

# Comparative Morphology and Development of *Poa pratensis* Infected by *Ustilago striiformis* and *Urocystis agropyri*

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## ABSTRACT

Morphological and developmental characteristics of *Poa pratensis* 'Merion' varied in response to infection by *Ustilago striiformis* (stripe smut) and *Urocystis agropyri* (flag smut). Laminae of stripe- and flag-smutted plants were more erect than those of control plants. Laminae of stripe-smutted plants were wider than those of control and flag-smutted plants. Lamina thickness was equal for stripe- and flag-smutted plants, and greater than that of control plants. Stripe-smutted plants were taller than controls, whereas, flag-smutted plants were noticeably stunted. Total branches produced by stripe- and flag-smutted plants were less than that of control plants. Stripe-smutted plants showed the greatest reduction

in total branching. Flag-smutted plants produced proportionally more intravaginal branches than did either control or stripe-smutted plants, indicating premature axillary crown bud break in response to *U. agropyri*. Extravaginal branching was proportionally higher among control and stripe-smutted plants. Total dry-matter production of flag-smutted plants was less than that of control and stripe-smutted plants. Root dry weight of stripe- and flag-smutted plants was less than that of controls; however, reduction of roots among flag-smutted plants was greater than among stripe-smutted plants. Phytopathology 60:1794-1797.

Leaf smuts of Kentucky bluegrass, *Poa pratensis* L., are caused by two different pathogens, *Ustilago striiformis* (West.) Niessl var. *poae* Thir. & Dick. (stripe smut) and *Urocystis agropyri* (Preuss) Schroet. (flag smut). Both pathogens produce linear sori in leaf mesophyll that may extend the entire length of the leaf (4, 5, 8, 12, 13, 15). Sori mature and rupture the leaf epidermis; leaves become necrotic, and release teliospores. Similar leaf symptoms resulting from these pathogens make macroscopic field identification of the respective diseases difficult. It has been reported that the brownish color of *U. agropyri* sori in early spring may provide a fairly reliable means of distinguishing flag-smutted plants from stripe-smutted plants (14). It is now generally recognized that this characteristic is not reliable and that positive diagnosis requires microscopic examination of teliospores (8, 11). Absence of symptomological differences in mowed *P. pratensis* infected by these pathogens has stimulated research on morphological and developmental characteristics of infected plants. Preliminary studies of field-collected *P. pratensis* infected by these pathogens showed differences in leaf and branching characteristics indicative of the pathogen present (7). This initial research did not, however, take into account potential variability of morphological and growth characteristics inherent in field-grown *P. pratensis* of unknown origin. The research reported herein was initiated to determine if morphological and developmental differences existed also in a selected cultivar (Merion) of *P. pratensis* with known growth characteristics when infected by *U. striiformis* or *U. agropyri*.

**MATERIALS AND METHODS.**—Merion Kentucky bluegrass was used for all studies. Merion is characterized by a wide lamina-to-sheath angle, and is a plant with vigorous vegetative branching and rooting characteristics (10). All control and diseased plants were col-

lected from 2-year-old Merion stock plants grown in steamed soil; the soil from which diseased plants were collected had been infested with teliospores of *U. striiformis* or *U. agropyri*. One hundred stripe- and flag-smutted crowns each, without lateral branches, were potted individually in a steamed, 2:1 loam-peat soil mixture in 3-inch pots. Each plant was cut back to a height of 7 cm. One hundred control plants were established in the same way. All plants were grown in the greenhouse at 12.5 to 29.0 C, and were provided a 16-hr day with supplemental incandescent light. Plants were examined for morphological and developmental differences after 60 days.

The influence of the respective pathogens on lamina-to-sheath angle, lamina width, and lamina thickness was studied. Lamina-to-sheath angle was determined by direct measurement with a 4-inch protractor. Three measurements were taken on each of 50 control, stripe-, and flag-smutted plants. Acute angle measurements were taken at the midvein of laminae to the nearest 5 degree interval; more precise measurements were not feasible because of lamina flexibility.

