POTENTIAL OF *PYRENOPHORA AVENAE* FOR BIOLOGICAL CONTROL OF WILD OATS, *AVENA FATUA*

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Summary. This study examines the potential of *Pyrenophora avenae* for the control of wild oats, *Avena fatua* L., by investigating its host range, growth requirements and host-plant interactions. Despite devastating effects of primary infection arising from seed or soil inoculation, it is concluded that inundation of seedlings with *P. avenae* shows most promise.

INTRODUCTION

The control of annual weeds using endemic fungal pathogens, the mycoherbicide strategy, has been extensively reviewed (2, 8, 11, 16). The mycoherbicide approach may have the potential to control wild oats, which is an important annual grass weed in cereal crops worldwide. Cereal yield losses are estimated at 2.2 million tonnes in Europe and 6.4 million tonnes in North America (6). The object of this study, therefore, was to investigate the feasibility of the endemic fungal pathogen *Pyrenophora avenae* Ito and Kurib. to control wild oats. Observations have shown this fungal disease to have a devastating effect on the spring emergence and survival of wild oat seedlings (N.C.B. Peters, pers. comm., 1984).

Pyrenophora avenae is a seed-borne fungus found as dormant mycelium between the pericarp and glumes of the caryopsis. When the seed germinates the fungus will begin to grow and colonize the coleoptile. Symptoms of primary infection include a twisted and distorted coleoptile and brown stripe lesions on the unfolding leaves. Primary infection can cause pre or post-emergence death of the wild oat seedling. If the seedling survives it may be stunted, produce fewer tillers, and in growing away from primary infection only the older leaves will display stripe symptoms. Later in the growing season, July-August, these lower leaves produce cylindrical, multi-septate conidia (30-170 x 10-20 um) that can cause secondary infection on the upper leaves seen as red-brown spot lesions. Inflorescences may also become infected and therefore allow *P. avenae* to continue its life cycle (3, 12).

To be a potential biological control agent, *P. avenae* must satisfy the following requirements: (a) produce abundant and durable inoculum in artificial culture; (b) be specific to the target weed; and (c) control the weed over a wide range of environmental conditions (2). Therefore this study investigated: (a) the growth requirements of *P. avenae*; (b) host range of *P. avenae*; and (c) the effect of *P. avenae* on the growth and development of wild oats.

METHODS

1. Growth requirements.

Media. Different nutrients were used to investigate their effect on the growth and sporulation of *P. avenae*. Cultures of *P. avenae* isolated from wild oat leaf tissue were subcultured onto potato carrot (PCA), V-8 juice, half concentration V-8 juice, oatmeal (OMA), half concentration OMA, green bean, wild oat leaf extract, or Sachs agar. All cultures were grown at 16°C, under 16 h near ultra violet light/8 h dark for 3 weeks.

Temperature range. Agar plugs of *P. avenae* were placed on PCA and grown at temperatures ranging from 4–32°C at 4°C increments; no light was provided. A spore suspension of *P. avenae* was applied to PCA and placed under the same temperature regime to determine the effect of temperature on the germination of spores after 24 h incubation.

2. Host range.

The plant species tested were wild oats, *A. sterilis* ssp. *ludoviciana*, *A. sativa*, *Triticum aestivum* and *Hordeum vulgare*. A spore suspension of *P. avenae* (5.5×10^4 spores/ml) in 0.1% Agral^R solution was sprayed on test plants to run off. Growth stage of plants was 12 on the Zadoks scale. A humid environment was provided for 48 h at 20°C.

3. Effect of *P. avenae* on wild oats.

Seed inoculation. Two pot experiments were conducted, one in a glasshouse where the average temperature was above 15°C and the other outside where it was below 15°C. Wild oat seed prior to sowing were placed in *P. avenae* mycelium homogenate prepared from a blended PCA culture. Plant material was harvested after 130 days and oven dry weights of shoots and roots were noted. The seed was harvested and weight determined from the number of seeds in a 2 g sample.

Soil inoculation. In a pot experiment, 100 seeds were sown per pot in non-sterilized soil either inoculated with homogenized OMA on which *P. avenae* had grown (10% w/w homogenate/soil), or homogenized OMA alone. The effect of inoculum level on emergence was investigated using 0, 20 and 50% homogenate/soil (w/w).

RESULTS AND DISCUSSION

The study has shown that *P. avenae* can grow well on PCA, V-8, half concentration V-8, OMA, green bean and wild oat leaf extract agar. Maximum spore production was achieved on green bean agar (Table 1).

Table 1. The effect of different media on *P. avenae* spore production (spores/plate)

Media	Spore production
Green bean	2.47×10^5
Oatmeal	2.19×10^5
V-8	1.94×10^5
Potato carrot	1.21×10^5
50% V-8	1.04×10^5
Wild oat leaf extract	9.6×10^4

Alternative spore production methods should be investigated. For instance, *Alternaria macrospora* can be grown in liquid culture, blended, poured into pans and incubated to produce spores (14). Aqueous mixtures of 1% (w/v) sodium alginate and homogenized mycelium of *Alternaria* spp. and *Fusarium* spp. can be pelletised by dropwise additions to 0.25 M $CaCl_2$ and air-dried. The pelletised fungi sporulate readily under a specific light-dark regime (15). These techniques should show potential for *P. avenae* spore production.

