PATHOGENICITY OF FUSARIUM SPECIES FROM ALSIKE CLOVER

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Abstract

Three pathogenic species of Fusarium were obtained from alsike clover affected by root rot. Of these, F. oxysporum Schlecht. appeared most commonly followed by F. &-morum (W. G. Sm.) Sacc. and F. avenaceum (Fr.) Sacc. On the other hand, their pathogenicity was not related to their frequency since F. avenaceum was more virulent to alfalfa and alsike clover than the other two species isolated, and F. culmorum was the most virulent on ladino clover. Red clover was relatively non-susceptible to the Fusarium species. The significance of the disease distribution in Quebec is discussed and its occurrence in fields planted with red clover shows that it should receive more attention with respect to the competition of plant species. The outbreak of root rot in Quebec is significant if alsike is to be kept in the mixture with timothy and red clover, The need for further investigations along this line is suggested.

Resume

Trois espkces de Fusarium pathogbnes ont été isolés de plants de trbfle alsike malades. F. oxysporum Schlecht. a été isolé le plus souvent, suivi de F. culmorum (W. G. Sm.) Sacc. et de F. avenaceum (Fr.) Sacc. Cependant, la pathogénicité de ces isolats fut sans rapport avec leur fréquence, i.e., F. avenaceum fut plus pathogkne à la luzerne et au trkfle alsike que les deux autre espbces, tandis que F. culmorum le fut plus au trbfle ladino. Le trèfle rouge semblait, par contre, resistant au Fusarium isolés du trbfle alsike. L'auteur traite de l'importance de la distribution de cette maladie et émet l'hypothkse que sa presence dans les champs de tr'efle rouge peut être due à la competition qui pourrait exister entre celui-ci et le trkfle alsike. L'éclosion de cette maladie au Québec est importante si on continue de semer cette plante en mélange avec le mil et le trkfle rouge. L'auteur sugg'eredonc que l'on devraitporter plus d'attention à ce problbme.

Introduction

Alsike clover, Trifolium hybridum L. is an important forage legume in Quebec, although its use fulness is limited by a lack of persistance of stands. This plant is commonly sown in mixture with red clover and timothy in the proportions of 8 lb timothy, 5 lb red clover, and 2 lb alsike clover per acre. During a disease survey made throughout the province of Quebec in the summer of 1965, alsike clover plants were observed to be dying following attack by Fusarium spp. Specimens of wilted plants were collected from 23 farm fields. Diseased and dead plants have been foundinmost of the regions where alsike clover is grown. This disease of alsike clover has not heretofore been reported in Canada. This paper presents the results of the survey and of pathogenicity tests of the isolates on forage legumes.

Materials and methods

Isolates of Fusarium were identified according to Gordon's system (3). Eightpercent of the isolates (3 isolates) were identified as Fusarium avenaceum (Fr.) Sacc., 19% (8 isolates) as Fusarium &-morum (W. G. Sm.) Sacc., and 73% (26 isolates) as Fusarium oxysporum Schlecht. All the isolates were kept on potato sucrose agar.

The legumes studied in experiments on the host range and pathogenicity included alfalfa (Medicago sativa L. var. 'Vernal'), alsike clover (Trifolium repens L. var. 'Pilgrim'), and red clover (Trifolium pratense L. var. 'Dollard'). Twenty plants of each species were inoculated with each isolate of Fusarium.

The Fusariumisolates were grown on a sterilized 3:1 mixture of soil and sand plus 10% by weight of corn meal, in Erlenmeyer flasks, They were allowed to grow at room temperature for two weeks before being used to inoculate the plants. Sterile distilled water was added, and the cultures were gently shaken. The resulting suspension contained spores, some mycelial fragments, and a few soil particles.

Inoculation consisted of dipping the roots of the seedlings for 10 minutes in the spore suspension containing approximately 100,000 cells per ml. All the experiments were terminated 60 days after transplanting.