Lamina width and thickness were determined from 30 laminae, each taken from control, stripe-, and flag-smutted plants. Maximum lamina width was determined by direct measurement to the nearest 0.5 mm. Lamina thickness was determined from free-hand cross-sections placed in lactophenol on microscope slides and measured with an ocular micrometer. Two measurements were made on diseased laminae: max thickness of laminae at sori and max thickness between sori (midvein excluded from measurement). Maximum length of plants was determined by measuring the longest living tiller (from soil surface to leaf apex) to the nearest 0.5 cm.

Differences in branching characteristics were determined by counting total branches produced by each plant. Branches were separated into intra- and extra-

vaginal types to further determine influence of the respective pathogens on development of branch types (6).

Total plant wt, leaf wt, and root wt was determined for all plants. Each plant was washed and air-dried for 48 hr. Roots were separated from crowns and rhizome nodes; leaves, crowns, and rhizomes were combined for leaf wt. Leaves and roots of each plant were then dried for 48 hr in a forced-air oven at 58 C.

**RESULTS.—Leaf characteristics.**—Distinct differences in leaf characteristics occurred between control and smutted plants and between stripe- and flag-smutted plants. Mean acute lamina-to-sheath angle of control plants was 38.2 degrees, which is characteristic of the wide, open growing habit of Merion (Fig. 1,2). Mean lamina-to-sheath angles of both stripe- and flag-smutted plants were greater than those of control plants (Table 1). The very wide acute angle of stripe-smutted plants resulted in narrow, upright plants relative to control and flag-smutted plants (Fig. 1, 2).

Lamina width and thickness varied between control and smutted plants and between stripe- and flag-smutted plants. Control and stripe-smutted plants had mean lamina widths greater than those of flag-smutted plants (Table 1). Mean lamina thickness of stripe- and flag-smutted plants was greater than that of controls (Table 1). There appeared to be no differences between stripe- and flag-smutted plants in over-all leaf thickness; however, lamina thickness of flag-smutted plants was slightly greater at sori and less between sori.

**Maximum tiller lengths.**—Tillers of control, stripe-, and flag-smutted plants differed markedly in max elongation (Fig. 2). Average max length of control tillers was less than that of stripe-smutted tillers and greater than that of flag-smutted tillers (Table 1). Although both diseases resulted in plants with tiller lengths different from those of controls, the greatest differences were between stripe- and flag-smutted tillers. Flag-smutted tillers were distinctly dwarfed, whereas stripe-smutted tillers were stimulated (Fig. 2, Table 1).

**Branching characteristics.**—Branching characteristics of smutted plants were distinctly altered. Smutted plants produced fewer branches than controls; stripe-smutted plants had fewer branches than flag-smutted plants and approximately one-half that of control plants (Table 1). Proportionally, flag-smutted plants produced more intravaginal branches than did either control or stripe-smutted plants; i.e., 93.9%, 69.6%, and 62.5%, respectively. Extravaginal branching was proportionally higher in stripe-smutted plants (37.5%), as compared to control plants (30.4%) and flag-smutted plants (6.4%).

**Dry-matter production.**—Total dry-matter production was lower among stripe- and flag-smutted plants as compared to control plants. Total dry wt of stripe- and flag-smutted plants was 35.3% and 29.4%, respectively, of control plants (Table 1). Proportionally, diseased plants produced more leaf dry-matter than did control plants. Total leaf dry wt of control, stripe- and flag-smutted plants was 70.1%, 81.2% and

TABLE 1. Effect of *Ustilago striiformis* and *Urocystis agropyri* on various morphological and development characteristics of 60 day old Merion Kentucky bluegrass (*Poa pratensis*) plants

