RELATIONSHIP BETWEEN *SITONA LEPIDUS* LARVAL NUMBERS AND WHITE CLOVER SEED PRODUCTION

P. J. GERARD

AgResearch, Ruakura Research Centre, Private Bag 3123, Hamilton

Corresponding author: pip.gerard@agresearch.co.nz

ABSTRACT

Plots of two cultivars of white clover (*Trifolium repens*), Grasslands Prestige (small-leaved, early flowering) and Grasslands Kopu (large-leaved, later flowering) were established in cold-frame beds. Clover root weevil (*Sitona lepidus*) larval numbers were manipulated to obtain winter larval densities ranging from 0 to over 1000/m². Increasing winter larval densities increased Prestige seed production (g/m²) when harvested on 28 December and 10 January and the trend was still evident on 24 January. No such relationships were found in Kopu for the early and middle harvests, but at the late harvest, increasing winter larval densities had a negative impact on seed production. The results indicate that the effect of clover root weevil larvae on second year clover seed crops may vary substantially between cultivars, and may even be positive rather than negative.

Keywords: *Sitona lepidus*, white clover, seed production.

INTRODUCTION

Clover root weevil (CRW) (*Sitona lepidus* Gyllenhal (Coleoptera: Curculionidae)) was first identified in New Zealand in 1996 (Barratt et al. 1996). It is now found in farmland from Whangarei to south of Te Kuiti and will eventually spread throughout the country, including the white clover seed production regions in the South Island. First year clover seed crops are likely to be rapidly colonised by CRW adults if there are infested pastures in the district. Although feeding by adults is unlikely to have any negative impact on flowering and seed production (Gerard et al. 1999), larvae resulting from eggs laid by invading adults may jeopardise a low-cost, high yielding second crop.

This paper summarises results from the second year of a two-year project investigating the impact of CRW on two contrasting clover cultivars. Year 1 had focused on the impact of adult damage. The aim in Year 2 (2000/01) was to quantify the impact of CRW larvae on second crop clover seed production.

METHOD

The trial used clover plots established in six 10 × 1 m cold-frame beds at Ruakura Research Centre, Hamilton in the 1999/2000 season. Each cold-frame bed had been divided into ten 1 × 1 m plots using 15 cm deep wooden barriers, giving a total of 60 plots. Grasslands Prestige (a small-leaved, early flowering clover) was established in 30 randomly selected plots and Grasslands Kopu (a large-leaved, later flowering clover) in the remaining 30 plots.

The inter-plot variation in winter 2000 CRW larval densities for each cultivar was generated and maintained as follows.

1. Nil larvae plots: Ten plots of each cultivar had been caged over summer in Year 1 to exclude adults. Once cages were removed in January 2000, raised barriers with a Stickem[®] strip were placed around the plot perimeters to prevent weevil adult entry, and pyrethrum (Yates) was applied to the foliage and barrier surfaces in the early evening at weekly intervals to kill any adults that may have flown in.

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2. High larvae plots: Ten plots of each cultivar had been exposed to high number of adults (500 adults/cage) over summer. Supplementary eggs, collected from weevil adults held in the laboratory, were added in April 2000, and plots were watered to enhance early instar establishment.
3. Based on plant cover, adult damage and larval data collected in April, the remaining plots were designated as likely to have low, medium and medium-high larval numbers. To maintain this variation, medium-high plots received water at the same time as the high larvae plots. Conversely, the top canopy of leaves was removed from the low larvae plots to increase drying of the soil surface and thus egg and neonate larval mortality, and to deter further adult oviposition activity.

This approach was used rather than varied application rates of a soil insecticide against the larvae, as soil insecticides would have impacted on other pests attacking clover roots in the plots, such as whitefringed weevil, grass grub and nematodes. The effect of one additional watering or light clipping during mild autumn conditions was considered unlikely to influence plant development such that it would impact on seed production eight months later.

On 2 August 2000, larval numbers were assessed by taking four 76 mm diameter soil cores/plot to a depth of 10 cm. Each core was sorted by hand and the number of larvae recorded. August sampling gives optimal estimates of damaging larval winter populations prior to larvae entering non-feeding prepupal and pupal stages in late spring. Early instar summer larval populations were not sampled because flower production commenced before adult emergence, and adults could move freely across all plots. Foliage samples (lamina, petiole and some shoots) were taken from each plot on 27 September and 1 November and submitted to Celentis Analytical for analysis of % foliar N.

Inflorescence density (those with at least 50% fully-formed florets) was assessed at weekly intervals from mid December until 18 January. Ten ripe seed heads were harvested from all plots on 28 December (early harvest), and 50 seed heads on 10 January (middle harvest) and 24 January (late harvest). These terms relate to this trial only, not to timing of harvests by the clover seed industry. Both cultivars were harvested on the same date. Seeds were removed from the seed heads, and the weight of seed/head collected determined for each plot. An estimation of seed yield/m² was obtained by multiplying weight of seed/seed head by plot inflorescence density. Inflorescence densities corresponding to the early, middle and late seed harvests were determined on 14 December, 21 December and 4 January respectively.

Data was analysed by regression with and without the nil larvae plots. As the treatments to exclude adults from the original nil larvae plots were not 100% effective, the nil larvae plots used in analyses consisted of only 13 of the original 20 plots treated with pyrethrum and one untreated plot. Cultivar means were compared using t tests.

RESULTS

A good distribution of larval densities was achieved across the trial site with at least six control plots containing no larvae and at least three plots exceeding 1000 larvae/m² for both cultivars.

Mean % foliar N for Prestige was significantly higher than Kopu, both on 27 September (5.35 vs 4.94%, $P < 0.001$) and on 1 November (5.09% vs 4.69%, $P < 0.001$).

