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Biology of insects that feed in the inflorescences of *Chionochloa* (Poaceae) in New Zealand and their relevance to mast seeding

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Abstract Inflorescences of New Zealand *Chionochloa* species are attacked by at least three insects, two flies and a moth. There has been disagreement about the identity of various life stages of these insects. We followed the seasonal pattern of occurrence of the two fly species that fed in *Chionochloa pallens* inflorescences in a population on Mt Hutt, Canterbury. Eggs and larvae of *Diplotoxa similis* (Diptera: Chloropidae) appeared in the inflorescences as soon as they emerged. The larvae are principally flower feeders, and most *D. similis* individuals had pupated by the end of the flowering period. *Diplotoxa similis* adults emerged from the puparia at the end of the season, and probably overwintered as adults. The second fly was an undescribed cecidomyiid (Diptera: Cecidomyiidae). Eggs of the cecidomyiid are laid into the *C. pallens* florets at the time of flowering, and hatch into mobile, translucent, early-instar larvae. Late-instar larvae were less mobile and opaque orange, and probably dropped from the inflorescences late in the season. The third species, *Megacraspedus calamogonus* (Lepidoptera: Gelechiidae) has large mobile caterpillars which appear early in the season, but there is doubt about its egg morphology and oviposition sites. On two dates there was a negative correlation between densities of the two fly species among plants. Since *D. similis* appears first, it may be able to usurp resources and reduce densities of the cecidomyiid. A review of known occurrences of the three insects suggests that, compared to *D. similis* and *M. calamogonus*, the cecidomyiid (1) has a greater geographic range and (2) occurs on more *Chionochloa* species; however, these trends might be due to poor sampling of *D. similis* and *M. calamogonus* early in the season. The cecidomyiid appears to be less easily satiated than *D. similis* by masting in *Chionochloa*.

Keywords Cecidomyiidae; *Chionochloa* (Poaceae); *Diplotoxa similis* (Diptera: Chloropidae); distribution; host range; insect flower feeding; mast seeding; *Megacraspedus calamogonus* (Lepidoptera: Gelechiidae); New Zealand grassland; predispersal seed predation; seasonal phenology

INTRODUCTION

New Zealand *Chionochloa* species are characterized by mast seeding, i.e., great interannual variation in seeding that is synchronized among individuals within populations (Connor 1966; Mark 1968; Kelly et al. 1992; McKone et al. 1998). The variation in reproductive intensity among years in *Chionochloa* spp. is the highest reported for any plant species (Webb & Kelly 1993; Kelly 1994; McKone et al. 1998; Kelly et al. 2000).

Predator satiation (Janzen 1971; Silvertown 1980; Kelly 1994) is the leading hypothesis to explain the evolutionary origin of mast seeding in *Chionochloa*

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(Kelly et al. 1992; Kelly & Sullivan 1997; McKone et al. 1998; Tisch & Kelly 1998). According to this hypothesis, mast seeding would be selectively advantageous if specialized seed predators were satiated during the occasional mass-flowering years and their populations were kept relatively low during years of low plant reproduction.

A number of insect species feed in *Chionochloa* inflorescences (White 1975; Kelly et al. 1992; Sullivan 1993; Cone 1995; Kelly & Sullivan 1997) and thus could be important in the evolutionary origin and maintenance of mast seeding. These include a chloropid fly (*Diplotoxa similis*; Spencer 1977), an undescribed cecidomyiid fly, and a gelechiid moth (*Megacraspedus calamogonus* Meyrick 1885). To assess the role that each of these species might have played in the evolution of masting, it is important to know the nature of their life cycles and the type of damage they cause to *Chionochloa* plants.

There has been some confusion in the literature (reviewed in Cone 1995) about the identity of these insects, particularly in the egg and larval stages. One reason for this is that all previous studies of the insects (e.g., White 1975; Kelly et al. 1992; Sullivan 1993; Cone 1995) were done on *Chionochloa* samples collected at the end of the growing season (usually February). Late-season samples are useful in assessing total damage to *Chionochloa* seed production for a season (White 1975) or for counting late stages in the insect life cycle, but could miss important earlier stages. To clarify the identity and seasonal phenology of the insects, we conducted a longitudinal study on *Chionochloa* inflorescences at one site through one flowering season. We also collected supplementary information in a second season at several sites, and collated all available information about the host plant ranges and geographic distributions of the three insects.

METHODS

The main study site was a population of *Chionochloa pallens* Zotov at 1070 m altitude on Mt Hutt, Canterbury (43°32′S, 171°33′E). The population is on a southeast facing slope adjacent to the Mt Hutt skifield road, and has been the site of ongoing research on this system (McKone 1990; Kelly et al. 1992; Kelly & Sullivan 1997). Vegetation here is a relatively undisturbed tussock grassland with tussock cover of 94% *C. pallens* and 6% *C. macra* Zotov (McKone 1990; Kelly & Sullivan 1997). All three insects discussed above have been recorded on *C. pallens* from this site, although *M. calamogonus* has only been observed in very low numbers (Kelly et al. 1992; Sullivan 1993; Cone 1995).

The phenology study took place in the 1995–96 flowering season. We have measured flowering intensity in this population since the 1985–86 season (Kelly et al. 1992; McKone et al. 1998; Kelly et al. 2000). 1994–95 was a very heavy flowering year, the second-highest flowering intensity recorded in 12 years (including 1996–97). In contrast, the 1995–96 flowering season was the second-lowest flowering intensity in 12 years. Thus our study took place in a very low flowering year immediately following a masting season.

Sampling of inflorescences was carried out five times at approximately 14 day intervals from 19 December 1995 until 16 February 1996. On the first sample date in December, very few inflorescences had emerged from their sheaths, so the small sample (Table 1) was collected haphazardly from most of the plants with visible inflorescences. After that date, when inflorescences had appeared widely in the population and were fully exserted, approximately 20 plants were sampled from four parallel 40m transects that ran southwest to northeast across the slope. At 8m intervals along the transects, a single inflorescence was taken from the nearest flowering *C. pallens* plant. The starting point of the transect was varied on different sampling dates so that the same plants usually would not have been resampled. Each inflorescence was placed in a paper bag and frozen within 4 hours of collection to impede insect movement between florets, spikelets and inflorescences.

