THE EFFECT OF THE ENDOPHYTIC FUNGUS *ACREMONIUM LOLII* ON ADULT BLACK BEETLE (*HETERONYCHUS ARATOR*) FEEDING

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SUMMARY

Choice and no choice feeding tests using *Acremonium lolii*-infected and *A. lolii*-free perennial ryegrass plants were performed on adult black beetle. Tiller counts showed that significantly more feeding damage occurred on *A. lolii*-free plants. The presence of *A. lolii* in perennial ryegrass therefore deters feeding by the adult black beetle.

Keywords: endophyte, black beetle, ryegrass, biocontrol.

INTRODUCTION

Considerable interest in the role of endophytic fungi as insect biocontrol agents was generated following the discovery that the presence of *Acremonium lolii* Latch, Christensen and Samuels in perennial ryegrass (*Lolium perenne* L.) conferred resistance to the grass from attack by Argentine stem weevil (*Listronotus bonariensis* (Kuschel)) (Prestidge *et al* 1982). In New Zealand the presence of *A. lolii* in perennial ryegrass has been shown to have similar effects on the cutworm, *Graphania mutans* (Dymock *et al* 1988), and the pasture mealy bug, *Balanococcus poae* (Pearson 1988).

The effect of *A. lolii* infection on the black beetle, *Heteronychus arator* (F.) (Coleoptera:Scarabaeidae), an introduced pest of northern North Island pastures, has not been reported. The present study was therefore undertaken to determine whether *A. lolii* has potential as a biocontrol agent against black beetle attack on perennial ryegrass.

METHODS

Choice and no choice feeding tests with respect to *A. lolii* infection in perennial ryegrass were performed on adult black beetle. Mature perennial ryegrass plants (c.v. Ellett) were used in both tests and the endophyte status of each plant was assessed. Sheath material was stained in aniline blue plus lactophenol and the presence or absence of *A. lolii* hyphae determined microscopically.

Black beetle were collected using pitfall traps, between April and June 1991 from pastures at the Ruakura Agricultural Centre. During the winter, the black beetles were maintained on a carrot diet in liverpails of soil, prior to experimentation in the spring.

The choice and no choice feeding tests were carried out in a greenhouse (12°C-28°C) and were terminated when damage became too excessive for black beetle to continue feeding.

Choice test

In June 1991, ten liverpails (170 mm x 170 mm x 190 mm) were filled to within 50 mm of the top with sieved Horotiu sandy loam soil. One *A. lolii*-infected and one *A. lolii*-free perennial ryegrass plant, each with between three to ten tillers, were planted into each pail. In September 1991, three male black beetles were introduced into half of the liverpails, and three females were introduced into each of the remainder. Each liverpail was covered with nylon mesh supported by a wire frame to prevent the black beetle from escaping, while allowing plant growth. The number of healthy and black beetle-killed tillers on each plant was assessed every 3 to 7 days, until the conclusion of the trial (day 24). Tillers which were shredded at their base resulting in their collapse, were classed as black beetle-killed. All other tillers were classed as healthy.

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No choice test

In June 1991, 16 liverpails were filled with sieved Horotiu sandy loam as in the choice experiment. Half of the liverpails were planted with two *A. lolii*-infected perennial ryegrass plants, and the remainder with two *A. lolii*-free perennial ryegrass plants. In September 1991, two male and two female black beetle were introduced into each liverpail, which were then covered with mesh as before. Black beetle induced tiller mortality was recorded periodically on each plant. Black beetle mortality was assessed at the conclusion of the trial (day 27).

Data were analysed using a chi-squared test.

RESULTS

Choice test

From day 11 onwards, significantly ($P < 0.001$) more *A. lolii*-free tillers were killed by black beetle than *A. lolii*-infected tillers (Fig. 1). By day 22, 74.5% of the *A. lolii*-free tillers had been killed, compared to 10.5% in the *A. lolii*-infected plants. The effect of beetle sex on tiller mortality was not significantly different.

No choice test

Black beetle killed significantly ($P < 0.001$) more *A. lolii*-free tillers than *A. lolii*-infected tillers (Fig. 2). Tiller mortality on *A. lolii*-free plants increased steadily from day 8 and by day 27, 89.3% had been killed. Tiller mortality on *A. lolii*-infected plants was very low and by day 27, only 1.6% of the tillers were dead. There was no significant effect of *A. lolii* on black beetle mortality.

Fig. 1: Choice Test: Mortality of *A. lolii*-free (E-) and *A. lolii*-infected (E+) Ellett perennial ryegrass due to adult black beetle. (Bars represent S.E.M.)

DISCUSSION

In order to reduce pesticide use, insect pest control using non chemical factors is becoming an important focus of research. The current study showed that the presence of *A. lolii* significantly reduced perennial ryegrass tiller death due to feeding by adult black beetle. Black beetle adults feed at the tiller bases just above the crown of the plant where *A. lolii* and several of the toxins associated with its presence, are known to be concentrated (Gallagher *et al* 1987), indicating a possible reason as to why the effect of *A. lolii* was so marked. The black beetle thus joins the list of insects affected

Fig. 2: No Choice Test: Mortality of *A. lolii*-free (E-) and *A. lolii*-infected (E+) Ellett perennial ryegrass due to adult black beetle. Bars represent S.E.M.).

by the presence of *A. lolii* in perennial ryegrass and is the first member of the Scarabaeidae to be recorded as such. However, mortality was not significantly higher in the black beetle confined to the *A. lolii*-infected plants, even though feeding activity was considerably reduced. This may have been because the no choice test was terminated before the effects of a depleted diet manifested itself as an increase in black beetle mortality. Similarly reduced feeding of black beetle maintained on *A. lolii*-infected ryegrass may be expected to lead to a reduction in fecundity.

The alkaloids peramine, ergovaline, ergotamine, ergocryptine and lolitrem B, are usually present in *A. lolii*-infected perennial ryegrass. All have been shown to have feeding deterrent effects or to be toxic to larval and/or adult Argentine stem weevil (Gaynor and Rowan 1985; Prestidge and Gallagher 1985; Dymock *et al* 1989; Popay *et al* 1990). *A. lolii*-infected perennial ryegrass may also contain many other potentially toxic/deterrent compounds (Miles *et al* 1992). Studies are underway to determine the effects of these on adult black beetle.

It is not known whether black beetle larvae, which often feed on perennial ryegrass roots, are also affected by the presence of *A. lolii*, although they can discriminate between roots from different plant species and show a marked preference for perennial ryegrass and paspalum (*Paspalum dilatatum*) roots (King *et al* 1981).

Although serious outbreaks occur only sporadically, black beetle is present in many free-draining soils at low densities and contributes to ryegrass loss and poor pasture vigour. Therefore, New Zealand ryegrass breeding programmes involving the development, or selection, of new strains of *A. lolii* should take the effects of *A. lolii* on black beetle, as well as the Argentine stem weevil, into account.

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