

# Enhancing nectar provision in vineyard habitats for the endemic New Zealand butterfly, *Lycaena salustius*

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**Abstract:** Nectar is an important factor influencing the level and persistence of butterfly populations, but particular sources of nectar may not be optimal for all species. In a farmland context, it is not always clear whether nectar sources used by butterflies are good quality species. They may be used opportunistically in the absence of true preferences, therefore possibly limiting maximal reproduction. This study investigated the use of nectar by adults of the endemic New Zealand butterfly, the common copper *Lycaena salustius*, in two ways: (1) a choice experiment in the field using a replicated design of different plant species, and (2) a greenhouse no-choice bioassay examining fitness enhancement by different flower species. In the field experiment, only *Lycaena salustius* males were observed in large numbers, and they spent a significantly longer time on flowers of *Veronica 'Youngii'* and *Fagopyrum esculentum* than on species already available in vineyards. In the laboratory, *Veronica salicifolia* and *Fagopyrum esculentum* flowers significantly enhanced the fitness of females over *Achillea millefolium* and the water control. These findings together imply that superior and preferred floral resources are not yet available to adult *Lycaena salustius* in vineyard landscapes. The no-choice greenhouse experiment suggests that the plant group with which the butterfly may have co-evolved is more beneficial than other exotic species, and that such plants could enhance populations in vineyards. The conservation of other butterfly populations in farmland and other ecosystems may benefit from similar investigations.

**Keywords:** conservation; farmland; fecundity; flower preference; longevity

## Introduction

Nectarivorous butterfly species rely on the sugars and water in nectar for optimal longevity and reproduction (Baker & Baker 1975; Boggs 1997; Fischer & Fiedler 2001). In the field butterfly species vary widely, however, in their preferences for flower species (Watt et al. 1974; Faegri & van der Pijl 1979; May 1992), and in farmland conservation of butterflies, catering for this variation in nectar preferences has been attempted through a general approach, albeit with some positive results. 'Wild flower mixes' can increase populations of butterflies, bumblebees, and other invertebrates spatially and temporally (Feber et al. 1996), and conserving non-native species in field margins contributes to increasing abundance of butterfly species on farmland (Dover 1990, 1996, 1997; Feber et al. 1996). In other ecosystems, research to determine the importance of certain nectar sources for butterfly habitat conservation is based on observed feeding patterns in the field (Brakefield 1982; Thomas 1984; Tudor et al. 2004) or the effect of nectar availability on butterfly populations (Shepherd & Debinski 2005; Nelson & Wydoski 2008).

However, questions are rarely asked about whether flower species used in the field by adult butterflies are nutritious food sources or are only common but low-quality resources that are used in the absence of true optimal nectar sources. This issue has also been raised in biological control research; for example, Wäckers (2004) argued that for parasitoid wasps, a useful nectar supply is not always guaranteed by the mere presence of floral resources, and that flower screening is an important step in understanding foraging principles.

This study investigated the current floral preferences of the endemic New Zealand butterfly, the common copper *Lycaena*

*salustius* Fabricius (Lepidoptera: Lycaenidae) (hereafter *L. salustius*), as a representative member of the butterfly fauna. The study employed, for the first time on a butterfly species, a no-choice laboratory-style bioassay using flowers, as opposed to artificial nectar, in an attempt to identify superior nectar sources. This is particularly important in New Zealand where conservation of alien, nectar-rich, 'weed' species is unlikely to gain support from landowners. Identification of a native flowering plant species or group that may be beneficial to butterflies and which is acceptable as an addition to farmland could assist in the continued development of native planting schemes such as 'Greening Waipara', a Government-funded project led by Lincoln University and Landcare Research in the viticultural region of Waipara, North Canterbury. The project is encouraging and assisting participating vineyard owners to replant once-common native plant species in and around their properties, with an emphasis on providing multiple ecosystem services (Fiedler et al. 2008). The inclusion of a native nectar source of high quality for butterflies would provide the additional ecosystem service of butterfly conservation, which will contribute to vineyard marketing and regional tourism, and may also attract beneficial insects.

Field surveys initially sought to identify the preferred nectar sources of *L. salustius* on farmland. Two experiments, one in the field and one in the laboratory, then attempted to establish whether alternatives are preferred or impart greater fitness benefits. This involved no-choice laboratory-style flower 'screening', a research technique that has been used frequently in conservation biological control to assess the fitness benefits of different nectar sources to insect parasitoids and pest moths (Baggen & Gurr 1998; Wäckers 2004; Lavandero et al. 2006; Wade & Wratten 2007; Winkler et al. 2009) but has not been

used to test the benefits of flower species on butterfly fitness. It is expected that *L. salustius* will prefer and receive most benefit from suitable native nectar sources when available. Subsequently identifying superior floral species will help to inform conservation efforts and help to determine whether the butterfly is likely to benefit from farmland habitats, or is surviving only in substandard refuges.