For each sampled inflorescence, florets from ten randomly selected spikelets were opened under a dissecting microscope. The presence of all insect eggs, larvae, pupae, puparia or adults was recorded for each floret. Each life stage was classified by type (e.g., "long white egg" or "orange larva") and eventually used to identify each of the stages. On 19 December, portions of the inflorescences had not yet emerged from the sheaths; counts of randomly sampled spikelets on that date were performed only on spikelets visible outside the sheath. In addition, we counted a haphazard sample of eight spikelets still enclosed within the sheath on 19 December.

Since the orange larvae found in *Chionochloa* inflorescences had never been definitively identified, we attempted to raise adults from the numerous larvae in our samples. Though we did not succeed in raising adults from Mt Hutt, we were able to rear two adult females from orange larvae from

Chionochloa spp. collected in Takahe Valley, Fiordland (45°16′S, 167°37′E). One of these females, as well as larvae from both Takahe Valley and Mt Hutt, were sent for identification to the cecidomyiid taxonomist R. J. Gagné of the Systematic Entomology Laboratory, US Department of Agriculture, Washington, DC, USA. On 13 January 2000 in the upper Otira Valley (42°54′S, 171°33′E, 1000 m altitude, 67 km north of Mt Hutt) numerous adult female cecidomyiids were observed ovipositing into the glumes and florets of *C. pallens* and *C. conspicua* right at the time of anthesis, and 18 females were collected.

To identify conclusively the insect responsible for the long white eggs found in florets, live material was collected from *C. pallens* and *C. macra* at the 1070 m site on 19 December 1999 and incubated in the lab before dissection. On the same date we also collected material from *C. rubra* at the base of Mt Hutt (450 m), where *M. calamogonus* has been more common (Cone 1995). With the assistance of E.G. White we examined preserved dry material collected by him in 1969, and also the negatives used to illustrate his 1975 paper on *Chionochloa* seed predators.

To see if the relative abundances of the cecidomyiid and *D. similis* vary in response to flowering intensity, we used the between-year data for predation and flowering in *C. pallens* at the 1070 m site from 1986 to 1999 (see Kelly et al. 1992; Kelly & Sullivan 1997). We calculated the number of florets showing definite or probable cecidomyiid damage, divided by the number of florets showing definite or probable *D. similis* damage. This ratio was regressed against flowering intensity (mean florets per tussock).

We also summarized all known observations of the three previously recorded insects that feed in *Chionochloa* inflorescences. The summary included published evidence as well as unpublished observations of D. Kelly and his various collaborators.

RESULTS AND DISCUSSION

We examined over 4000 individual florets for the presence of insects throughout the 1995-96 flowering and fruiting period of *C. pallens* at our Mt Hutt study site (Table 1). The two flies (*D. similis* and the cecidomyiid) were the dominant insects in the samples. There were no signs of *Megacraspedus calamogonus* larvae, shed head capsules or diagnostic feeding signs in any of the florets we dissected, although we did find *M. calamogonus* at this site in earlier years and in the 1999 *C. rubra* samples. We found thrips (Order Thysanoptera) in about 10% of florets in January and February samples. It is possible that thrips also damage florets as they are disproportionately common on damaged ovaries (Sullivan & Kelly 2000) but we do not consider thrips further in this paper. A very few florets (< 0.1%) contained Hymenoptera [possibly the parasitoids discussed by White (1975), Sullivan (1993) and Cone (1995)] or a larva tentatively identified by Cone (1995) as a pseudococcid. See Cone (1995) for illustrations and further discussion of these other insects.

Table 1 Sampling dates in 1995–96 and sample sizes in the study population of *Chionochloa pallens* at Mt Hutt, Canterbury. Sample size is the number of florets examined for the presence of insects, with the number of spikelets and plants from which these florets were taken.

Sample date	Reproductive stage[1]	No. of florets sampled	No. of spikelets sampled	No. of plants sampled
19 December	pre-anthesis	318	58	5
2 January	mid-flower	1114	200	20
16 January	end of flowering	1000	197	20
1 February	fruit maturation	976	207	21
16 February	fruit maturation	692	180	20

[1]pre-anthesis: most inflorescences not yet emerged from sheaths. None of the emerged florets had dehisced anthers.
mid-flower: most inflorescences emerged from sheaths. Stigmas visibly exsert from many florets. Anthers had dehisced in 40.6% of the florets that had no insect damage.
end of flowering: anthers had dehisced in 94.5% of the florets that had no insect damage.
fruit maturation: ovaries expanding and maturing into grains.

