OBSERVATIONS ON THE BIOLOGY OF THE PASTURE MEALYBUG, *BALANOCOCCUS POAE*, FROM HAWKE'S BAY PASTURES

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ABSTRACT

The seasonal biology of *Balanococcus poae* was examined in Hawke's Bay from 2006 to 2008. Adult females were found throughout the year, typically in wax cells ca 1–2 cm below the soil surface, with a peak density of ca 1300/m² during winter and early spring (June–October). Winter eggs were followed by neonate nymphs from spring through summer. The timing of life-stages indicates that there was a single generation each year, but a partial second generation may also have occurred in late summer. No males were found. Morphometric data did not reliably separate different immature stages. Neonate nymphs on potted plants crawled first to the tips of tillers and then moved down the blade to feed beneath the protective tiller sheath. No parasitoids or predators of mealybugs were found. In the absence of biological control, grasses with endophytic fungi that confer resistance to pasture mealybug would give effective control.

Keywords: seasonal phenology, *Ripersella rumicis*, behaviour, natural enemies, morphometric data, control.

INTRODUCTION

The mealybug *Balanococcus poae* (Maskell) (Hemiptera: Pseudococcidae) is endemic to New Zealand, and one of 23 species of the genus so far known in the country (Cox 1987). It was rather superficially described by Maskell (1879) as *Dactylopius poae* and by Brittin (1915) as *Ripersia globatus*. Maskell's *D. poae* was redescribed by De Boer (1968) as *Pseudantonina poae*, and it was given its current designation (as a new combination) by Cox (1987). It is naturally found on many native grasses and tussocks in the Poaceae, but has adapted well to a number of introduced pastoral grass species, particularly perennial ryegrass, *Lolium perenne* L. For this reason it is now commonly known as the pasture mealybug.

The biology and life-history of *B. poae* have not been well studied. Maskell (1879) found it on the roots of the common tussock grass, "or rather on the stems close to the ground". Most of the adult females identified have been from roots, or "roots and bases of host plants" (Cox 1987), and Pearson (1988) noted that the insect attacked the crowns, upper roots and lower tillers of ryegrass, and that crawlers climbed to the tips of tillers to disperse passively by wind.

Pearson (1988) described the insect's life-cycle as "straightforward and parthenogenetic", with adults and eggs occurring within ovisacs on the crown, upper roots and lower tillers of plants from late-September through to October. It was estimated

that females produced from 50 to 400 eggs each through one generation a year, but more recent evidence suggests that *B. poae* is bi-sexual and develops through two generations a year, at least in Canterbury (Pennell et al. 2004).

High numbers of *B. poae* were frequently associated with unthrifty or dead grasses during the 1960s to 1980s, particularly during periods of drought (Pearson 1988). Hence most of the field research on *B. poae* has centred on measuring its status as a pasture pest, particularly in Canterbury. Populations on native grasses may be naturally reduced by a native endoparasitoid, discovered by Pearson (1988) and described as *Odiaglyptus bififormis* (Hymenoptera: Encyrtidae) by Noyes (1988). Some control in pasture may be achieved also by the use of strains of *Neotyphodium lolii*, a fungal endophyte of perennial ryegrass (Pearson 1988; Pennell et al. 2004, 2005).

In this study aspects of the development, seasonal biology and phenology of *B. poae* were examined in populations collected from Hawke's Bay.

METHODS

Seasonal phenology, biology and natural enemies of *B. poae*

Mealybugs, identified as *B. poae* by R.C. Henderson (Landcare Research, Tamaki, Auckland), were collected from spade samples of permanent pasture and soil from two Hawke's Bay farms in June 2005. One farm, at Kereru, was under the western foothills (altitude 300 m above sea level), and the other, at Maraetotara, was in high country (altitude 600 m above sea level) close to the eastern coast. A further seven samples were collected from Kereru from December 2005 until February 2007, after which regular sampling was switched to Maraetotara (following a trial sample in December 2006). A further nine samples were then collected regularly (every 4 to 6 weeks in summer and 6 to 12 weeks in winter) from Maraetotara from March 2007 to May 2008.

On each sampling occasion after June 2005, ca 40 soil cores (5 cm diameter) to a depth of ca 10 cm were collected from a known infested area of paddock. The cores were 2 metres apart in four lines of 10 replicates/line. The cores were couriered to the Mt Albert Research Centre, where they were carefully pulled apart and live insects examined under a binocular microscope. The different life-stages of *B. poae* present were noted, as was the depth in the core from which any mealybug was recovered, and any other relevant biological event. Mealybugs were identified to species by eye (with the identity of representative individuals being confirmed by R.C. Henderson). The insects were examined in this manner, rather than by wet extraction, because the search was primarily for the ecological relationships between mealybugs and their host plant and natural enemies. The numbers of adult female *B. poae* in a wax cell in the cores were recorded. The presence of eggs and crawlers (neonate nymphs) within the cells, but not their numbers, was also recorded.