Morphological and developmental characteristics observed	Treatments					
	Control plants		<i>U. striiformis</i> -infected plants (stripe smut)		<i>U. agropyri</i> -infected plants (flag smut)	
	Range	Means	Range	Means	Range	Means
Lamina-to-sheath angle, degrees <sup>a</sup>	20-70°	38.2°	40-85°	64.7°	35-75°	52.7°
Maximum tiller length, cm <sup>b</sup>	9.0-23.5	15.4	9.5-30.0	19.5	6.0-15.5	11.6
Lamina width and thickness, mm <sup>c</sup>						
Lamina width	3.0-5.0	4.3	3.5-6.0	4.8	2.0-4.5	3.1
Lamina thickness	0.2-0.3	0.2	0.2-0.4	0.3	0.2-0.5	0.3
Lamina thickness at sori			0.2-0.4	0.3	0.3-0.5	0.4
Lamina thickness between sori			0.2-0.4	0.3	0.2-0.3	0.2
Branching characteristics, no. <sup>d</sup>						
Total branches	17-46	29.9	7-26	15.2	10-44	26.5
Intravaginal branches	9-32	20.9	4-18	9.5	9-43	24.9
Extravaginal branches	3-18	9.1	2-12	5.7	0-5	1.7
Dry-matter production, g <sup>e</sup>						
Total plant wt	0.4-2.8	1.74	0.20-1.3	0.64	0.20-1.1	0.52
Leaf wt	0.3-1.9	1.22	0.20-1.1	0.52	0.10-0.9	0.46
Root wt	0.1-1.0	0.52	0.04-0.2	0.11	0.01-0.2	0.06

<sup>a</sup> Based on 150 leaf samples each; all means significantly different (.05).

<sup>b</sup> Based on 100 tiller samples each; all means significantly different (.05).

<sup>c</sup> Based on 30 lamina samples each, except for leaf thickness of smutted plants which were based on combined at-and-between sori thickness (60 samples). Mean width of stripe-smutted plants was significantly greater (.05) than that of controls and flag-smutted; controls were significantly greater (.05) than flag-smutted. Stripe-smutted and flag-smutted leaf thickness was significantly greater (.05) than controls.

<sup>d</sup> Based on 100 plants each. Mean total branch production for controls was significantly greater (.05) than diseased plants; mean total branch production of flag-smutted plant was significantly greater (.05) than stripe-smutted.

<sup>e</sup> Based on 100 plants each. Mean differences between control and smutted plants and between stripe- and flag-smutted plants significantly different (.05).

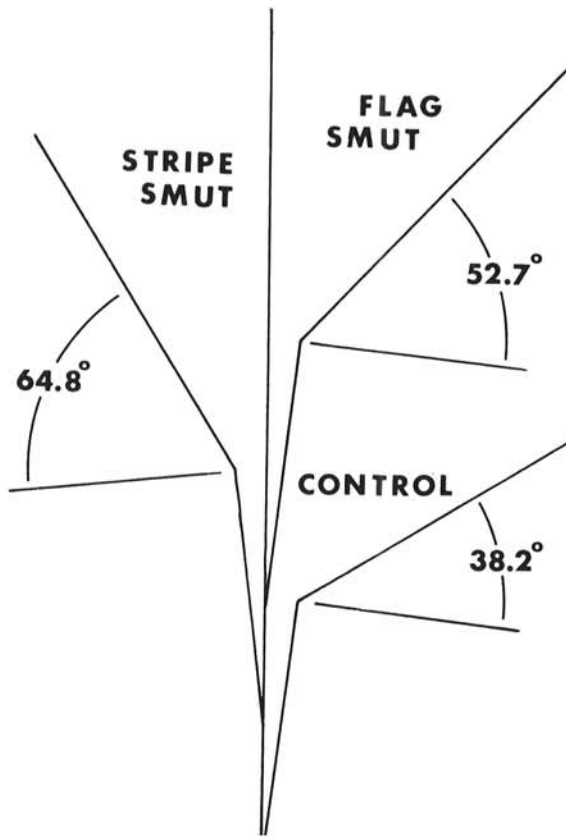


Fig. 1. Schematic diagram showing changes in lamina-to-sheath angle of Merion *Poa pratensis* in response to infection by *Ustilago striiformis* (stripe smut) and *Urocystis agropyri* (flag smut).