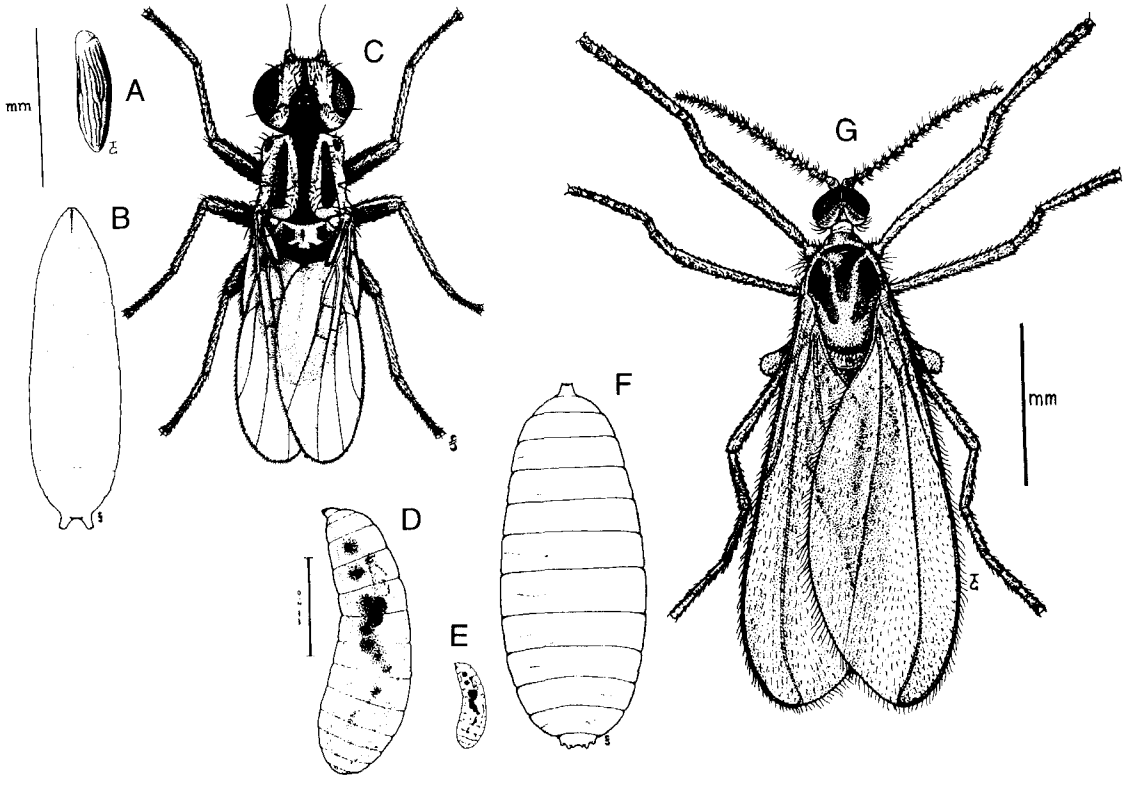

Fig. 1 Morphology of flies that attack *Chionochloa* inflorescences. *Diplotoxa similis* (Chloropidae): (A) egg; (B) larva; (C) adult. Undescribed cecidomyiid: (D, E) early-instar orange larva, shown at two magnifications; (F) late-instar orange larva; (G) adult female. All drawings are to the same scale (bar is 1 mm), except for D where bar is 0.2 mm. Drawings by J. Sullivan and T. Galloway.

Diplotoxa similis (Chloropidae)

There were often elongate, white eggs (Fig. 1A) in the spikelets of *C. pallens* collected from December 1995 through February 1996 (Fig. 2). The eggs are visible with the naked eye in the field, but seem to fall readily out of the spikelets if samples are handled roughly after collection. Most of the eggs were found in the glumes (Fig. 2). A few eggs had been laid in florets in a minority of spikelets (Fig. 2), but even in these spikelets there were often eggs also in the glumes. Eggs were found singly or in groups of up to 15 eggs, though we suspect the groups were not laid in a single oviposition event as the egg is large in relation to the fly (see below). We were able to distinguish hatched and unhatched eggs in counting, since hatched eggs appeared empty and flattened. On 19 December 1995 less than half of the eggs (45.6%) were hatched, but by 2 January 1996, 91.2% of the eggs had hatched. After that date, all of the eggs were hatched (with the exception of a single unhatched egg found on 1 February). These eggs resemble the eggs that White (1975; see below) identified as *Megacraspedus calamogonus*, but we found consistent associations between the white eggs and the presence of *D. similis* larvae or feeding signs, and no association with *M. calamogonus* larvae, head capsules or feeding signs.

The *C. pallens* material collected on 19 December 1999 also contained abundant white eggs (mostly hatched) and assorted live *D. similis* larvae ranging in size from 0.48 to 2.05 mm long. Three of the unhatched eggs were incubated overnight and one hatched into a small larva (0.42 by 0.12 mm) with the paired spiracles which are a distinctive feature of *D. similis*. Therefore these elongate white eggs are those of *D. similis*. They are consistent with Ferrar's (1987) general description of the size (0.5–0.9 mm) and appearance (cylindrical with longitudinal striations) of chloropid eggs (Fig. 1; Table 2). One-

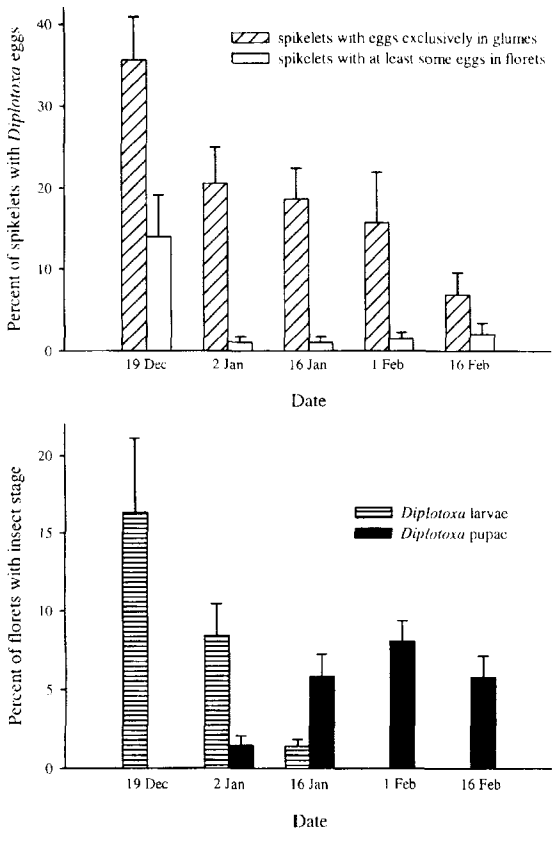

Fig. 2 Presence of *Diplotoxa similis* in *Chionochloa pallens* inflorescences through the season at the Mt Hutt study site in 1995–96. Top: the percentage of spikelets that had *D. similis* eggs either exclusively in the glumes or with at least some of the eggs in florets. Bottom: the percentage of florets with larvae or puparia. Error bars are standard error of the mean, with individual plants used as the replicates; see Table 1 for sample sizes at each date.

way ANOVAs revealed no significant differences in the length or width of eggs found inside *C. pallens* or *C. macra* florets in 1999, or in samples from *C. pallens* on 2 January 1996 (length: $F_{2, 123} = 0.64$, NS; width $F = 0.15$, NS). There was also no difference in length between eggs found in glumes or in florets ($F_{1, 100} = 0.01$, NS). Eggs from material collected by White in 1969 and measured in 1999 were identical in appearance with the same longitudinal striations and were fractionally smaller (Table 2). The size difference from our samples was significant (length: $F_{3, 169} = 6.53$, $P = 0.004$; width $F = 3.78$, $P = 0.012$), but could perhaps be partly attributed to shrinkage in 30 years of dry storage.

Our 1995-96 samples contained many of the distinctive green larvae (Fig. 1B) and dark puparia detailed in previous work on late-season samples (White 1975; Sullivan 1993; Cone 1995). The *Diplotoxa* puparia were within the florets, in contrast to the cecidomyiid. Contrary to the proposal of Kelly et al. (1992) that the larvae of *Diplotoxa similis* were orange (now known to be the cecidomyiid, see below), there can be no doubt that these larvae and puparia were *Diplotoxa similis*. Both larvae and puparia showed distinctive paired spiracles and a black cephalopharyngeal skeleton near the mouth (Ferrar 1987; see Cone [1995] for detailed descriptions); transitional forms are known between larvae and puparia; and *D. similis* adults readily eclose from the puparia in the lab. The adults are black with yellow markings.