The plant species in each core were not identified, but in a separate study a year previously the 5-year old sown pasture at Maraetotara was dominated by introduced grasses. Species included *L. perenne* (perennial ryegrass) (in 34% of cores), *Agrostis capillaris* (brown top) (37%), *Holcus lanatus* (Yorkshire fog) (15%), *Cynosurus cristatus* (crested dog's-tail) (11%) and *Anthoxanthum odoratum* (sweet vernal) (2%) (Slay 2006). These species and their distribution were probably similarly represented in this study.

Laboratory measurements of *B. poae* development

The behaviour and development of young mealybugs was measured by transferring a single *B. poae* female plus eggs to a mealybug and endophyte-free perennial ryegrass plant (grown from seed supplied by Dr. A. Popay, AgResearch, Ruakura) in a 10 cm diameter pot, so that newly emerged crawlers dispersed on to the tillers. Five newly infested tillers were marked per pot, and the presence and development of the mealybugs were observed periodically over the following weeks until they could no longer be found. Fifty *B. poae* crawlers on 10 pots were examined.

Morphometric characterisation of *B. poae* instars

An attempt to characterise the instars of *B. poae* morphometrically was made. This exercise can be a valuable aid to determining seasonal population dynamics through

knowledge of the presence of growing life-stages – especially of small or cryptic insects. The leg-length of hemimetabolous insects has frequently been used to characterise different instars. Here, size was measured under a binocular compound microscope from two parameters, leg length (tibia+tarsus) and body length, of freshly collected (i.e. not after storage in any preservation fluid) specimens held under a coverslip on a microscope slide.

The smallest mealybugs were collected directly from wax cells in association with ovipositing females, and were designated as crawlers. Adult females were designated as those that were found in wax cells but had not yet produced eggs (and so were not moribund). Other mealybugs, of unknown age but presumably including other life-stages, were collected from the field and laboratory and measured against these benchmarks to characterise second and third instar females.

RESULTS AND DISCUSSION

Mealybug species

More than 600 core samples were received. About half were examined in detail for mealybug and natural enemy activity. Two species of mealybugs, *B. poae* and *Ripersella rumicis*, were found. The grass species in individual cores were not identified, but the two mealybug species occupied quite separate niches, with *B. poae* rarely found deeper than 2 cm below the grass crowns in the soil, while *R. rumicis* (a true 'root mealybug') were usually found towards the bottom of the soil cores, 5–7 cm below the surface (Fig. 1). No biological interaction between them was ever observed.

FIGURE 1: Mean depth (cm) of individual *Balanococcus poae* and *Ripersella rumicis* from soil cores collected from Hawke's Bay farms at Kereru and Maraetotara from 2006 to 2008. Values are the mean from 25–43 cores (see Table 1).

The collection of *B. poae* in June 2005 was the first record of this mealybug from Hawke's Bay. Numbers and presence of *B. poae* in the cores from the Kereru site were consistently low from 2006–07, despite the high numbers found in the June 2005 samples. Consequently, sampling was switched to Maraetotara, where *B. poae* presence then increased significantly from May 2007 (Fig. 2). It is not known whether the increase

in *B. poae* at Maraetotara (and the decline in *R. rumicis*) reflected structural population differences at the two sites, or seasonal environmental differences between 2006-07 and 2007-08 that affected populations at both sites. A single datum point (March 2007) suggests the latter. This was the first collection from Maraetotara, but the ratio of the two mealybugs in the cores was similar to those at Kereru in previous samples. The subsequent decline in *R. rumicis* (as a percentage of occupied soil cores) at Maraetotara (Fig. 2) may indicate that optimum population growth of the two species occurs under different environmental conditions – e.g. soil temperature and/or moisture – such that one or other of the mealybugs in any pasture may be more or less common in different years. The reversal of the species frequency in March 2008 is probably a sampling anomaly, but this datum point illustrates that further studies are necessary to understand the population dynamics of these two mealybug species.

FIGURE 2: The percentage of soil cores containing *Balanococcus poae* and *Rippersella rumicis* collected from farms at Kereru and Maraetotara, Hawke's Bay, from 2006 to 2008. Values are the mean from 25-43 cores (see Table 1).

Natural enemies

The mature adult female *B. poae* within their wax cells below the soil surface appeared to be completely protected from natural enemies. No predators or parasitoids of mealybugs were found in association with them, and no signs of predation or parasitism (i.e. partly eaten or mummified mealybugs) were found. No predators or parasitoids of *R. rumicis* were found either.