Method of assessing plant damage

The disease severity rating was based on the damagecaused to tap root and secondary roots and recorded as follows: 0 = no root discoloration, no disease in this case: 1 = trace of root browning, mostly on secondary roots; 2 = moderate root browning, considerable necrosis on secondary roots: 3 = severe rottingof secondary and tap roots: 4 = severe rotting of the entire root system; 5 = death of the plant. The disease rating presented in Table 1 is the mean rating of 20 plants per isolate, i.e. these data represent the mean disease rating of 60 plants in the case of F. avenaceum, 160 plants in the case of F. culmorum, and 520 plants in the case of F. oxysporum. The percent plants killed in the final recording is also based on the same number of plants,

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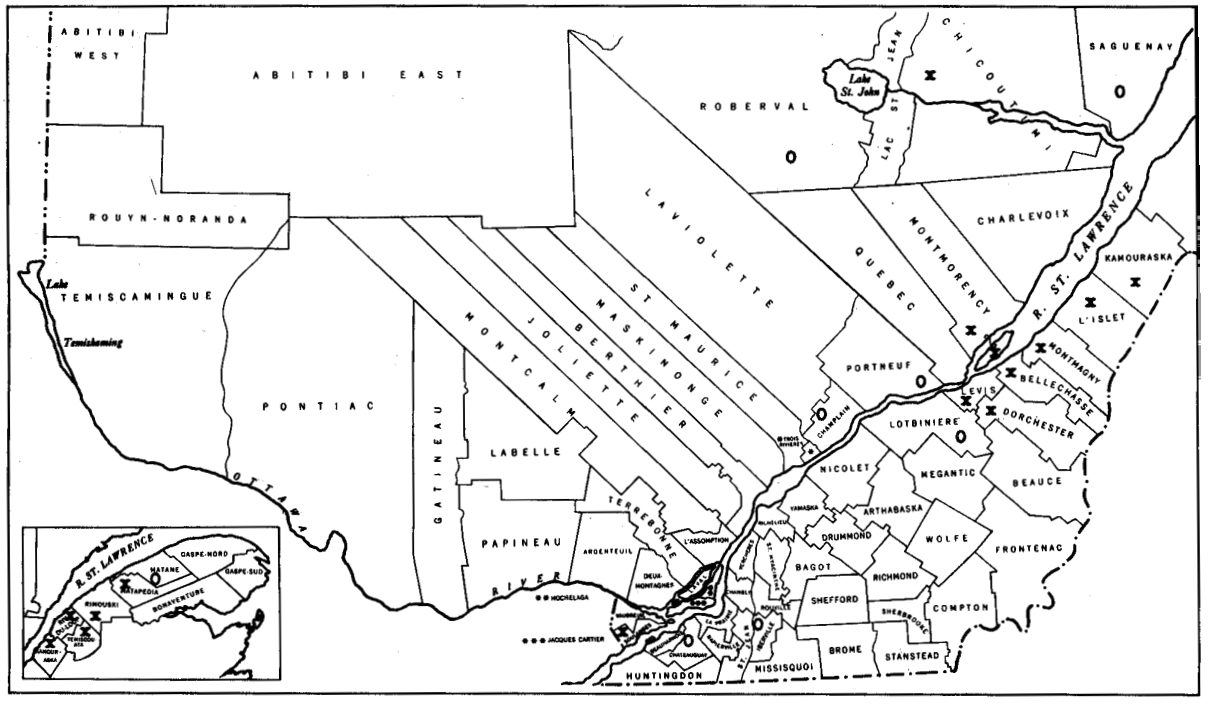

Figure 1. Map of the province of Quebec showing the distribution by counties of alsike clover and its disease caused by Fusorium spp.

O: alsike clover not attacked by Fusorium spp.

x: alsike clover attacked by Fusorium spp.

Results

Figure 1 shows that alsike clover plants attacked by Fusarium spp. were found only from Levis to Bonaventure counties on the south shore of the St. Lawrence River and in two other counties, Chicoutimi and Montmorency on the north shore. In Montmorency the disease was found on Orleans Island.

The results presented in Table 1 indicate that the four plant species so far tested show varying degrees of susceptibility to attack by Fusarium spp. F. avenaceum is the most pathogenic to alfalfa followed by F. culmorum and F. oxysporum in that order. Alsike clover is more susceptible to F. avenaceum than to F. culmorum and F. oxysporum. Although the data obtained with F. culmorum and F. oxysporum do not lend themselves to statistical analysis, the differences do not appear to be particularly significant. Ladino clover is quite susceptible to attack by F. culmorum followed in that respect by F. avenaceum and F. oxysporum. Whereas no plants of red clover were killed by any of the Fusarium spp. tested the disease intensity was higher following inoculation with F. avenaceum than with the other two Fusarium spp.

