Influence of Temperature on Growth of Stripe-Smutted Creeping Bentgrass and on Sorus Development of Ustilago striiformis

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ABSTRACT

Temperature influenced number and length of stripe-smutted stolons proliferated from axillary node buds of stripe-smutted stolons. Under all temperature regimes, the number of stripe-smutted stolons produced exceeded that of healthy stolons, but average length of stripe-smutted stolons was reduced, except at day-night temperatures of 21.1 and 15.6 C, respectively. Day-night temperatures of 21.1 and 15.6 C were optimum for production and growth of stripe-smutted stolons; temperatures of 29.4 and 23.9 C were optimum for sorus formation. Production of stripe-smutted stolons grown at 21.1 and 15.6 C from axillary node buds of stripe-smutted stolons grown at high (37.8 and 32.2 C) and low (12.8 and 7.2 C) day-night temperatures for 40 days established that low temperature slowed intercalary formation of teliospores but did not inhibit growth of the pathogen in stolons. High temperature inhibited pathogen growth but did not eradicate it from stolons. Phytopathology 60:665-668.

Stripe smut, *Ustilago striiformis* (West.) Niessl var. *agrostidis* (Davis) Thir. & Dick., occurs on numerous cultivars of creeping bentgrass, *Agrostis palustris* Hud. (2, 3, 4). *U. striiformis* is perennial in creeping bentgrass and establishes "mycelium reservoirs" in nodes (5). From such infected nodes, hyphae grow into stolons produced from axillary node buds and keep pace with, or lag behind, stolon growth.

Research on stripe-smutted creeping bentgrass has revealed developmental characteristics of *U. striiformis* within the plant, but has not examined the epiphytology of the disease as influenced by environment. Influence of environment on growth of stripe-smutted stolons and on the pathogen within such stolons is unknown. This study was initiated to investigate the influence of temperature on growth of stripe-smutted creeping bentgrass and on development of the stripe-smut pathogen within the grass.

MATERIALS AND METHODS.—Cultivars selected for study were Arlington C-1, Congressional C-19, Old Orchard C-52, and Penncross. All cultivars, controls and smutted, were vegetatively propagated in a steamed 2:1 shredded sphagnum-soil mixture at greenhouse temperatures ranging from 15.6 to 28.9 C.

Experiments for determining influence of temperature on growth of stripe-smutted stolons were divided into four treatments: day-night temperatures of 12.8 and 7.2 C; 21.1 and 15.6 C; 29.4 and 23.9 C; and 37.8 and 32.2 C; respectively. Percival PT-80 Phytotrons with 76 to 95% relative humidity (RH), approximately 4,800 ft-c (51.668×10^3 lux), and a 14-hr daylength were used for all experiments. Smutted stolons were propagated in each treatment from 3-cm lengths of stolon, each possessing one axillary node bud. Each treatment contained 48 replicates grown in plastic-flat inserts divided into 48 compartments (Cell-Pak Inserts, Geo. J. Ball, Inc.), and the inserts were placed in plastic flats. Stolons were grown in each treatment for 40 days; then new stolons were counted and measured to the nearest 0.5 cm. Branch stolons and stolons less than 3 cm in length were not included. At the end of 40 days, replicates of each treatment were also examined for stolons with mature or immature sori. Plants from each treatment were then removed from the Phytotrons to a 15.6 to 28.9 C greenhouse, and further observed for sorus development. Observations were made at 70 and 100 days after initiation of treatments. Control plants were maintained under the same conditions.

Experiments to determine influence of high and low temperatures on growth of *U. striiformis* and development of sori were conducted by establishing 3-cm lengths of stripe-smutted stolons as previously described. Forty-eight smutted stolon lengths of each variety were grown at day-night temperatures of 12.8 and 7.2 C and at 37.8 and 32.2 C, respectively. Surviving stolons were grown at the respective temperatures for 40 days; each replicate was then examined for stolons with sori. Twelve stolons with 3 nodes each, not displaying sori, were selected from each of the respective day-night temperature treatments. Stolons were then dissected into 3 lengths, each possessing one node, then lined-out in flats and grown at day-night temperatures of 21.1 and 15.6 C. Sorus development was observed in stolons produced from each axillary node bud over a 60-day period. Control plants consisted of maintaining the remaining plants at the original temperature ranges and recording sorus development for the 100-day period.