Seasonal phenology

Adult female *B. poae* were recorded in every sample except for one (November 2006), and so were present throughout the year (Table 1). All of the adult female *B. poae* found in every core sample were counted on each occasion. The mean number per core was calculated and, by simple extrapolation, the approximate density of adult females/m² of pasture was calculated. An estimated peak density of 1000-1300 adult females/m² of pasture was recorded in late autumn of 2007 at Maraetotara (Table 1).

TABLE 1: Mean numbers of adult female *Balanococcus poae*/core and /m² on different sample dates, and presence or absence of eggs and nymphs. Samples were taken from Kereru (K) or Maraetotara (M).

Sample date	No. cores examined	No. <i>B. poae</i> adults		Presence of	
		Mean/core sample	Estimated/m ² of pasture	Eggs	Neonates
June 2006 (K)	40	0.42 ($\pm$ 0.23)	212.5	No	No
October 2006 (K)	40	0.08 ($\pm$ 0.04)	37.5	No	No
November 2006 (K)	40	0.00	0.0	No	No
January 2007 (K)	40	0.15 ($\pm$ 0.07)	75.0	Yes	Yes
February 2007 (K)	40	0.05 ($\pm$ 0.05)	25.0	No	Yes
March 2007 (M)	40	0.23 ($\pm$ 0.08)	112.5	No	No
May 2007 (M)	25	2.08 ($\pm$ 0.40)	1040.0	No	No
June 2007 (M)	25	2.58 ($\pm$ 0.45)	1340.0	Yes	No
September 2007 (M)	43	1.42 ($\pm$ 0.22)	709.3	Yes	No
October 2007 (M)	40	1.53 ($\pm$ 0.24)	501.5	Yes	Yes
December 2007 (M)	30	0.90 ($\pm$ 0.27)	450.0	No	Yes
January 2008 (M)	40	0.40 ($\pm$ 0.09)	200.0	Yes	Yes
March 2008 (M)	40	0.28 ($\pm$ 0.11)	137.5	No	Yes
May 2008 (M)	40	0.80 ($\pm$ 0.16)	400.0	No	Yes

The presence of eggs and nymphs in the cores, but not their numbers, was recorded. Adult numbers at Kereru were low, and few eggs or nymphs were found. At Maraetotara, eggs were most commonly found in winter and spring, from June–October, and coincided with peak numbers of females (Table 1). Young nymphs were then found from spring through the summer and into autumn (Table 1). Although eggs were first found in June 2007, neonate nymphs were not seen until October 2007, suggesting that eggs may require warmer spring soil temperatures before hatching, and that the season's first eggs may not emerge for nearly 4 months after being laid. These dates are similar to those recorded in a separate study in 2005–06, when crawlers above ground were recorded in sticky traps from December to March, peaking in January (Slay 2006). The presence of eggs throughout the winter and early spring, followed by neonate nymphs from spring and over summer, indicates that there is typically a single generation each year in Hawke's Bay. However, the presence of eggs in January, following a sample in which none was found, raises the possibility that there was a partial second generation in summer resulting from the first cohorts of eggs that emerged in spring. The crawler presence between October and May may also have included two generations, as previously reported in Canterbury (Baxendale et al. 2004; C.G.L. Pennell, AgResearch, pers. comm.). Finer definition of the age-structure of the *B. poae* population from the Maraetotara data was not possible, and further data are required to clarify the voltinism of *B. poae* in Hawke's Bay.

Regardless of the number of generations per year, the consistent presence of adults throughout the year indicates that the life-cycle of an individual *B. poae* in these grazed pastures featured a 3–4 month period of nymphal development, followed by a relatively lengthy period of adult female pre-oviposition, oviposition and egg development in the shelter of a subterranean wax cell.

Peak numbers of *B. poae* reported here (ca 1300 adult females/m² (Table 1)) were possibly at least as high as those reported in Christchurch by Pennell et al. (2005), who found up to 11,710 *B. poae* of all stages/m² in endophyte-free ryegrass. Laboratory experiments to measure physiological development over time at different temperatures may be required to confirm the voltinism of this mealybug throughout its range, and on different host plants, in New Zealand.

Life-stages of *B. poae* in the field and laboratory

The signature white flocculent wax excreted by *B. poae* about 1-2 cm below the soil surface almost always formed part of a protective wax 'cell', usually found close to a root but separate from it. Adult female *B. poae* within the cell laid eggs, which hatched to first instar crawlers. Eggs and young nymphs were sometimes present together, but the young mealybugs soon dispersed, and were rarely, if ever, seen feeding on roots beneath the surface.