The optimum temperature for growth on PCA ranged from 12 - 24°C, with no measurable growth at 4°C. Spore germination (normally 80-90%) was reduced to below 50% by a temperature of 4°C. *P. avenae* can grow well at temperatures that usually occur in late spring-summer in Great Britain.

P. avenae was shown to produce symptoms on wild oats, *A. sterilis* and *A. sativa*, and could be re-isolated from the leaf tissue of these plant species. Small necrotic spots were observed on *T. aestivum* and *H. vulgare* but *P. avenae* could not be re-isolated from the inoculated plants. These symptoms are probably the result of a hypersensitive response. *P. avenae* could therefore be used as a control agent of wild oats when the weed is growing in wheat and barley but not cultivated oats.

At temperatures below 15°C primary infection reduced the emergence of wild oats by 50% and subsequently killed 75% of those seedlings that emerged (Table 2). There was also a 60% reduction in shoot and root dry weight of seedlings that survived primary infection. The seed of infected wild oats is significantly smaller and this may reduce the competitiveness of these seedlings when emerging in a crop (9).

Table 2. The effect of primary infection on the emergence and mortality of wild oats.

Infection	Temperature	Emergence	Mortality
Healthy	>15	81	-
	<15	65	-
Infected	>15	89	7
	<15	30	75
(s.e)		6.1	0.08

Primary infection had little or no effect on the emergence and mortality of seedlings at temperatures above 15°C although both tiller number and shoot dry weight were increased, 25 and 17%, respectively, and root dry weight reduced by 15%. These observations suggest wild oats is producing healthy tillers in response to *P. avenae* infection. The reduction in root dry weight noted in both experiments agrees with observations by others (4) on *A. sativa* treated with *P. avenae* culture filtrate.

Soil inoculation can introduce primary infection and result in mortality of germinating wild oat seed (Table 3).

Table 3. The effect of inoculation of the soil in April with *P. avenae* on wild oats.

Soil treatment	Emergence (%)	Primary infection (%)	Mortality (%)
Control	16	8	2
Inoculated	12	98	30

Soil inoculation has several disadvantages as a potential method for controlling wild oats.

1. It assumes all germinating seed will come into contact with *P. avenae* and become infected. Emergence was shown to decline from 70% in the control soil to 30% where inoculum level was 50% (w/w), thus the higher the soil inoculum level of *P. avenae* the greater the probability of pathogen and seed contact and lower wild oat emergence.

2. It assumes *P. avenae* can survive in a soil for the period that wild oat seed are able to germinate. In fact viability of *P. avenae* in this soil dropped rapidly after 1 month to give only 10% primary infection and no mortality. The reason for the decline in viability is most probably the presence of antagonists and hyperparasites in the soil. Related fungi e.g, *Helminthosporium* spp. are known to be hyperparasitised by *Hymenula cerealis* (5), amoebae (7), and there are various antagonists, soil bacteria (10) and other fungi (13, 1).

The answer to control of wild oats may lie in the application of *P. avenae* as inundative inoculum to the entire target weed population. This study has not been able to achieve the spore concentration required for inundation. Further work must be undertaken on spore production, application methods, e.g., humectants, stickers and wetters, and perhaps the use of plant growth regulators or low doses of herbicide to enhance the disease effect.

REFERENCES

1. Campbell, W.P. 1956. Can. J. Bot. 34, 865-874.
2. Daniel, J.T., Templeton, G.E., Smith, R.J. and Fox, W.T. 1973. Weed Sci. 21, 303-307.
3. Dennis, R.W.G. 1973. Trans. Brit. Mycol. Soc. 18, 223-238.
4. Graniti, A. and Porto Puglia, A. 1984. Phytopath, Medit, 33, 39-42.
5. Kenneth, R. and Isaac, P.K. 1964. Can. J. Plant Sci. 44, 182-187.
6. O'Donovan, J.T. and Sharma, M.P. 1984. Can. Plains Proc. 12, 27-42.
7. Old, K.H. 1978. Ann. Appl. Biol. 89, 128-131.
8. Scheepens, P.C., Van Zon, J.C.J. 1982. In: Microbial Pesticides (Ed. E. Kurstak). (Marcel-Dekker: New York). pp. 623-664.
9. Sharma, M.P., McBeath, D.K. and Vanden Born, N.H. 1977. Can. J. Plant. Sci. 57, 811-817.
10. Simmonds, P.M. 1947. Sci. Agric. 27, 625-632.
11. Templeton, G.E., Smith, R.J., Tebeest, D.O., Beasley, J.N. and Klerk, R.A. 1981. Arkansas Farm Res. 30, 8.
12. Turner, D.M. and Millard, W.A. 1931. Ann. Appl. Biol. 28, 535-558.
13. Tveit, M. and Moore, M.B. 1954. Phytopath. 44, 686-689.
14. Walker, H.L. 1980. U.S. Dept. Agric. Science and Education Administration, Advances in Agricultural Technology, Southern Series, No. 12. 5 pp.
15. Walker, H.L. and Connick, W.J. 1983. Weed Sci. 31, 333-338.
16. Woodhead, S.W., Ridings, W.M. and Schoulties, C.L. 1981. Phytopath. 71, 913.