The newly hatched *D. similis* larva in 1999 was completely transparent apart from the cephalopharyngeal skeleton. Slightly larger early-instar *D. similis* were translucent and appeared light green, gold, or rose. The colour of the early larvae was

Table 2 Dimensions (mm) of *Diplotoxa similis* eggs and *Chionochloa* floral parts from different host species and seasons. Egg measurements within a column followed by the same letter did not differ significantly according to Tukey HSD means comparisons (see text); the White fig 2 data were excluded from the analysis. A blank means no data were collected. "White 1969" is from our measurements in 1999 of physical samples kept by E.G. White from his work published in 1975, whereas "White fig 2" is from our scaled measurements of the photo in the 1975 paper.

Source	Egg length	Egg width	N	Spikelet	Glume 1	Glume 2	N
C. pallens 1999	0.73 a	0.18 a	67	15.9	9.4	10.9	10
C. macra 1999	0.74 a	0.18 ab	29	16.4	9.8	12.5	10
C. pallens 1996	0.74 a	0.18 ab	30				
C. rubra 1999				18.1	9.0	10.7	10
White 1969	0.70 b	0.16 b	47	16.8	11.5	13.3	12
White fig 2	1.58		8	30.7	21.0	≥20.8	3

derived at least partly from the contents of their digestive tracts, which contained floral parts. *Chionochloa* ovaries and immature anthers are light green; as they mature, anthers become gold or rose. Later *D. similis* instars are less transparent, and appear whitish green to dark green as reported by previous investigators.

Neither eggs nor larvae of *D. similis* were found in the sample of spikelets that were still enclosed in the sheath on 19 December 1995. This suggests that the spikelets were not available to ovipositing females until after they emerged from the sheath. The highest proportion of spikelets with *D. similis* eggs and of florets with *D. similis* larvae was recorded on 19 December (Fig. 2), and we attribute this to the general lack of inflorescences available to ovipositing females at this time. This could also account for multiple females ovipositing in the same glume, as would be required to produce the clusters of eggs we observed. By the time most inflorescences had emerged in January, the abundance of *D. similis* eggs and larvae was at approximately the level it would remain for the rest of the season (Fig. 2). The decrease of *D. similis* eggs in the final sample on 16 February is probably due to the loss of hatched eggs from the glumes. The empty eggs might fall out when florets open widely for dispersal of the mature grains; as explained above, empty eggs fall readily from florets if they are roughly handled.

The first puparia appeared on 2 January, and by 16 January the majority of *D. similis* were puparia (Fig. 2). The February samples contained only puparia.

We conclude that *Diplotoxa similis* eggs are elongated and white, and were laid in *Chionochloa* inflorescences early in the season, as soon as the inflorescences emerged from their sheaths. Eggs hatched immediately and larvae began feeding on floral parts (both stamens and pistils) before anthesis. By the time *Chionochloa* flowering ended in mid-January, most *D. similis* larvae had already pupated and they were causing no further damage to the plants. Previous work has shown that adults emerge from puparia at the end of the season (White 1975; Kelly et al. 1992; Cone 1995; pers. obs.). Thus the available information suggests that there is a single generation of *D. similis* per year, and that the flies overwinter as adults. This is supported by a collection of adults in late June (Spencer 1977).

Cecidomyiidae (undescribed species)

Starting in mid-January 1996 (Fig. 3), our Mt Hutt samples contained the immobile orange larvae that have been identified previously as cecidomyiids (Mark 1965a,b; Burrows 1968; Cone 1995). The orange larvae inside florets are clearly visible in the field if the spikelets are wet. R. Gagné (pers. comm.) identified the larvae as Cecidomyiidae, Supertribe Cecidomyiidi, and confirmed that these larvae appeared to be the same species as those from Takahe Valley. These larvae, and the adult reared from Takahe Valley, were from a species that is undescribed and "does not belong to any genus previously reported from grasses elsewhere in the world" (R. Gagné, pers. comm.). The adult (Fig. 1G) is black with a distinctive bright orange abdomen.

We agree with Cone (1995) that Kelly et al. (1992) were incorrect in their identification of the orange larvae as those of *Diplotoxa similis*.

We found what we believe to be the eggs and early-instar larvae (Fig. 1D, E) of the cecidomyiid. In our mid-flowering sample on 2 January 1996 (Fig. 3), we found small orange eggs. These eggs were rare by 16 January when flowering was ending, and were not found after that. Female cecidomyiids were observed ovipositing into florets at anthesis in the Otira Valley on 13 January 2000. Also in the January 1996 samples, a mobile larva appeared that was different in appearance from the larvae of *D. similis* (Fig. 1). The mobile larvae were small and mostly translucent, except for a distinctive orange centre. These were almost always located within the florets, near or on the ovary.

These were almost certainly the eggs and early-instar larvae of the undescribed cecidomyiid, based on their orange colour, time of appearance, and position within the inflorescence. Cecidomyiid larvae typically have three instars (Gagné 1989). We propose that the early instars are mobile, and partly translucent with an orange centre. Later instars (possibly the third instar only) are relatively less mobile, completely orange, and opaque. Most previous reports of the orange larvae (Kelly et al. 1992; Cone 1995) were from samples that apparently contained only the late instars, collected late in the season. Though all of Sullivan's (1993) *Chionochloa* samples were collected in February or March, some florets were still in early stages of development; these occasionally had small eggs or mobile larvae that he also identified as early stages of the immobile cecidomyiid larvae.

The cecidomyiid eggs must hatch rapidly, since they were common only in the 2 January sample (Fig. 3). Cecidomyiid eggs generally hatch within days of oviposition (Gagné 1989). Unlike the larger *D. similis* eggs, which remain visible in the inflorescence after hatching, there is apparently no easily

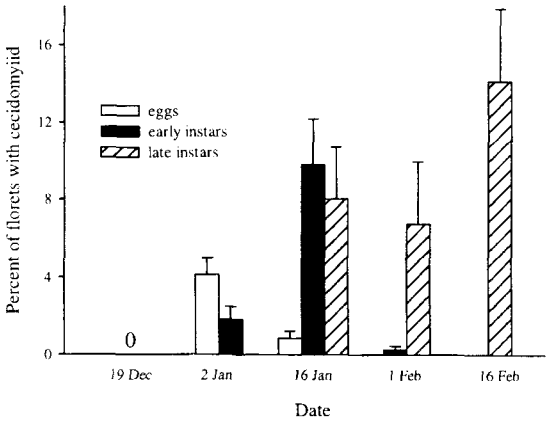

Fig. 3 Presence of the undescribed cecidomyiid in *Chionochloa pallens* inflorescences through the season at the Mt Hutt study site in 1995–96. Error bars are standard error of the mean, with individual plants used as the replicates; see Table 1 for sample sizes at each date.

visible remnant of the cecidomyiid eggs after hatching. The mobile instars must also be of relatively short duration, since they were common only in the 16 January sample (Fig. 3).

