Introduction

Soilborne plant pathogens are regarded as important causes of failures of newly established and mature stands of forage legumes in the North Central Region of the U.S. A Mycoleptodiscus-like fungus was recovered from decaying roots and stems of birdsfoot trefoil plants sampled from two-year-old yield trials in 1994 and establishment year stands in 1995 at the Arlington Agricultural Research Station, Arlington, WI. Although recognized in states south of Wisconsin, Mycoleptodiscus terrestris has not been implicated in poor health of forage legumes in Wisconsin. M. terrestris has been previously reported to be pathogenic on alfalfa, red clover and birdsfoot trefoil in Illinois (Gerdemann, 1954). The fungus has been reported to be pathogenic on birdsfoot trefoil in Missouri (Pettit, et al., 1969) and eastern U.S. (Carroll and Whittington, 1991). Only the trefoil cultivar Dawn (Beuselinck, 1994) and the germplasm CAD (Beuselinck and Steiner, 1994) have been reported to have some degree of resistance (tolerance) to M. terrestris. However, no resistance has been identified in trefoil germplasm adapted to the northern area of the Midwest. Forage legume germplasm has not been characterized extensively for reaction to M. terrestris. Our goal is to characterize the isolates for phenotypic and genotypic traits in relation to isolates from other geographic regions and host origins. This information will be used in a program to improve forage legumes for resistance to M. terrestris. Forage legume germplasm improved for resistance to M. terrestris can be used to assess the importance of this fungal pathogen in the upper Midwest.

Materials and Methods

Pathogen identification. A fungus resembling Mycoleptodiscus terrestris was recovered from root and stem tissue excised from birdsfoot trefoil plants. Excised tissue was placed on acidified potato dextrose agar (APDA) and incubated for 5-7 days. Isolates were grown on APDA, corn meal agar (CMA), microwaved soybean leaflets and irradiated carnation leaves for 7 days under fluorescent lights (12 hr light: 12 hr dark).

Isolates and germplasm evaluation. The virulence of eight isolates, four derived from field grown birdsfoot trefoil plants; one each from greenhouse grown red clover and alfalfa plants; and three from alfalfa-derived isolates provided by L.H. Rhodes, Ohio State University, Columbus, OH, was evaluated on eight forage legumes: alsike, berseem, kura, red and white clovers, alfalfa, birdsfoot trefoil and sweetclover. Three-week-old seedlings were inoculated with a mycelium/sclerotium suspension (one 100 mm standard plate/1 water) as a drench at a rate of 20 ml per 10 seedlings and incubated for three weeks at 25°C. Six-week-old seedlings were evaluated for reaction to the respective isolates on a scale of 1 to 5; 1 = no symptoms and 5 = a dead plant.

Results and Conclusions

Pathogen identification. All isolates recovered from birdsfoot trefoil produced sclerotia and conidia that were morphologically similar to Mycoleptodiscus terrestris isolates recovered from alfalfa. All birdsfoot trefoil isolates and two of the three alfalfa-derived isolates produced
conidia on carnation leaves but did not sporulate on PDA. One isolate from birdsfoot trefoil was tested on CMA but did not sporulate. Light was required for sporulation. All isolates produced sclerotia on carnation leaves, PDA and CMA.

**Isolate-species evaluation.** All forage species were susceptible to the *M. terrestris* isolates used in this study (Table 1), but alsike and kura clovers and alfalfa were less susceptible to this set of isolates compared to the other forage legumes evaluated. Symptoms varied from black decay of root tissues to severe wilting of the stem tissue resulting from root decay or black water-soaked lesions at the base of stems which completely girdled the stem.

**Future Research/Plans**

1. Isolates of *Mycoleptodiscus* recovered from forage legumes in Wisconsin will be compared to isolates from other regions of the United States and other species of *Mycoleptodiscus*. Isolates will be compared for differences in phenotypic traits such as spore and sclerotium morphology and virulence to specific forage legumes and compared to genotypic traits related to molecular markers.

2. A standardized method will be developed to select and characterize forage legume germplasm for resistance to *M. terrestris*.

3. An attempt will be made to improve forage legume germplasm for resistance to *M. terrestris*. This germplasm will be used to determine the role of *M. terrestris* on forage legume performance in the upper Midwest.