## Materials and methods

### Study species

*Lycaena salustius* is endemic to New Zealand and is not an endangered species (Patrick & Dugdale 2000). The adults are relatively sedentary, have a wingspan of 24–33 mm, and are found in most areas of the country. There are two broods during early and late summer, depending on climate. Often associated with shrubland and coastal environments, its larval host plants consist of members of the genus *Muehlenbeckia* (Polygonaceae), particularly *M. complexa* and *M. australis* (Gibbs 1980)<sup>1</sup>. Apart from the early records (summarised by Gibbs (1980)) and observations of population dynamics by Flux (1968) and Craw (1975), there have been no published studies on the ecology of *L. salustius*. This species has been chosen for these experiments because it was prevalent in surveys (Gillespie & Wratten 2012) and was suitable for laboratory rearing, readily mating and ovipositing in caged environments.

### Field observations

We recorded nectar use by adult *L. salustius* in the summer of 2008/09, during transect survey work carried out using the standard method of the British butterfly monitoring scheme (Pollard & Yates 1993). In each of six vineyards in Waipara,

northern Canterbury, a fixed transect route was established, passing through different vegetation types in the vineyards. The total length of the six transects was 14 065 m and they were walked 13 times from October 2008 to April 2009, with intervals of 2–3 weeks between walks. For the nectar-use observations, each occasion an adult *L. salustius* was observed feeding was recorded, along with the flower species used. As some flowers are more abundant than others, however, a flower abundance scoring system was used also, to calculate a flower species preference ranking for adult butterflies. On most of the transect visits throughout the summer, the abundance of flowers of each species or group of species was scored for each section of transect passing through a different vegetation type, using the DAFOR estimating method (5 = dominant, 4 = abundant, 3 = frequent, 2 = occasional, 1 = rare) (Clausen et al. 2001). Numbers of inflorescences were estimated visually and a particular flower was considered dominant if more than 200 inflorescences were counted, abundant if 100–200 inflorescences were counted, frequent if 50–100 were counted, occasional if 20–50 were counted, and rare if counts were under 20 inflorescences. Nectar-source abundance categories were summed for each section of transect for an overall nectar abundance score. These scores were summed over the summer and a mean for each section calculated. The number of observations of butterfly feeding was then divided by this score to give a rank of preference for the flower species (Table 1).

### Field experiment

In the summer of 2009/10, a plot 15 × 8 m was marked out on one vineyard at the interface of the main *L. salustius* habitat (a small remnant of native vegetation) and the adjacent field margin of the vineyard. A rabbit-proof fence was built around the perimeter of the plot. A total of 32 plants, four each of eight species, were then planted within

<sup>1</sup> Plant names follow Allan Herbarium (2000)

**Table 1.** The 15 flower species on which *Lycaena salustius* was observed feeding throughout the summer survey of 2008/09, listed in descending order of observation frequency. Plant abundance scores are based on abundance categories averaged over the season and the flower rank is based on the number of feeding observations divided by the plant abundance score. Thus, the third most visited flower (*Geranium* spp.) is the top-ranking flower despite its relative scarcity, because of the large number of visits.

Species	Species name (Family) * indicates native plants	No. of feeding observations	Plant abundance score	Flower rank
Californian thistle	<i>Cirsium arvense</i> (L.) Scop. (Compositae)	120	2.91	4
Common yarrow	<i>Achillea millefolium</i> L. (Compositae)	59	1.89	5
Cranesbill species	<i>Geranium</i> L. spp. (Geraniaceae)	55	0.36	1
Sprawling wire-vine	* <i>Muehlenbeckia axillaris</i> (Hook.f.) Endl. (Polygonaceae)	48	0.44	2
Goosefoot species	<i>Chenopodium</i> L. sp. (Amaranthaceae)	28	0.39	3
White clover	<i>Trifolium repens</i> L. (Leguminosae)	24	4.83	10
Black medick	<i>Medicago lupulina</i> L. (Leguminosae)	24	1.30	6
Pōhuehue	* <i>Muehlenbeckia complexa</i> (A.Cunn.) Meisn. (Polygonaceae)	20	1.44	9
Hedge mustard	<i>Sisymbrium officinale</i> (L.) Scop. (Cruciferae)	13	0.73	7
Yellow Asteraceae	(= Compositae)	12	7.77	13
Viper's bugloss	<i>Echium vulgare</i> L. (Boraginaceae)	8	4.89	12
Lucerne	<i>Medicago sativa</i> L. (Leguminosae)	4	0.97	11
Large-flowered mallow	<i>Malva sylvestris</i> L. (Malvaceae)	4	4.32	14
Narrow-leaved vetch	<i>Vicia sativa</i> L. (Leguminosae)	2	0.12	8
Small-flowered mallow	<i>Malva parviflora</i> L. (Malvaceae)	1	2.30	15