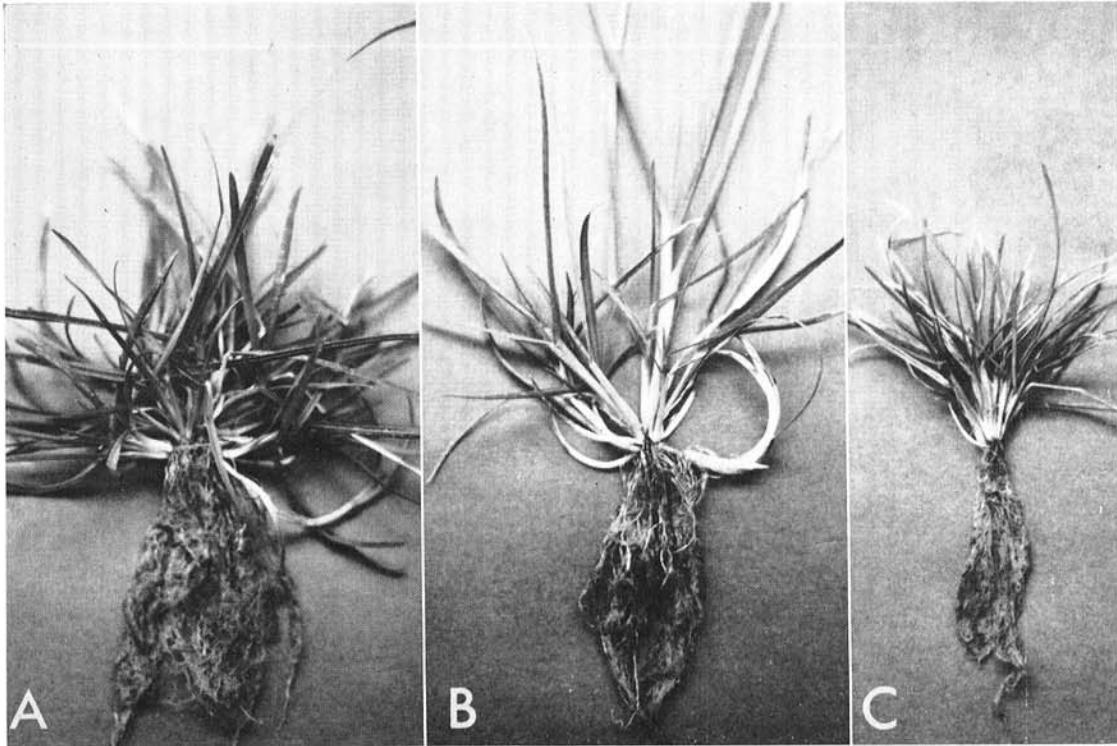
88.5% of their respective total dry wt. The most obvious differences in dry-matter production between control and smutted plants and between stripe- and flag-smutted plants was in root dry wt (Table 1). Root dry wt was less in both stripe- and flag-smutted plants, but was much less among flag-smutted plants. Proportionally, root dry wt of flag-smutted plants was 11.5% of their total wt; root dry wt among stripe-smutted and control plants was 17.2% and 29.8%, respectively.

