IMPORTATION AND QUARANTINE OF *MICROCTONUS HYPERODAE*, A SOUTH AMERICAN PARASITOID OF ARGENTINE STEM WEEVIL

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SUMMARY

The potential for biological control of Argentine stem weevil (*Listronotus bonariensis*) using the thelytokous parasitoid *Microctonus hyperodae* is discussed in the context of current efforts to develop resistant grasses. The collection in South America of 13447 weevils and their shipment to New Zealand is described. These in turn produced 513 *M. hyperodae* pupae which led to the establishment of 247 parthenogenetic lines. Although generally parthenogenetic, four males were also produced. Quarantine and rearing procedures employed to date are briefly outlined.

INTRODUCTION

The damage potential of the ubiquitous Argentine stem weevil (*Listronotus bonariensis*) to New Zealand pastoral and arable agriculture is well known (e.g. Kelsey 1958). This pest is estimated to cost the nation more than $150M every year in both yield losses and cost of pasture renovation (Prestidge *et al* 1985).

Over the last 35 years, considerable investigation into the use of insecticide to control *L. bonariensis* has been carried out (e.g. Pottinger 1983) and there are indications that in some specialist applications like seed crops and pasture establishment (Trought 1976), this may be a cost-effective approach. However, the mining habit of the larval stages and the frequently unpredictable flight activity in some areas of the country (Goldson 1981) mitigates against widespread insecticide use. In view of these considerations and the need to develop low input, sustainable agricultural systems, effort has been directed towards the development of grasses resistant to *L. bonariensis.*

The discovery that the fungal endophyte *Acremonium lolii*, when present in ryegrass plants, reduces *L. bonariensis* damage (Prestidge *et al* 1982) has greatly improved the understanding of grass resistance mechanisms, and has allowed exploitation of this resistance to the pest (e.g. Prestidge *et al* 1985). Unfortunately, *A. lolii* also causes ryegrass staggers (Fletcher and Harvey 1981) and other effects deleterious to stock development (Fletcher 1986). Analysis of endophyte-infected plants has shown lolitrem B to be responsible for the animal health problems (Gallagher *et al* 1981) while a different compound, peramine, confers most of the plant's resistance to *L. bonariensis* (Gaynor and Rowan 1985). Since this original work, considerable effort has been devoted to discovering and developing endophytes which may confer resistance to *L. bonariensis* without incurring stock health problems (R.A. Prestidge, pers. comm.).

Biological control agents would be highly complementary to plant resistance. In 1966-67 the *L. bonariensis* egg parasitoid *Anaphes atomarius* was introduced and released in New Zealand by DSIR but apparently failed to establish (Dymock 1989). A more recent initiative involves a partnership between MAF Technology and DSIR Plant Protection. This work is focused on the South American adult parasitoid *Microctonus hyperodae*. Apart from a single descriptive paper by Loan and Lloyd (1974) nothing was known about this species although considerable experience has been acquired by the

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MAF/DSIR group working with the *Sitona discoideus* parasitoid *Microctonus aethiopoides* (Goldson *et al* 1990). Life table and modelling analysis of the impact of *M. aethiopoides* on *S. discoideus* has clearly demonstrated the value of parasitoids that attack the adult stage of a pest species rather than its early stages (Barlow and Goldson in prep.).

METHODS

Exploratory collection of biological material

The availability of *M. hyperodae* in South Brazil (Rio Grande do Sul) was originally drawn to the attention of the authors by an EMBRAPA entomologist at Passo Fundo, Mr Dirceu Gassen. Consequently an initial visit was made to South Brazil between mid-November 1988 and mid-January 1989 by S.L. Goldson. Sampling was also carried out in various locations in Argentina and Chile (Fig. 1). The best sites for weevil and parasitoid collection were invariably low-lying damp or ornamental areas (town plazas etc) which contained a high proportion of volunteer *Lolium multiflorum*. The presence of the parasitoids and their intensity of attack were determined at each location by dissection of *L. bonariensis* adults. During these visits it was also arranged for local technical assistants to forward regularly to New Zealand samples of *L. bonariensis* adults preserved in alcohol. These were subsequently dissected to ascertain levels of parasitism in an attempt to determine an optimum time for future collection and live transhipment.

Collection and transhipment of live material

A return visit to South America was made by S.L. Goldson and M.W. Stufkens in September-October 1989 with the aim of collecting parasitised populations of weevils for live shipment back to New Zealand. After ascertaining parasitism by dissection,

Fig. 1: ***Microctonus hyperodae*** **collection sites.**

populations of adult *L. bonariensis* were sent from the areas around Porto Alegre (South Brazil), Ascasubi, General Roca and Bariloche (Argentina) and La Serena (Chile) (Fig. 1). These were supplemented by follow-up collections in November-January (1990) by R.P. Pottinger who also made new collections at Colonia (Uruguay) and in Concepcion (South Chile) (Fig. 1).

Each time an adequate population was acquired, the material was immediately carried (by air where possible) to Buenos Aires for on-shipment to New Zealand by Aerolineas Argentinas. On arrival in New Zealand the insects were quickly moved to the DSIR quarantine facility at Lincoln. Arrival in quarantine was usually within 20 hours of dispatch from Buenos Aires.