Discussion

Although root rot ranks among the most important diseases of forage legumes, the Fusarium spp. inciting it are generally considered as weak pathogens. They cause little damage to vigorously growing plants, but are extremely destructive to plants lacking vigor. Under field conditions plants become predisposed to attack in many ways. In Quebec, root reserves are largely consumed during the long winter dormancy, so that the plants are less able to resist attack. In forage legume growing areas, fungus pathogens, moisture, drought, and most important, excessive grazing in the fall, predispose plants to attack. The Fusarium spp. were isolated from alsike clover collected in regions where the rainfall and consequently the soil moisture is relatively high.

Table 1. Virulence of three Fusarium isolates on four species of legume seedlings as indicated by disease severity ratings and percent plants killed.

Isolate and plants tested	Disease severity rating	Percent plants killed
F. avenaceum		
Alfalfa	3.8	76.2
Alsike clover	2.9	55.5
Ladino clover	1.4	13.3
Red clover	1.3	0.0
F. culmorum		
Alfalfa	2.4	41.1
Alsike clover	2.0	31.1
Ladino clover	2.3	40.0
Red clover	0.4	0.0
F. oxysporum		
Alfalfa	1.8	16.2
Alsike clover	2.1	33.8
Ladino clover	0.7	8.7
Red clover	0.6	0.0
Check		
Alfalfa	0.8	0.0
Alsike clover	0.6	0.0
Ladino clover	0.2	0.0
Red clover	0.2	0.0

The distribution of the disease on alsike clover is very interesting since it was foundonly in an area extending from Levis to Bonaventure and in Chicoutimi. Generally, diseases caused by Fusarium are classed as high soil temperature diseases because the pathogen is most active during the warmest weeks of the growing season. In the regions where the disease was noticed, the temperature is relatively low as compared to that of the Montreal area where the disease was not found. On the other hand, the rainfall is quite high in the Lower St. Lawrence and in the Lake St. John areas where the disease was observed; we would be inclined to think that this factor might have contributed to the development of the disease if we consider the heavy incidence of alfalfa root rot caused by F. avenaceum in 1964 when the soil moisture was very high (1). Since alsike clover is sown in mixture with timothy and red clover, could these two plants weaken alsike clover through competition, rendering it more susceptible to attack by microorganisms? We are inclined to think so since agronomists generally contend that it is difficult to keep alsike clover stands for many years in Quebec.

Fusarium isolates fromalsike clover differed in virulence; Chi (2), using Fusarium isolates from red clover has recorded similar findings. On the average, of all the isolates tested, F. avenaceum was the most pathogenic to alfalfa and alsike clover, and F. &-morum the most pathogenic to ladino clover. Furthermore, the isolates of F. oxysporum, which were the most numerous, were less pathogenic to alsike than the isolates of F. avenaceum and had about the same virulence as F. culmorum. A singular lack of specificity was shownby the isolates of Fusarium obtained from alsike clover since they attack roots of M. sativa, T. repens and T. pratense. However, the present results indicate that the use of red clover in mixture with alsike clover may have certain limitations although the strains of Fusarium attacking alsike clover do not appear very virulent to red clover in the present studies. These results open up a field for further investigations of the competition of the two clover species with the emphasis on red clover weakening alsike clover.

The discovery of Fusarium spp. onalsike clover in eastern Quebec may mean that the disease has been missed in earlier surveys since it has not been reported previously. However, if that plant is to be kept in our mixtures in the future, it is urgent to look for varieties resistant to this disease. Furthernore, if alsike clover proves to be unsuitable to our regions due to climatic factors governing the occurrence of this disease, is it wise to retain it any longer in our forage crop?

Literature cited

1. Aubé, C. 1965. Studies on the biology of Verticillium and Fusarium species and effects of environment on development of alfalfa wilt and root rot. XIII. Fusarium rot of alfalfa. Ph. D. Thesis, McGill University, Montreal, Quebec.
2. Chi, C. C. 1965. Pathogenicity of Fusarium species from red clover. Can. Plant Dis. Surv. 45: 3-7.
3. Gordon, W. L. 1952. The occurrence of Fusarium species in Canada. 11. Prevalence and taxonomy of Fusarium species in cereal seed. Can. J. Botany 30: 209-251.