the plot in a randomised block design, with each plant placed approximately 1.5 m from the edge of the plot and/or its nearest neighbour. The eight plant species were: *Veronica* 'Youngii' (sometimes known as *Veronica/Hebe* 'Carl Teschner') (Plantaginaceae), *Trifolium repens* (Leguminosae), *Fagopyrum esculentum* (Polygonaceae), *Muehlenbeckia complexa*, *Phacelia tanacetifolia* (Boraginaceae), *Achillea millefolium* (Compositae), *Chrysanthemum maximum* (Compositae) and *Leptinella minor* (Compositae). The plants were selected either because they were attractive to *L. salustius* adults in the previous field season and were relatively easy to grow in a nursery (*Trifolium repens*, *Muehlenbeckia complexa*, *Achillea millefolium*), because they were native species absent from Waipara but potentially attractive to butterflies (*Veronica* 'Youngii', *Leptinella minor*; M. Gillespie, pers. obs.), or because their presence would allow the testing of plant species prescribed to vineyards for conservation biological control of insect pests (*Fagopyrum esculentum*, *Phacelia tanacetifolia*; Berndt et al. 2006). Finally, *Chrysanthemum maximum* was chosen as a readily flowering last-minute replacement for *Cirsium arvense* (Asteraceae), which failed to flower at the time of proposed data collection. Seeds of *Chrysanthemum maximum*, *Fagopyrum esculentum* (cv. Katowase) and *Phacelia tanacetifolia* (cv. Balo) were obtained commercially from New Zealand suppliers: Kings Seeds, Katikati, Bay of Plenty (*C. maximum*); Midland Seeds, Ashburton, Canterbury (*F. esculentum*); and Kiwi Seeds, Blenheim (*P. tanacetifolia*). These seeds were sown in potting mix at the nursery at Lincoln University. All other plants were grown from Canterbury seed stock in potting mix either in the nursery at Lincoln University (*Trifolium repens*, *Achillea millefolium*), or by Hurunui Natives (*Veronica* 'Youngii', *Leptinella minor*, *Muehlenbeckia complexa*). Plants were transplanted in September 2009 to allow them to overcome root shock and reach flowering stages to coincide with peak butterfly populations. Plants were watered weekly as required.

Only four of the plant species (i.e. one a cultivar) were in flower at the time of data collection: *Veronica* 'Youngii', *Phacelia tanacetifolia*, *Trifolium repens*, and *Achillea millefolium*. The four remaining species were therefore replaced by potted plants of these species in flower. Pots were sunk into the ground so that the top of the pot was level with the soil surface and the soil covered the pots' rims. On 4 December 2009, the grass around the plants was mown at 0830 hours, to ensure a uniform sward and to remove other flowering species in and around the plot. From 0930 to 1200 hours, the plot was observed continuously from a distance of 3 m. The weather on this day was cloudless and still, with a mean morning air temperature of 25°C. When a wild *L. salustius* adult entered the plot, a stopwatch was started and the butterfly was followed until it began feeding on a plant species in the experiment. The duration of feeding activity, assessed through binoculars, was recorded for each individual flowering plant until the butterfly left the plot, at which point it was caught with a butterfly net and kept in a small, dark container so that it would play no further part in the experiment. All butterflies were subsequently released at the end of the day. The experiment was repeated 2 weeks later on 18 December. It is assumed that the adults involved in the second experiment were independent of the first as the 2-week interval is longer than the field lifespan of adult *L. salustius* (10.4 days; Craw 1975). In total, 36 butterflies were observed.