We found no puparia of the cecidomyiid in any of our *Chionochloa* samples, in agreement with the results of Sullivan (1993) and Cone (1995) on late-season samples. Larvae probably drop from the inflorescences late in the season (as suggested by Cone 1995). Larval diapause is common in cecidomyiids (Gagné 1989), so it seems likely that larvae overwinter in the soil, although we do not know when they pupate. Adult eclosion from the puparium is almost certainly in summer during flowering; the Otira observations confirm that adults oviposit into open florets at that time. The few adults which we have successfully reared emerged from larvae which were kept at 4°C in a fridge from collection in autumn until spring; the adults emerged when the material was moved to room temperature about October. Many cecidomyiids have larval diapause that is extended over several years (Barnes 1956; Gagné 1989) so it is possible that some adults emerge more than one season after the larvae dropped from *Chionochloa* inflorescences.

We conclude that the orange larva that has been observed frequently in *Chionochloa* inflorescences is an undescribed species of cecidomyiid. In our population it oviposits at the time of anthesis in early January. Most feeding is done on the developing *Chionochloa* ovaries. Larvae drop from *Chionochloa* inflorescences late in the season, probably to overwinter in the soil. Adults emerge before or during flowering, in the next year or possibly after extended (multi-year) larval diapause in the soil.

Megacraspedus calamogonus (Gelechiidae)

In our 1999 sample of 40 *C. rubra* spikelets from Mt Hutt, there were abundant *M. calamogonus* larvae of various sizes; the larvae are pale with reddish longitudinal stripes. However, we saw no *D. similis* larvae in these samples, and no white eggs, or eggs of any other kind. White (1975) describes the eggs of *Megacraspedus calamogonus* as being laid singly or in groups under the glumes of the florets, and illustrates some in his fig. 2. In location and appearance from his photograph they are similar to the eggs we have shown to belong to *D. similis*. In White's stored material we found numerous white *Diplotoxa* eggs averaging 0.70 mm long, which is about 5% shorter than the eggs from our samples (Table 2). However the scale on fig. 2 of White (1975) suggests that the eggs in the photograph were 1.55–1.70 mm long (Table 2). This two-fold size difference would suggest that the eggs in his fig. 2 were different from *Diplotoxa* eggs and could be the eggs of *M. calamogonus*. However, we found no eggs in White's stored material longer than 0.79 mm. We suspect that the scale stated in the figure captions in White (1975) may be wrong by a factor of two, as the plant parts also seem too large. We measured the lengths of spikelets and the first and second glumes in White's fig. 2, in White's stored material from 1969, and in fresh material of *C. pallens*, *C. rubra* and *C. macra* in 1999. For all three plant parts we found close agreement between the lengths of our plant material and White's stored samples, whereas our scaled measurements from the photos were about twice as large (Table 2). The lemmas shown in White's figs 5–7 are also about twice as large as lemmas on material from 1999 and 1969 (data not shown). It is hard to understand how the photo scales could be wrong, since we have seen the originals and the tick marks in all his figures are included in the negatives, but we believe unless a physical specimen of a 1.5 mm white egg can be found their existence must be seriously in doubt.

The only other apparent invertebrate eggs we have seen in *Chionochloa* florets were found in material of *C. rigida* from Hawkdun Runs Road near St Bathans (44°50′S, 169°52′E) in central Otago, collected on 30 December 1998. This material had frequent larvae of *M. calamogonus* but no definite sign of either *D. similis* or the cecidomyiid. We

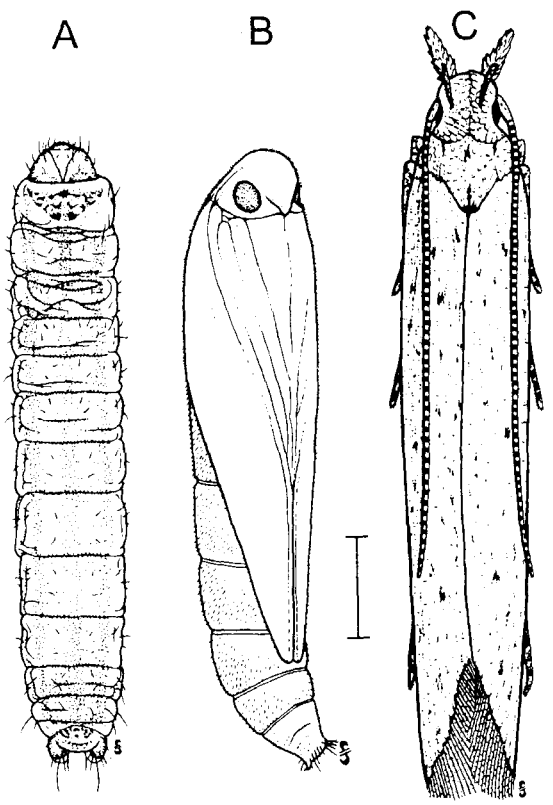

Fig. 4 Morphology of the moth that attacks *Chionochloa* inflorescences. *Megacraspedus calamogonus* (Gelechiidae): (A) larva; (B) pupa; (C) adult. All drawings are to the same scale (bar is 1 mm). Drawings by J. Sullivan.

found three elongate green eggs within florets. The surface of the eggs was smooth and undecorated; the wall of the eggs was transparent, and the contents were mid green in colour but undifferentiated in structure. The most clearly intact egg was 1.53 × 0.28 mm; the others were similar in length but had collapsed sideways. If these are insect eggs, they are different from those described by White, but could represent the eggs of *M. calamogonus*. More early-season sampling is necessary to verify this. In the meantime we conclude that the eggs of *M. calamogonus* are currently not known with any certainty.