RESULTS.—*Influence of temperature on production and growth of stolons.*—Mortality of smutted stolons was low at all day-night temperatures, but increased slightly at 37.8 and 32.2 C (Table 1). With one exception (Arlington C-1, 29.4 and 23.9 C), axillary node buds of stripe-smutted stolons of all varieties at all temperature ranges produced more stolons than did unsmutted stolons. An analysis of variance revealed that the number of stripe-smutted stolens produced at all temperatures was significantly greater than that of all the controls ($P < .01$). On the basis of stolon production, day-night temperatures of 21.1 and 15.6 C were optimum for smutted and unsmutted stolons. Day-night temperatures of 12.8 and 7.2 C and 37.8 and 32.2 C

TABLE 1. Number and length of stolons produced by healthy and stripe-smutted creeping bentgrass (*Agrostis palustris*) as influenced by temperature over a period of 40 days[a]

Temp range (76-95% RH)		Surviving stolons of 48 replicates		No. stolons produced[b]		Avg length of stolons (cm)	
Day Night	Cultivars	Control	Smutted	Control	Smutted	Control	Smutted
12.8- 7.2 C	Arlington C-1	47	48	78	116	6.3	6.1
	Congressional C-19	47	48	80	102	6.4	4.7
	Old Orchard C-52	48	48	78	136	6.4	6.2
	Penncross	47	48	117	119	7.2	5.2
21.1-15.6 C	Arlington C-1	48	48	156	160	21.5	21.4
	Congressional C-19	48	48	133	160	27.1	26.7
	Old Orchard C-52	48	48	168	274	21.4	27.9
	Penncross	48	48	215	289	20.8	21.4
29.4-23.9 C	Arlington C-1	48	48	114	113	22.6	14.4
	Congressional C-19	48	48	106	132	20.4	19.9
	Old Orchard C-52	48	48	122	163	25.9	23.2
	Penncross	48	46	163	197	11.3	8.4
37.8-32.2 C	Arlington C-1	47	46	55	75	7.6	6.1
	Congressional C-19	45	32	56	57	6.4	4.9
	Old Orchard C-52	47	43	53	95	8.3	6.7
	Penncross	46	42	66	67	6.2	4.3

[a] All stolons in each replicate were propagated from axillary node buds on 3-cm lengths of stolon.
[b] Stolons measuring less than 3 cm not included.

reduced production of smutted and unsmutted stolons.

Average stolon length of unsmutted plants exceeded smutted plants at all day-night temperatures except 21.1 and 15.6 C. In this temperature regime, smutted stolons of Old Orchard C-52 and Penncross outgrew controls by an average of 6.5 and 0.6 cm, respectively (Table 1). All other unsmutted stolons at all temperature ranges were an average of 0.2 to 8.2 cm longer than smutted stolons. No leaf necrosis was observed among smutted or unsmutted plants under any temperature range during the 40-day experiment.

Influence of temperature on sorus development.— After 40 days' growth at the respective temperatures, plants were examined for mature and immature sori. Stolons displaying sori were recorded, and all plants were then placed in a greenhouse at 15.6 to 23.9 C and again examined for sori at 70 and 100 days. Six to 28 replicates (12.5 to 58.3%) of cultivars grown at day-night temperatures of 12.8 and 7.2 C displayed visible sori within 40 days (Table 2). Sorus formation was greatest in replicates at temperatures of 21.1 and 15.6 C and 29.4 and 23.9 C. Optimum sorus development within 40 days occurred at 29.4 and 23.9 C, averaging 38 smutted replicates/cultivar. Except for some escapes, all replicates of all cultivars at day-night temperatures of 12.8 and 7.2 C, 21.1 and 15.6 C, and 29.4 and 23.9 C displayed smutted stolons at 100 days after removal to the greenhouse. Sorus development was reduced at the 37.8 and 32.2 C temperature range; 0 to 2 replicates (0 to 4.3%) displayed sori among the cultivars with no appreciable increase at 70 days (Table 2).

TABLE 2. Influence of temp on development of stripe smut sori within infected stolons of creeping bentgrass (*Agrostis palustris*)

Temp range (76-95% RH)		Sori at the end of 40 days[a]	Sori after removal to 15.6 to 23.9 C greenhouse[a]	
Day Night	Cultivars		70 Days	100 Days
12.8- 7.2	Arlington C-1	25	35	46
	Congressional C-19	10	37	48
	Old Orchard C-52	6	39	48
	Penncross	28	48	48
21.1-15.6	Arlington C-1	37	41	46
	Congressional C-19	35	48	48
	Old Orchard C-52	23	39	45
	Penncross	40	48	48
29.4-23.9	Arlington C-1	44	47	47
	Congressional C-19	48	48	48
	Old Orchard C-52	38	48	48
	Penncross	22	35	46
37.8-32.2	Arlington C-1	2	4	17
	Congressional C-19	2	2	22
	Old Orchard C-52	1	3	25
	Penncross	0	0	31

[a] 48 Replicate stolons used; number of replicates showing mature and immature sori were recorded.

At 100 days, however, smutted replicates had increased 17 to 31 (36.9 to 73.8%) among cultivars.