Overall, spring growing conditions contributed to no significant difference being found between cultivars in inflorescence timing and production although Prestige peaked at a higher density ($423 \pm 17/\text{m}^2$) than Kopu ($296 \pm 8/\text{m}^2$) ($P < 0.001$) (Fig. 1). Increasing numbers of larvae present in Prestige plots in August appeared to have a positive effect on the peak number of inflorescences produced (Fig. 2., $r^2 = 0.29$, $P < 0.01$). No such relationship was found for Kopu.

When the early (28 December) seed weight/head and 14 December flower density data were combined to obtain seed weight/m², regression analysis showed a positive linear relationship between seed weight/m² and August larval numbers for Prestige ($y = 0.009x + 14$, $r^2 = 0.27$, $P = 0.01$). No such relationships were found for Kopu.

FIGURE 1: Kopu and Prestige mean inflorescence density at five sampling dates in summer 2000/01.

There was a significant relationship between August larval numbers and g seed/50 heads at the peak harvest on 10 January for Prestige ($y = 0.0007x + 1.96$, $r^2=0.30$, $P<0.01$). Using the 21 December flower numbers, the estimated seed produced per plot was highly significantly related to the number of larvae infesting that plot in the preceding winter (Fig. 3. $r^2=0.30$, $P<0.001$). As with the early harvest, no such relationships were found for Kopu.

No relationship was found between winter larval numbers and g seed/50 heads for Prestige at the 24 January harvest, although seed/m² maintained a tendency to increase

FIGURE 2: Winter CRW larval numbers and peak inflorescence density on Prestige plots.

with increasing larval numbers in the preceding winter (omitting nil larvae plots $P=0.059$). Conversely, for the first time a negative relationship was found between seed yield data and larval numbers for Kopu plots. The weight of seed/head in Kopu declined in response to the larval numbers present six months previously (Fig. 4. $P<0.01$). However, this did not significantly impact on the overall weight of seed/m² harvested from Kopu plots.

FIGURE 3: Winter CRW larval numbers and seed weight/m² from Prestige plots at peak harvest (omitting nil larvae plots).

DISCUSSION

The presence of CRW larvae in winter appeared to have a beneficial effect on Prestige seed production. Over the range of larval densities in this trial (maximum density 1370/m², cf. maximum pasture population recorded 1500/m², B. Willoughby, unpubl. data), higher larval densities in winter were associated with greater production of seed/m² in the early and middle harvests. Kopu did not show a similar response. Although overall production was not affected, this cultivar showed a negative response in weight of seed/head to winter CRW larval numbers, but only at the final harvest (24 January).

This marked difference in cultivar response to the abundance of CRW larvae in pure

FIGURE 4: August CRW larval numbers and weight seed/head from Kopu plots on 24 January (omitting nil larvae plots).

clover swards is noteworthy. CRW larvae attack the roots, stolons and nodules and thus affect nutrients, particularly nitrogen, available for vegetative and reproductive growth. In the trial area, the clover was healthy, and even in the plots with highest larval numbers, 100% clover cover was maintained and new nodules continued to be produced on the roots. However, nodule damage was very evident in many plots, and both cultivars showed significant decreases in % foliar N in November in response to higher winter larval numbers (P. Gerard, unpubl. data).

Grasslands Prestige is a small-leaved early flowering clover and Grasslands Kopu is a large-leaved later flowering clover. It is known that clover cultivars do differ widely in sensitivity to N when in mixed swards. For example, Wilman & Asiegbu (1982) found applied N to pasture had a very large adverse effect on stolon length in small-leaved varieties but only a modest effect in medium-large leaved varieties. Furthermore, the application of N increases vegetative growth to the detriment of seed production (Clifford 1987) by increasing the ratio of vegetative to reproductive nodes on stolons. It is possible that Prestige was more sensitive than Kopu to the larval-induced changes in available N within the plant, even in the absence of competing pasture species, and that this sensitivity to decreased available N was expressed in some of the components contributing to seed yield, such as numbers of vegetative nodes (inflorescence density) and seeds/head.

The disappearance of the positive influence of increasing larval numbers on Prestige seed yield components at the final harvest, and the appearance of a negative effect on seed weight/inflorescence in Kopu, could be related to seasonal depletion of plant reserves. For example, mean foliar N levels for both cultivars on 1 November were 5% lower than the means on 27 September. If low levels of available N within a plant impair some seed yield components, such as pollen or ovule fertility, any limitation is likely to increase as the season progresses. In addition, as Kopu has lower initial N levels, it is likely to show evidence of depletion before Prestige.

The relationships found between larval numbers and some of the seed yield components were not linear at very low larval densities, as plants in the nil larvae plots responded differently to those with larvae. It is possible that measures used to achieve nil larval densities may have affected the incidence of other pests establishing in these plots, or as with some other root-feeding pests, low pest numbers can enhance vegetative growth. Low infections of plant parasitic nematodes have been shown to increase availability of plant nutrients (Yeates et al. 1999). For simplicity of presentation in this paper, most analyses presented have omitted the nil larvae plots. The low correlation values (r^2 range 0.27–0.57) for most relationships show that larval numbers were not the only factors influencing seed production in this trial.

In summary, the results indicate that the impact of CRW on second year crops varied between cultivars. Crops of early flowering, small-leaved cultivars that have high inherent % foliar N levels, such as Prestige, may benefit from infestation by CRW larvae and specific measures against CRW are not warranted. Conversely, infestations in crops of late flowering medium-large leaved cultivars with lower inherent % foliar N, such as Kopu, may have minor negative impacts on crops harvested late in the season. Growers of these crops may need to include CRW when considering their overall pest management strategies, but specific controls are unlikely to be cost-effective.

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