We found no evidence of *M. calamogonus* in any of the longitudinal samples in 1995-96, but have seen it occasionally in other studies at Mt Hutt, especially at lower elevations (Sullivan 1993; Cone 1995, Sullivan & Kelly 2000). Our data are too sparse to indicate conclusively when *M. calamogonus* feeds relative to *D. similis*; Sullivan (1993) found *M. calamogonus* fed earlier than *D. similis*, whereas Cone (1995) found the opposite. We can conclude, however, that both *M. calomogonus* and *D. similis* feed earlier than the cecidomyiid. Our *M. calomogonus* larvae ranged in size from 0.8–6.0 mm long, and large specimens readily pupate in the lab. Pupae are in pale brown silk cocoons, usually outside of the florets. The white-coloured adults successfully emerged from these pupae after about two weeks (illustrated in Fig. 4). White (1975) notes that the larvae mature rapidly (in about 3 weeks) and move readily from floret to floret. He considered that larvae do not pupate in *Chionochloa* florets, contrary to Meyrick's (1885) report from *Cortaderia*; our observations support White as we did not find pupae in florets in the field. Adults apparently overwinter to lay eggs the following spring (White 1975); White (1964) found five adults in August in the base of a *Poa cita* tussock.

Although we found few *M. calamogonus* at our main study site, at other sites this species may be the most important seed predator and can destroy more than 50% of *Chionochloa* florets (White 1975; Sullivan & Kelly 2000).

We conclude that the eggs of *M. calamogonus* are not known with certainty, but the larvae feed on floral parts early in the flowering season. Larave may be very common at some sites, especially at lower elevations, but are rare at others. They usually pupate outside the florets. The adults eclose soon afterwards, and presumably overwinter as adults.

Interaction between the fly species

To determine the relationship between populations of *D. similis* and the cecidomyiid on individual host plants, we compared the percent of florets with each of the two flies on the last three sample dates (when the cecidomyiid populations had reached high levels; Fig. 3). We analyzed the data by means of the non-parametric Spearman rank correlation, using individual plants as the unit of replication. There was a significant negative correlation between densities of the two flies on both 16 January ($r_s = -0.665$, n = 20 plants, $P = 0.003$) and 1 February ($r_s = -0.643$, n = 21 plants, $P = 0.002$). The correlation was not significant on 16 February ($r_s = -0.257$, n = 20 plants, $P = 0.27$). The relationship between insect densities is shown in Fig. 5 for 16 January and 1 February. Except for a single plant on 16 January, the cecidomyiid density was *greater* than 10% of florets only in plants with a *D. similis* density of *less* than 10% of florets.

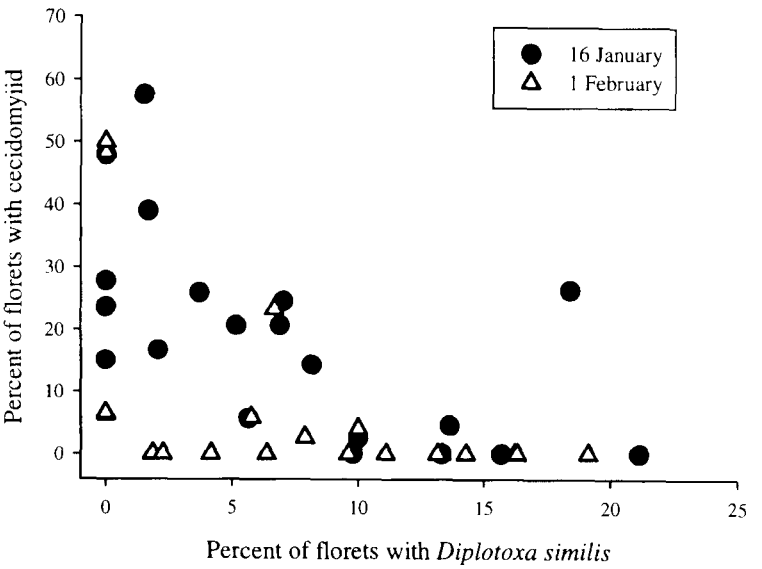

Fig. 5 Comparison of the density of *Diplotoxa similis* and of the undescribed cecidomyiid on individual *Chionochloa* plants. Data are from the Mt. Hutt study site in 1995–96. There was a significant negative correlation between the densities of the two species on the dates shown (Spearman rank correlation, $P < 0.01$), but not on 16 February. Approximately 50 florets were examined from each plant sampled; see Table 1 for details of sample sizes.

Since the two fly species both occur commonly in *Chionochloa* inflorescences and have some overlap in their food resources, it is possible that they compete with each other. If there is competition, the difference in seasonal phenology makes it likely that *D. similis* has a much stronger impact on the cecidomyiid than vice versa. *Diplotoxa similis* is active several weeks earlier in the season (compare Fig. 2 and 3), so plants heavily damaged by *D. similis* might have fewer intact florets to support cecidomyiids later in the season. This interaction is similar to that proposed by Harris & Chung (1998) for two insect species that feed on masting pecan trees (Juglandaceae: *Carya illinoensis*); *Acrobasis nuxvorella* (Lepidoptera: Pyralidae) feeds on developing nutlets early in the season, and thus removes potential resources for the late-season feeder *Curculio caryae* (Coleoptera: Curculionidae).

Alternatively, the negative correlation between densities of the two insects might have been caused by variation in the phenology of individual plants in the population. Plants with inflorescences that emerged early might have been exposed to high *D. similis* populations, since oviposition occurred as soon as inflorescences appeared. If the oviposition season of *D. similis* is short, later-emerging inflorescences might have been available at a time when only the cecidomyiid was ovipositing.

It is not clear why the negative correlation between insect densities was no longer significant on 16 February. This is late enough in the season that adults may have begun to emerge from some of the *D. similis* puparia, *Diplotoxa* eggs could drop from florets as the season progresses, or some cecidomyiid larvae may have already dropped from the inflorescences to the soil.

Geographical distribution and host range of the insects

Based on our revised criteria for the identification of the three invertebrate species, we have summarized known observations of the three *Chionochloa* feeding insects. Despite earlier confusion in the literature, we were able to determine the identity of insects in most previous reports. Observations of white eggs on spikelets, and white or green larvae, were taken to refer to *Diplotoxa similis*, while reference to orange larvae were taken to be the undescribed cecidomyiid. *Megacraspedus calamogonus* was identified either from presence of the distinctive lepidopteran larva, its head capsule, or distinctive feeding damage (White 1975). Because there were no obvious differences among insects taken from different *Chionochloa* species or from different locations (e.g., Table 2 and R. Gagné pers. comm.), we have assumed for the present that there are just three widespread insect species. This may not necessarily be so, and there remains the possibility of cryptic species or host races.