Dispersing nymphs observed on potted ryegrass plants in the laboratory crawled upwards through the grass root mass and on to the tillers. They then crawled to the tiller tips, from where, under favourable wind conditions, they would probably disperse – literally to pastures new – as observed by Pearson (1988). In the laboratory, in the absence of wind and after an undetermined time, crawler behaviour changed to a post-dispersive phase, and they moved back down the tillers to feed. Crawlers on marked tillers frequently disappeared between one observation and the next, indicating that developing nymphs were quite mobile. Removal and dissection of tillers showed that young mealybugs frequently fed beneath the tiller sheath around the ligule. The cryptic nature of the mealybugs and difficulty in distinguishing one instar from the next meant that it was not possible to map their development over time. All monitored mealybugs had vacated the marked tillers after about 3 weeks, presumably to complete their life-cycle on or below the plant crown.

No males were found in either field-collected or laboratory-observed *B. poae*.

Morphometric characterisation

Measurements from a total of 101 mealybugs were collected. Of these, 38 were designated crawlers (first instars) and nine were adult females. The remaining 54 were nymphs of unknown instar. Hind tibia+tarsus length increased from a minimum of 0.08 mm in known crawlers to a maximum of 2.7 mm in the largest adult females (Fig. 3a). The leg lengths of most nymphs were very similar, and the variation in size of crawlers was almost as great as that for the unknown instars. Consequently leg length could not be used to discriminate between different instars. Legs of *B. poae* are abnormally small and 'somewhat distorted' compared with those of other *Balanococcus* species (Cox 1987). These features, in combination with the limitations of measuring fresh specimens unprepared for microscopic examination, may have obscured incremental increases in length with instar. Body size provided a better indication of the different instars, but again, failed to discriminate clearly between them (Fig. 3b). However, all adults had a body length greater than 1.5 mm and most nymphs smaller than 0.5 mm long were first instars, while the remainder (including second and third instar nymphs) were between 0.5 and 1.5 mm long (Fig. 3b).

Male mealybug nymphs are typically longer and thinner than females, as observed by Pennell et al. (2004), and usually have wingbuds that are progressively more pronounced in the third and fourth instars. No males were found during the study. Their absence may simply be because the Hawke's Bay sampling regime was inadequate to detect them, but it may also indicate that they were naturally rare, or that *B. poae* is facultatively parthenogenetic, with population differences due to region or host-plant variation throughout the country.

FIGURE 3: Measurements of (a) hind tibia+tarsus length (mm) and (b) body length (mm) from 101 *Balanococcus poae* individuals collected from Hawke's Bay or reared in a laboratory colony.

Distribution of *B. poae* in New Zealand

The sites at Kereru and Maraetotara are rather typical examples of farmlands in Hawke's Bay and the presence of *B. poae* in both indicates that the mealybug is widespread through the region, and possibly throughout the east coast of the North Island. Maskell (1879) did not provide collection data for his specimen. De Boer (1968) noted specimens from Palmerston North, Nelson, Port Hills (Canterbury) and Oamaru, and Cox (1987) recorded collection data in the New Zealand Arthropod Collection from the regions of Wanganui, Wellington, Nelson, Mid-Canterbury, Central Otago and Dunedin; and on host plants in the Poaceae, Cyperaceae and Ericaceae. The breadth of these records, coupled with the Hawke's Bay data here, makes it likely that *B. poae* is widely distributed throughout both islands of New Zealand, although its origins on tussock may mean that it will prove to be more common in the North Island in cooler climates (e.g. at higher altitudes).

Control of *B. poae* in pastures

Odiaglyptus biformis, which to date is the only known parasitoid of *B. poae*, has been found throughout New Zealand, from Northland to Stewart Island, and from a wide range of habitats (Noyes 1988). Such a wide distribution suggests either that

B. poae is similarly widespread, or that *O. biformis* is polyphagous. However, neither *O. biformis* nor any other parasitoid or mealybug predator was found in the soil cores from this study, suggesting that biological control of *B. poae* in these Hawke's Bay pastures was effectively non-existent. Clearly, adult females in wax cells below the sward are very well protected against any natural enemies other than soil predators. Immature mealybugs feeding under tiller sheaths are also protected. Yet *O. biformis* has been collected from pasture (Pearson 1988; Noyes 1988) so the pasture environment itself is not completely inhospitable to these parasitoids. It may be that there are significant differences between managed pastures with discrete boundaries and landscape features (such as hedges) and extensive permanent pastures such as those sampled in Hawke's Bay. It is also not known how animal stocking rates and rotations affected either mealybug or natural enemy populations. A study of the population dynamics of both *B. poae* and its natural enemies in their natural tussock environments may throw some light on any significant differences in pastures.

In the meantime, recent studies have clearly demonstrated the negative effects of endophytes on *B. poae* population dynamics (Pennell et al. 2005), suggesting that plant breeding for mealybug resistance through endophyte manipulation may be the most effective way to manage this pest in pastures.

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