap water for 2 hr then separating the palea and lemma from the caryopsis with forceps and surface treating them similar to whole seeds. The washing of whole seeds for two hours facilitated the easy separation of the palea and lemma from the caryopsis. Because the palea and lemma have less mass than the whole seed or the caryopsis, different treatment times of .5, 1, 3, and 5 min in the NaOCl and ETOH solution were attempted with 3 min being the most satisfactory.

Seeds, palea and lemma, and caryopsis were incubated at 22-26 C under cool, white flourescent lights and beginning at 7 days, exposed for the next 7 days to UV light (360 nm) for 30 min daily to enhance sporulation. Cultures were identified to species after 14 days. Cultures not identified immediately were placed on APDA slants and identified when convenient.

One sample of wild rice seed obtained from a lake that had been stored at -20 C and three F. graminearum Schwabe cultures isolated from this seed source were examined for presence of nivalenol and dioxynivalenol as follows. Samples ground in a mill for 1 minute then placed in flask to which extraction solvent (acetonitril:water, 84:16 v/v) added and shaken for 1 hr. Extract filtered through Whatman # 4 filter paper and added to a charcoal column previously activated with extraction solvent. The eluate was evaporated to dryness and analysized by HPLC (Shimadzu SLC-6A at 229 nm)

Identification of Fusarium to species was done using the key by Nelson et al (5). Identification of fungi other than Fusarium spp. was done using the key by Barnett (1).

RESULTS

Seed sources stored either at -20 or 4 C were considered representative of wild rice seed sources grown in Minnesota in 1993. Fusarium spp. were consistently isolated from all seed sources regardless of their origin (Table 1). Additionally, there was no difference in incidence of Fusaria isolated from wild rice seed dried to 20-21 % moisture content and stored at either -20 or 4 C; however, Fusarium spp. were not isolated from seed stored in water after harvest. Instead Mucor sp., Geotrichum sp., and yeast (Saccharomyces spp.) were consistently isolated from all seed samples stored in water. Although Fusarium spp. were infrequently isolated from processed seed, F. graminearum was still isolated from .6 % of the seed that was sampled even after being subjected to the high temperatures necessary for the parching process.

Fusarium spp. were consistently isolated from all seed structures and were present in the caryopsis regardless of seed source or temperature of storage (Table 2). Generally, if there is a high incidence of Fusaria isolated from whole seed, Fusaria will be isolated from the palea and lemma, and caryopsis also.

Fusarium graminearum was the predominant species isolated from whole seed, palea and lemma, and caryopsis of wild rice seed (Table 3). Other Fusarium spp. that were present were F. anthophilum (A. Braun) Wollenw., F. culmorum (Wm. G. Sm.) Sacc., F. moniliforme J. Sheld., F. sporotrichioides (Sherb.), and F. subglutinans (Wollenweb. & Reinking) P. E. Nelson, T. A. Toussoun, & Marasas. However, these species were infrequently isolated. Fusarium anthrophilum, F. culmorum, F. sporotrichioides and F.

subglutinans were isolated only from seed gathered from paddies and were not isolated from lake wild rice sources.

Nivalenol and dioxynivalenol were obtained from all three F. graminearum cultures but not the seed sample from which the cultures were isolated.

Because scab symptoms on wild rice have not been readily noticed, seed samples were examined for whole seeds that displayed typical scab symptoms common on other small grains, such as wheat. Light brown, bleached, or otherwise discolored and shrunken seed was examined and isolations made. Typical symptoms are a white to light brown color; however, infected seeds do not appear to be shrunken (Fig. 1). Some seeds have a light pink discoloration that is apparently due to mycelial growth of Fusaria. Fusarium spp. were isolated from 100 % of seeds that displayed scab symptoms and from 1-26 % of seeds that did not display symptoms.

DISCUSSION

Scab of wild rice is reported here for the first time. To date, no comparable disease has been reported from white rice (Oryzae sativa L.); however, F. semitectum Berk. & Ravenel has been implicated in a necrosis that partially or totally covers the surface of kernels. A similar necrosis develops from flowering to ripening (7). Fusarium moniliforme has been commonly isolated from white rice seed where it has been implicated in reducing germination (3).

The unusally high incidence of scab on wheat in northern Minnesota in 1993 may have accentuated the incidence of Fusarium spp. on wild rice. However, conditions of high humidity that favor the develop of scab on small grains are commonly found in wild rice stands regardless if they are located in a river, lake, or cultivated paddy. Disease observation notes (Unpublished) and anecdotal evidence suggest the presence of scab symptoms on wild rice from previous years. Therefore, it is likely that scab has been a common disease of wild rice in the past regardless of where the plants were grown.

The production of the toxins nivalenol and dioxynivalenol by F. graminearum cultures isolated from wild rice seed suggests similar cautions used in placing other small grains in the human food chain may have to be excercised with wild rice. However, in a preliminary study, no toxins were found in one wild rice sample. A more extensive survey is needed to determine the presence of toxins in wild rice seed; particularly processed wild rice and wild rice grown in environmental conditions condusive to development of scab.

LITERATURE CITED

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Table 1. Percentage of Fusarium spp. isolated from whole wild rice seed obtained either from paddies or lakes and subjected to different storage conditions after harvest or processed

Source	Storage condition	Percentage Fusarium spp.[a]
Lake	-20 C, 20-21 % seed moisture	51
Paddy	-20 C, 20-21 % seed moisture	18
Lake	4 C, 20-21 % seed moisture	81
Lake	4 C, 20-21 % seed moisture	46
Paddy	4 C, 20-21 % seed moisture	62
Paddy	4 C, 20-21 % seed moisture	99
Lake	4 C, immersed in water	0
Paddy	4 C, immersed in water	0
Paddy	4 C, immersed in water	0
Lake	Processed	.6
Paddy	Processed	.6

[a] Isolations were made from 300 seeds per sample.

Table 2. Percentage of Fusarium spp. isolated from whole seed, palea and lemma, and caryopsis of wild rice seed harvested from a paddy and a lake source. Seed was dried to 20-21 % moisture for 2-3 days at 21-24 C then stored either at -20 C or 4 C.[a]

Source (Storage Temperature)	Percentage Fusarium spp. Whole seed	Palea and lemma	Caryopsis
-20 C			
Paddy	16	24	3
Lake	51	28	4
4 C			
Paddy	62	23	34
Paddy	99	96	97
Lake	81	35	54
Lake	46	36	2

[a] Isolations were made from 300 whole seeds and 100 palea and lemma, and caryopsis.

Table 3. Percentage of Fusarium spp. isolated from whole seed, palea and lemma, and caryopsis of wild rice seed gathered either from paddies or lakes

Species (source)	Whole seed [a] %	palea and lemma %	caryopsis %
Paddy			
anthrophilum	>1	0	0
culmorum	>1	0	0
graminearum	58	48	45
moniliforme	2	>1	>1
sporotrichioides	>1	0	0
subglutinans	1	0	0
Lake			
anthrophilum	0	0	0
culmorum	0	0	0
graminearum	59	33	19
moniliforme	>1	>1	1
sporotrichioides	0	0	0
subglutinans	0	0	0

[a] Isolations were made from bulked 900 whole seeds, 300 palea and lemma, and 300 caryopsis stored at -20 C and 4 C.