All three insects are found in a variety of *Chionochloa* species (Table 3). The cecidomyiid occurs in all eleven species for which we have data, while *D. similis* and *M. calamogonus* have been found in about half of these. The lack of records of

the latter insects from some *Chionochloa* species should not be taken as definitive evidence that the insects do not feed on these plants. In some cases authors reported on only one or two of the three insects, but have no definitive statement that the others are absent in that host species. Also, adults emerge in late summer in both *D. similis* and *M. calamogonus*, and so these insects may be missing from samples taken later in the season. Early-season samples would be necessary to make sure that these insects are not present at a particular location.

The three insects are recorded most often from *Chionochloa* inflorescences, and we think it likely that they are *Chionochloa* specialists. However, there is some evidence that they occur in other grass genera. *M. calamogonus* was first recorded from inflorescences of *Cortaderia* (Meyrick 1885). Cone (1995: 84) found what appeared to be a *Diplotoxa similis* individual feeding in a *Rytidosperma* inflorescence from Mt Hutt. A complete determination of host range for the insects would require more thorough sampling of other grasses. However, there are few other large-seeded grasses present at most of our *Chionochloa* study sites.

The geographical distribution of the insects is shown in Fig. 6, including *Chionochloa* samples in which particular species were found to be absent. The cecidomyiid appeared to be the most widespread, recorded in almost all samples, from the central North Island to Stewart Island. As with the distribution among host species, the apparent lack of *D. similis* and *M. calamogonus* may be due partly to poor early-season sampling. Nonetheless, based on the available evidence *D. similis* and *M. calamogonus* appear to be relatively uncommon in the southern South Island, especially in the wetter western parts of Otago and Southland. More sampling would be worthwhile. All three insects are readily observed in the field: *D. similis* eggs and the orange cecidomyiid larvae are visible with the naked eye in early- and late-season inflorescences respectively, while *M. calamogonus* larvae are easily shaken out of early-season inflorescences onto white paper.

Implications for mast seeding and reproduction in *Chionochloa*

The three insects have quite different life cycles, and could be expected to act differently as selective agents for the evolution of masting. As suggested by earlier investigators (Sullivan 1993; Cone 1995), it is clear that *D. similis* and *M. calamogonus* appear earlier in the reproductive cycle of *Chionochloa* than does the cecidomyiid. It is probably more accurate to refer to *D. similis* as a flower-feeder rather than a seed predator, since its larvae were mostly present before and during flowering and often fed on anthers.

Table 3 *Chionochloa* species in which the three seed predators (two dipterans and one lepidopteran) have been found. Identifications are based on presence of insects or insect remains and on characteristic feeding damage. They follow the sources cited, verified against the morphological descriptions given in this paper. Incorrect identifications in Kelly et al. (1992) were corrected to include both *Diplotoxa* and the cecidomyiid.

Chionochloa species	*Diplotoxa similis*	undescribed cecidomyiid	*Megacraspedus calamogonus*
C. australis		1, 3	
C. conspicua		8, 9	
C. crassiuscula		1, 8	
C. flavescens	4, 6	1, 3, 4, 6, 9	4, 6
C. macra	4	4, 7	4
C. oreophila		3, 9	
C. pallens	4, 5, 6	1, 3, 5, 6, 8, 9	5, 6
C. rigida	4	1, 2, 8, 9	4, 8
C. rubra	4, 6	6, 8, 9	4, 6
C. spiralis		8	
C. teretifolia		8, 9	

1 = Burrows (1961); 2 = Mark (1965 a,b); 3 = Burrows (1968); 4 = White (1975); 5 = Kelly et al. (1992); 6 = Sullivan (1993); 7 = Tisch and Kelly (1998); 8 = D. Kelly and A.L. Harrison (unpubl. obs.; 1995-96–1998-99, Takahe Valley); 9 = A.M. McCall (unpubl. obs.; 1998–99)

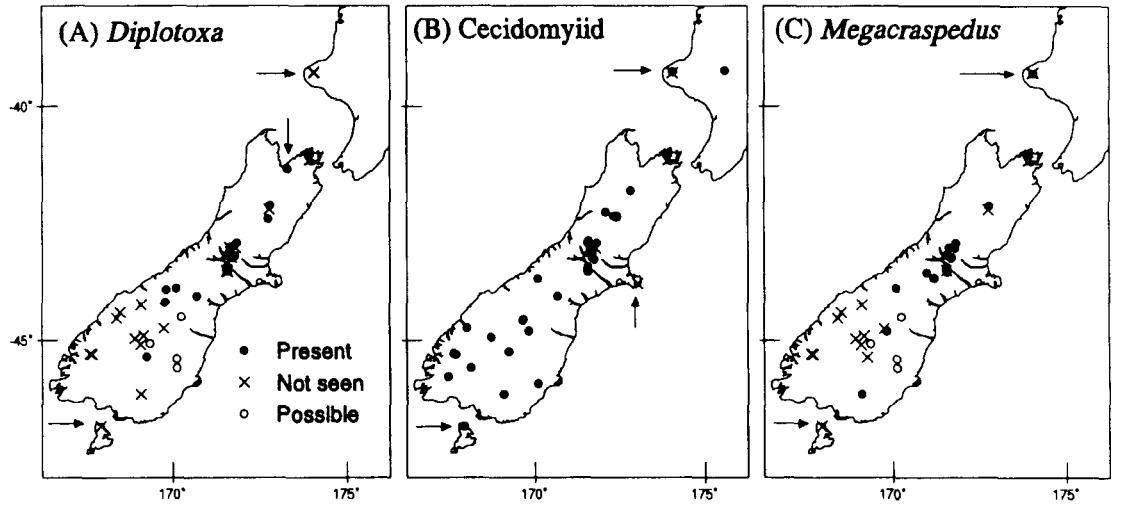

Fig. 6 Known distribution throughout New Zealand of three flower- or seed-feeding insects found on *Chionochloa*. "Not seen" indicates that observers with experience identifying that insect had sampled there and did not find the particular species. The four "possible" sites in eastern Otago are locations where White (1975) reported damage that showed at least one of *Diplotoxa similis* or *Megacraspedus calamogonus* was present, but could not be attributed definitely to either. Arrows point to outlying sites. Sources are as follows: Burrows (1961, 1968), 5 sites; Mark (1965 a, b), 2 sites; White (1975), 22 sites; Spencer (1977), 1 site; Sullivan (1993), 29 sites; D. Kelly and A. L. Harrison (unpubl. obs.), 9 sites; A. M. McCall (unpubl. obs.), 22 sites.