**References**


<table>
<thead>
<tr>
<th>SPECIES*</th>
<th>Tre 94.1</th>
<th>Tre 95.1</th>
<th>Tre 95.2</th>
<th>Tre 95.3</th>
<th>Red 94.1</th>
<th>Alf 94.1</th>
<th>Rodalf 1</th>
<th>Rodalf 2</th>
<th>Rodalf 3</th>
<th>MEAN</th>
<th>LSD (5%)</th>
<th>CV (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alsike Clover</td>
<td>3.50</td>
<td>2.54</td>
<td>2.57</td>
<td>2.02</td>
<td>2.57</td>
<td>2.66</td>
<td>2.95</td>
<td>3.14</td>
<td>3.01</td>
<td>2.77</td>
<td>0.51</td>
<td>15.30</td>
</tr>
<tr>
<td>Alfalfa</td>
<td>2.86</td>
<td>2.92</td>
<td>2.97</td>
<td>2.28</td>
<td>2.57</td>
<td>3.11</td>
<td>2.92</td>
<td>2.65</td>
<td>2.74</td>
<td>0.44</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Berseem Clover</td>
<td>4.04</td>
<td>3.19</td>
<td>3.38</td>
<td>2.50</td>
<td>3.43</td>
<td>3.37</td>
<td>3.50</td>
<td>3.81</td>
<td>3.61</td>
<td>3.42</td>
<td>0.42</td>
<td></td>
</tr>
<tr>
<td>Kura Clover</td>
<td>3.44</td>
<td>2.67</td>
<td>2.91</td>
<td>2.47</td>
<td>2.91</td>
<td>2.89</td>
<td>3.32</td>
<td>3.00</td>
<td>3.17</td>
<td>2.97</td>
<td>0.40</td>
<td></td>
</tr>
<tr>
<td>Red Clover</td>
<td>3.95</td>
<td>3.09</td>
<td>3.20</td>
<td>2.72</td>
<td>3.39</td>
<td>2.88</td>
<td>3.83</td>
<td>3.88</td>
<td>2.96</td>
<td>3.32</td>
<td>0.33</td>
<td></td>
</tr>
<tr>
<td>Sweetclover</td>
<td>3.08</td>
<td>3.75</td>
<td>3.40</td>
<td>2.62</td>
<td>3.79</td>
<td>3.50</td>
<td>3.50</td>
<td>3.68</td>
<td>3.21</td>
<td>3.40</td>
<td>0.67</td>
<td></td>
</tr>
<tr>
<td>Trefoil</td>
<td>3.73</td>
<td>3.37</td>
<td>3.10</td>
<td>2.55</td>
<td>3.14</td>
<td>2.96</td>
<td>3.63</td>
<td>3.62</td>
<td>3.42</td>
<td>3.27</td>
<td>0.65</td>
<td></td>
</tr>
<tr>
<td>White Clover</td>
<td>3.67</td>
<td>3.19</td>
<td>3.30</td>
<td>2.36</td>
<td>3.25</td>
<td>2.79</td>
<td>3.58</td>
<td>3.24</td>
<td>3.01</td>
<td>3.16</td>
<td>0.59</td>
<td></td>
</tr>
<tr>
<td>MEAN</td>
<td>3.53</td>
<td>3.09</td>
<td>3.10</td>
<td>2.43</td>
<td>3.11</td>
<td>2.95</td>
<td>3.41</td>
<td>3.41</td>
<td>3.13</td>
<td>3.13</td>
<td></td>
<td></td>
</tr>
<tr>
<td>LSD (5%)</td>
<td>0.76</td>
<td>0.58</td>
<td>0.24</td>
<td>0.39</td>
<td>0.42</td>
<td>0.42</td>
<td>0.4</td>
<td>0.45</td>
<td>0.48</td>
<td>0.22</td>
<td>0.20</td>
<td></td>
</tr>
<tr>
<td>CV (%)</td>
<td>15.30</td>
<td>13.30</td>
<td>4.00</td>
<td>3.30</td>
<td>10.80</td>
<td>10.10</td>
<td>9.80</td>
<td>9.30</td>
<td>11.00</td>
<td>11.20</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Disease Severity Index:** 1 = healthy plant, 5 = dead plant.

**ISOLATES:**
- Tre 94.1 = Original trefoil isolate from 1994 field nursery
- Tre 95.1 = Isolate obtained from field in Sept 95
- Tre 95.2 = Isolate obtained from field in Sept 95
- Tre 95.3 = Isolate obtained from field in Sept 95
- Red 94.1 = Isolate from red clover inoculated with Tre 94.1
- Alf 94.1 = Isolated from alfalfa inoculated with Tre 94.1
- Rodalf 1 = Alfalfa isolate #1 from Rhodes
- Rodalf 2 = Alfalfa isolate #2 from Rhodes
- Rodalf 3 = Alfalfa isolate #3 from Rhodes