DISCUSSION.—Morphological and developmental differences in *P. pratensis* infected by *U. striiformis* or *U. agropyri* were shown to be in direct response to the respective pathogens. Both pathogens reduced total branch production; stripe-smutted plants produced wide-leaved, elongated tillers and flag-smutted plants produced fine-leaved, dwarf tillers (Fig. 2). These growth characteristics suggest a possible growth-regulator imbalance in diseased plants. Various genera of Basidiomycetes are implicated in increased or decreased levels of growth-regulators in plants they infect (3). Within the genus *Ustilago*, at least two species have been studied. *Ustilago maydis* has been shown to increase auxin, probably IAA (16), in *Zea mays* (9); and *U. hypodytes* causes stem elongation in *Bromus erectus*

suggestive of increased auxin production (3). If symptoms produced by *P. pratensis*, in response to *U. striiformis* and *U. agropyri*, are due to growth-regulator imbalances, they are atypical of those responses usually associated with auxins and gibberellins (GA). Elongation of stripe-smutted tillers (primarily laminae, not internodes) could be a typical GA response; however, lateral branch production was lowest among stripe-smutted plants; such inhibition of lateral branching is atypical of GA (3). The dwarfing of flag-smutted tillers could be associated with deficiencies in auxin and/or GA; however, root development among flag-smutted plants was greatly reduced, a condition commonly associated with excessive auxin (3). On the basis of research contained herein, this discussion is speculative; however, the symptomological differences found between stripe- and flag-smutted plants and knowledge of the ability of some Basidiomycetes to modify growth-regulator physiology strongly suggest that growth-regulator research is warranted.

Although total branch reduction probably was the direct result of altered growth physiology induced by the respective pathogens, the influence of the pathogens on intra- and extravaginal branch types was believed to be a secondary indirect effect. Extravaginal branching in *P. pratensis* results from axillary crown buds, especially those associated with older phytomers (a piece of stem with a leaf on the upper end and a bud at the lower end), growing through surrounding dead leaf sheaths; intravaginal branches, however, arise within the confines of a living leaf sheath (6). It is possible, therefore, that inhibition of axillary crown bud development, which was responsible for total branch reduction in stripe-smutted plants, may have indirectly caused these plants to produce proportionally more extravaginal branches. Stripe-smutted tillers usually had large crowns with numerous axillary crown buds of different ages surrounded by leaf sheaths at various stages of senescence and necrosis. When axillary crown bud development occurred, buds grew through the senescing or necrotic sheaths to develop extravaginal branches. These same developmental characteristics may apply to the proportionally greater number of intravaginal branches produced by flag-smutted plants. The flag-smut pathogen caused premature break of axillary crown buds, resulting in a predominance of intravaginal branches. Tillers did not grow long enough before axillary crown bud development to allow adequate senescence or necrosis of sheaths for extravaginal branching. Therefore, the actively growing sheaths caused axillary buds to develop within the confines of living sheaths and to produce intravaginal branches.

The general inhibition of total branch and root growth in smutted plants was reflected in reduced dry-matter production. Although flag-smutted plants produced more branches than did stripe-smutted plants, total plant and leaf wt of flag-smutted plants were less (Table 1). This supports the explanation of growth characteristics relative to branch types produced. That leaf dry-matter of stripe-smutted plants exceeded that



**Fig. 2.** Comparative development of Merion *Poa pratensis* infected with *Ustilago striiformis* (stripe smut) and *Urocystis agropyri* (flag smut). **A**) Control plant. **B**) Stripe-smutted plant showing elongation of coarse textured leaves and tillers; total branching is reduced, but with a proportionally large number of extravaginal branches. **C**) Flag-smutted plant showing distinct dwarfing of fine textured leaves and tillers. Excessive intravaginal branching is reflected in the dense appearance of the plant. Root production is noticeably reduced.

of flag-smutted indicates that branches of stripe-smutted plants were heavier due to production of more leaves per tiller. The most marked differences in dry-matter production occurred in root wt (Table 1). Both smuts reduced root development; however, root wt was greatly reduced among flag-smutted plants. Such reduction in root development is seemingly characteristic of the flag-smut pathogen in that similar observations have been reported in wheat (1, 2). This reduction in roots of flag-smutted plants may also account for my observation that flag-smutted plants seem to become necrotic much faster than do stripe-smutted plants under conditions of high temp and drought.

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