Influence of temperature on growth of pathogen.—Pathogen development in smutted stolons grown at 12.8 and 7.2 C and 37.8 and at 32.2 C for 40 days and then dissected into 3 lengths (one axillary node bud/length) and grown at 21.1 and 15.6 C displayed distinctly different reactions to the respective temperatures (Table 3). With the exception of Penncross, all axillary node buds of stripe-smutted stolons grown for 40 days at 12.8 and 7.2 C produced stripe-smutted stolons at 21.1 and 15.6 (Table 3). Axillary node buds grown at 21.1 and 15.6 C from stripe-smutted stolons grown for 40 days at 37.8 and 32.2 C produced 1 to 4 smutted stolons from basal nodes (node 1) among cultivars; 0 to 2 were produced from node 2; and none from the terminal node (Table 3). At the end of 100 days, plants maintained at 37.8 and 32.2 C remained unsmutted; plants at 12.8 and 7.2 C were 97% smutted.

DISCUSSION.—Leaf necrosis of stripe-smutted plants was not directly related to temperature as has been reported for Kentucky bluegrass (*Poa pratensis*) (1, 6). Mortality increased slightly at day-night temperatures of 37.8 and 32.2 C; however, leaf necrosis was nonexistent at all temperature ranges. Temperature stress did not induce leaf necrosis as long as humidity and water were maintained sufficiently high to avoid wilting. Leaf necrosis and mortality of stripe-smutted creeping bentgrass plants probably results from a combination of temperature, humidity, and water stress.

Production of stripe-smutted stolons from smutted axillary node buds exceeded controls at all day-night temperatures, with the exception of Arlington C-1 at 29.4 and 23.9 C (Table 2). Greater production of stripe-smutted stolons may be indicative of growth regulators produced by the pathogen, or of interference with normal growth physiology of stolons due to the presence of the pathogen. With the exception of Old Orchard C-52 and Penncross at 21.1 and 15.6 C, however, check stolons had an average length greater than smutted stolons (Table 1). It appears that this temperature range is optimum for mutual development of plant and pathogen; i.e., smutted stolons appear to grow as well as unsmutted stolons.

Reduced sorus development at 40 days at 12.8 and 7.2 C and at 37.8 and 32.2 C, and subsequent development of sori in these treatments within 100 days in the greenhouse at 15.6 to 23.9 C, were indicative of two distinct reactions of the pathogen to high and low temperatures (Table 2). Stripe-smutted stolons produced from axillary node buds of stripe-smutted stolons grown at low temperatures (12.8 and 7.2 C) for 40 days established that, under conditions of low temperature, *U. striiformis* grows throughout the plant, but intercalary formation of teliospores is slowed (Table 3). This was established by production of stripe-smutted stolons propagated at 21.1 and 15.6 C from axillary node buds of stripe-smutted stolons (selected prior to appearance of sori) previously exposed to low temperature; stripe-smutted stolons were produced from each of 3 nodes (only stolons possessing three nodes were used) of 12 stolons selected from each cultivar (Table 3). If nodes of stripe-smutted stolons grown at low temperatures for 40 days were not colonized by the pathogen, the new stolons produced would have been unsmutted. Failure of axillary node buds of stripe-smutted stolons grown at high temperatures (37.8 and 32.2 C) for 40 days to produce stripe-smutted stolons when grown at 21.1 and 15.6 C was indicative of high temperature inhibition of the pathogen (Table 3). No stripe-smutted stolons were produced from terminal nodes (node 3); 0 to 2 stripe-smutted stolons were produced from node 2, and 1 to 4 from basal nodes (node 1) among the cultivars when grown at 21.1 and 15.6 C (Table 3). This was indicative of pathogen inhibition; if the pathogen was present in nodes of stripe-smutted stolons grown at high temperatures, the axillary node buds of the stolons would have produced stripe-smutted stolons when grown at 21.1 and 15.6 C. High temperature inhibition of pathogen growth does not appear, however, to eradicate the pathogen from stolons, as indicated by gradual recovery of stripe-smutted stolons from high-temperature treatments (Table 2).

Three practical implications are evident from the research. Firstly, it is probable that stripe-smutted creeping bentgrass will survive periods of high temperature in golf greens, where water is adequate and where humidity is relatively high. Secondly, during periods of high temperature, the pathogen will probably remain in

TABLE 3. Growth and sorus development of *Ustilago striiformis* in creeping bentgrass (*Agrostis palustris*) stolons grown at 21.1 and 15.6 C from axillary node buds of stripe-smutted stolons grown 40 days at high (37.8 and 32.2 C) and low (12.8 and 7.2 C) day-night temperatures[a]

	No. axillary node buds from which stripe-smutted stolons were produced[b]					
	Basal node[b]		Middle node		Terminal node	
Cultivars	Low temp	High temp	Low temp	High temp	Low temp	High temp
Arlington C-1	12	1	12	0	12	0
Congressional C-19	12	4	12	2	12	0
Old Orchard C-52	12	3	12	1	12	0
Penncross	12	2	10	0	8	0

[a] Day-night temperatures of 21.1 and 15.6 C optimum for growth of stripe-smutted stolons (Table 1).

[b] Data collected from 12 stolons of each cultivar not displaying sori; each stolon possessed 3 nodes from which stolons were propagated.

a semidormant state within the stolons. Thirdly, proliferation of stripe-smutted stolons will be at a maximum during the cool periods of the spring and fall.

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