Specimens were collected from a wide range of habitats within latitudes similar to New Zealand in order to provide as much biotypical material as possible. Habitats varied from subtropical humid (Porto Alegre) to arid subalpine (Bariloche) to dryland (General Roca) (Fig. 1).

Quarantine and rearing procedures

Given the risk of foot and mouth disease and anthrax, imported material was immediately placed under maximum security quarantine conditions. Temperatures were maintained at around 17 °C with a 16:8 L:D photoperiod and 80-90% humidity. Careful attention was paid to ensure that no hyperparasites were accidentally reared, in particular *Mesochorus* spp. (Loan pers. comm.).

Since no laboratory research had been undertaken with *M. hyperodae* before, approaches to its handling were of necessity experimental. However, using the experience gained with *M. aethiopoides* (Goldson *et al* 1990), slightly modified techniques were applied to *M. hyperodae* and were found to be largely satisfactory. In each case fifty or more *L. bonariensis* adults were exposed to two parasitoids for 48 hours in a 110 cm circular cage with no furnishings. Thereafter the parasitoids were removed and the weevils transferred to a similar cage with a mesh floor; this allowed emergent prepupae of the parasitoid to drop into a container below and pupate on strips of paper towel. While the parasitoids developed, the weevils were fed sprigs of low endophyte ryegrass twice weekly. The emergent pupae were periodically collected and placed in petri dishes with a damp wick until adult eclosion.

M. hyperodae is parthenogenetic and each female adult that originated from the material collected in South America was used to found a clonal line. Such an approach ensured a reliable means by which to monitor and try to remedy any decline in genetic variability resulting from the quarantine process. In this way over 240 lines were established.

RESULTS AND DISCUSSION

All the weevils imported from all locations were identified as *L. bonariensis* by R. Craw (pers. comm.). While the weevil was relatively easily found in association with *Lolium* spp. in damp/low-lying areas, primitively it would seem that the distribution of *L. bonariensis* followed that of 'vega'-type areas (Lloyd 1966). These are characteristically stream-fed swampy areas and are equivalent to cushionbogs and flush vegetation swamps in New Zealand (S. Halloy, pers. comm.). The primitive floristic assemblage in these areas is dominated by Juncaceae, Cyperaceae and some Graminae such as *Poa* spp. and *Deyeuxia* spp. While *Poa* is a well known host, it is possible that the weevil can survive on young *Deyeuxia* spp. as well.

The details of the shipments from South America are summarised in Table 1. A total of 13447 weevils were collected, which produced 513 pupae leading to the establishment of 247 adult *M. hyperodae*. In addition four apparently impotent males were reared from the imported weevils. Although Loan and Lloyd (1974) have reported the species to be parthenogenetic, such very rare occurrence of males is consistent with species that show obligate thelytoky (Imms 1964).

Levels of parasitism of less than 5%, as determined by emergence, were remarkably low compared to those sometimes measured directly in South America by dissection. In August 1988, 61% parasitism was found in a population from Passo Fundo in South Brazil and 77% in La Serena in early January 1989 (Goldson

TABLE 1: Details of parasitoid shipments from South America.

Location collected	No. of weevils imported	Pupal yield	Parasitoid lines established	Percent parasitism
Brazil				
Porto Alegre (c.30°S)	960	72[1]	48	4.5
Uruguay				
Colonia (c.35°S)	2183	115	48	5.3
Argentina				
Ascasubi (39°S)	1200	65	31	5.4
General Roca (39°S)	1729	79	40	4.6
Bariloche	2625	27	19	0.7
Chile				
Concepcion (c.36°C)	1550	17	12	2.0
La Serena (c.30°C)	2500	138	49	5.4
Totals	13447	513	247	

[1] 29 pupae were forwarded directly

unpublished data). Similarly Loan and Lloyd (1974) observed the levels of parasitism in the Bariloche area to be 39% in October 1972. Dissection during the second visit again indicated that the levels of parasitism were consistently higher than the subsequent emergence data suggested (Goldson unpublished data). It is probable, therefore, that the relatively low yields in quarantine related to the rigours of transhipment.

Host range specificity testing is currently being conducted on native Curculionidae in quarantine prior to the preparation of an environmental impact assessment. Should *M. hyperodae* be found to be acceptable for release, there is a reasonable probability that it would be able to establish in at least some areas of New Zealand given the wide range of habitats in South America where it is found. These areas of establishment should include dry habitats as it was more readily collected in these environments (Goldson unpublished data). It is noteworthy that it is the dry eastern areas of New Zealand that sustain the most damage by *L. bonariensis* (Prestidge *et al* 1985).

While it is doubtful that *M. hyperodae* would singularly eliminate *L. bonariensis* as a problem, it is highly probable that it would provide a very valuable adjunct to plant resistance. This is particularly so when it is considered that, in ameliorating damage, the suppressive effect of integrated multiple factors may be more synergistic than simply additive in reducing pest damage (van Emden 1982).

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