By the time seeds were maturing, most *D. similis* had already pupated (Fig. 3). *M. calamogonus* also feeds early, often on flowers rather than seeds. In contrast, damage by the cecidomyiid was restricted to the time of seed maturation, so it was exclusively a seed predator.

Since *D. similis* can completely consume both stamens and pistils in a floret (Cone 1995; pers. obs.), a heavy infestation could severely reduce both the male and female reproductive success of a *Chionochloa* plant. In contrast, cecidomyiid larvae feed too late to have much direct impact on the male reproductive success of a plant, and a *Chionochloa* plant heavily attacked by the cecidomyiid could still have high male reproductive success (unless the seeds it had fathered on another plant were also eaten by cecidomyiids).

The differences between the life cycles of the two flies also suggests that the predator satiation strategy would be more effective on *D. similis*. We infer that *D. similis* overwinters as an adult and it is most unlikely that adults last over more than one winter. Although adults are winged, they are reluctant fliers (Kelly et al. 1992) so adult dispersal distances would be expected to be low. Therefore a local *D. similis* population is completely dependent on an annual

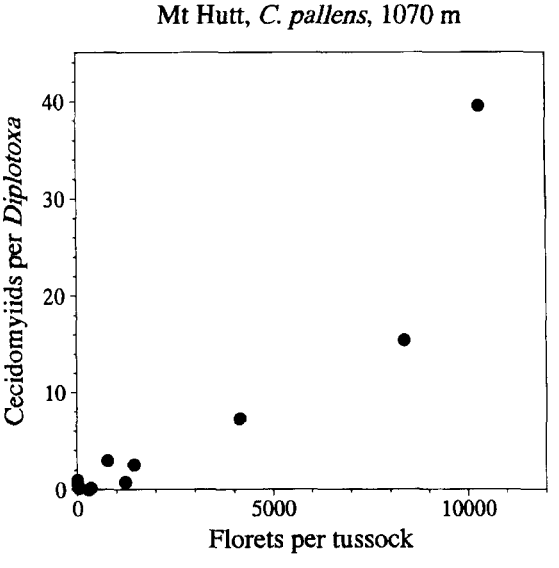

Fig. 7 Relative abundances of *D. similis* and the cecidomyiid in a temporal sequence of data, 1986–99 at Mt Hutt in *C. pallens*. The number of florets certainly or probably damaged by the cecidomyiid for each floret certainly or probably damaged by *D. similis* was positively related to the size of the flowering effort ($R^2 = 0.861$, $F = 61.97$, $n = 12$, $P < 0.001$).

supply of *Chionochloa* flowers. Low flowering years would presumably result in very small *D. similis* populations the following year. In contrast, if the cecidomyiid is capable of extended larval diapause, this would produce a temporal "refuge" in the soil by which the cecidomyiid population could survive during years of very low (or even no) *Chionochloa* flowering. There is evidence on variation from year to year in cecidomyiid predation from Takahe Valley which is consistent with the existence of extended diapause (Kelly et al. 2000).

The difference in the egg size between the two flies suggests a second reason why *D. similis* might be more easily satiated than the cecidomyiid. The eggs of *D. similis* are large relative to its body size (Fig. 1), which suggests that a female's reproductive capacity is relatively small. If a female *D. similis* can produce relatively few eggs, populations will be limited in their capacity to respond to superabundant floral resources in mast years. The eggs of the cecidomyiid are much smaller relative to adult body size, which suggests high potential numbers of eggs per female. Gagné (1989) states that there are often more than 100 eggs per female cecidomyiid. This high reproductive rate would mean that cecidomyiid populations would have a greater ability to track changing amounts of *Chionochloa* resources.

The temporal sequence of relative abundances of *D. similis* and the cecidomyiid from Mt Hutt support the suggestion that the cecidomyiid is less easily satiated. In years of low flowering (< 500 florets per tussock) there was always less than one cecidomyiid-damaged floret per *D. similis*-damaged floret. However, in higher-flowering years, the ratio rose steeply to peak at just under 40 cecidomyiid-damaged florets per *D. similis*-damaged floret in the highest flowering year (Fig. 7). This shows that the cecidomyiid increases much more than *D. similis* in high flowering years.

The egg size of *M. calamogonus* relative to the adult moth is unknown, but there are indications that it may also be harder to satiate by masting than *D. similis*. If *M. calamogonus* is better able to track varying *Chionochloa* resources, the proportion of florets destroyed by *M. calamogonus* relative to the fly should be highest in mast years. Such a trend is evident in White's (1975) data set from *C. flavescens*, *C. macra* and *C. rubra* at Porter River (30 km NW of Mt Hutt) from 1970–73. The ratio of ovaries eaten by *M. calamogonus* to those eaten by *D. similis* over all *Chionochloa* species was greatest (8.9) in 1971, a year of "heavy" flowering; intermediate (1.2) in 1972, a year of "moderate" flowering; and least (0.1 each) in 1970 and 1973, described respectively as years of "very light" and "sparse" flowering. This suggests that during White's study *M. calamogonus* responded to the increased abundance of florets in mast years more successfully than did *D. similis*. These qualitative data mirror exactly the pattern in the quantitative data of Fig. 7.

In summary, our data suggest that the different insects vary in their susceptibility to, and selective effects on, masting in *Chionochloa* species. The masting strategy appears not to satiate the cecidomyiid as effectively, presumably because of its greater reproductive capacity and the possibility of extended larval diapause. It would be instructive to determine whether reduced vulnerability of the cecidomyiid to predator satiation has allowed it to persist in more sites throughout New Zealand (especially where *Chionochloa* masting is most pronounced), and hence to show a wider geographic distribution than *Diplotoxa*. Also, since the cecidomyiid is very difficult to satiate and damages many seeds, this could help explain why the genus *Chionochloa* has such extremely high levels of between-year variability in flowering intensity compared with other masting species (Kelly et al. 1992; Webb & Kelly 1993; McKone et al. 1998; Kelly et al. 2000)

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