### Greenhouse bioassay

The effects of different nectar sources on adult female fitness as estimated by fecundity and longevity were investigated in a no-choice bioassay. Eight cages measuring  $0.8 \times 0.8 \times 0.8$  m and consisting of a metal frame and nylon mesh netting were erected and placed on a table in a greenhouse at the Lincoln University nursery. The greenhouse heating was controlled on a thermostat to prevent the temperature dropping below 15°C. No day-length or humidity controls were employed, but humidity varied between 49 and 73%. A length of black mesh was placed on the western half of the roof of each cage to provide periodic shade and prevent overheating. In each of the cages, a small potted plant of *Muehlenbeckia australis* was placed in a corner of the cage for oviposition. A small plastic vial filled with water and a dental wick protruding through a hole in the yellow lid was also placed in the middle of each cage on top of an upturned plant pot.

The eight cages were then randomly assigned to one of four treatments (two replicates at a time): *Achillea millefolium*, *Veronica salicifolia* G. Forst. (Scrophulariaceae), *Fagopyrum esculentum*, and a water control. These species were chosen because they were readily available at the time and represent the three groups of plants included in the field experiment (attractive to *L. salustius* in the field (*Achillea millefolium*), potentially attractive (*Veronica* spp.), potentially attractive and used for biocontrol (*Fagopyrum esculentum*)). For *Achillea millefolium* and *Fagopyrum esculentum*, potted flowering plants were grown in the nursery at Lincoln University from seeds obtained as above. However, *Veronica salicifolia* was too large for the small cages, so a flowering shoot was cut from a *V. salicifolia* bush in the nursery grounds, and placed in a plastic vial filled with water and held in place by a dental wick protruding through a hole in the lid. The flowering shoot was replaced daily and the vial was placed on an upturned flowerpot in the cage. This form of flower provision in cage experiments can act as a satisfactory substitute for intact inflorescences (Wade & Wratten 2007). For the water control, no flower was placed in the cage. All nectar treatments were placed in the unshaded corner of the cage opposite the oviposition treatment.

In addition to these plants, a potted *Fagopyrum esculentum* plant with flowers removed was placed in each of the cages. This was done because *L. salustius* females were observed laying a large number of eggs on *Fagopyrum esculentum* in a pilot study. As one of the treatments included a flowering *Fagopyrum esculentum*, the addition of a non-flowering individual to every cage was to account for the effect of this plant species on oviposition. There were therefore two plants acting as oviposition strata in each cage.

A newly eclosed, unfed and unmated female of laboratory-reared *L. salustius* was randomly assigned to each cage. Females were reared from eggs laid in the laboratory by wild-caught females from a colony near Lake Ellesmere (Te Waihora), Canterbury, an area local to Lincoln University and approximately 90 km from Waipara. Males caught from the same location were added to the cages (one per cage) to mate the females as required. The cages were revisited daily to check for survival, count the number of eggs on each plant, and water or replace plants where necessary. The males were removed from the cages when the females were observed to have begun to lay eggs. On the few occasions that this did not occur after 2 days, a new wild-caught male was introduced. The females were checked daily and the total number of eggs laid were counted when the female had died. Plants holding eggs were placed in the insectary at the Lincoln nursery and

checked regularly for egg hatching. When the females of all eight cages had died, the insects were dissected and the eggs remaining in the abdomen were counted under a  $\times 10$  binocular microscope. This number was added to the total score of eggs laid on the plants per butterfly to provide an estimate of 'potential fecundity'. The experiment was started in January 2010 and was repeated four times ( $n = 5$  cycles), with the final run ending in the second week of March 2010, although with only four cages on the last run (i.e. no duplication of treatments) ( $n = 9$  replicates per treatment in total). Unfortunately, records were not made of female body weight prior to the experiments, and as males were field-collected there was no way of knowing age or mating history. Therefore, results should be treated with caution, as these factors can affect fecundity and fertility in some species (Fischer & Fielder 2000; Gotthard et al. 2000).

### Statistical analysis

The data from the field experiment were analysed using the Friedman test, a non-parametric analogue of a two-way ANOVA that is used when there is a single observation for each factor combination. 'Flower species' was the grouping variable and 'Row' was the blocking variable. The response variables were frequency of flower visits to each individual flowering plant, and total time (in seconds) of feeding activity at each individual flowering plant. Only two females were recorded feeding in the field plot, so the sexes were not treated separately.

For the greenhouse bioassay, survival analysis was used to compare the effect of treatment on the lifespan (days) of butterflies. The Kaplan–Meier estimate of the survival function was calculated and Cox's Proportional Hazard Model (Afifi & Clark 1990) was used to compare the survival curves. Mean eggs laid, potential fecundity, reproductive period, and eggs laid per day were analysed using ANOVA after ensuring normality and homogeneity of variance, with cage number

and experiment run added as covariates. Pairwise comparisons were tested using Tukey's HSD. All analyses were performed in R version 2.9.2 (R Core Development Team, 2009).

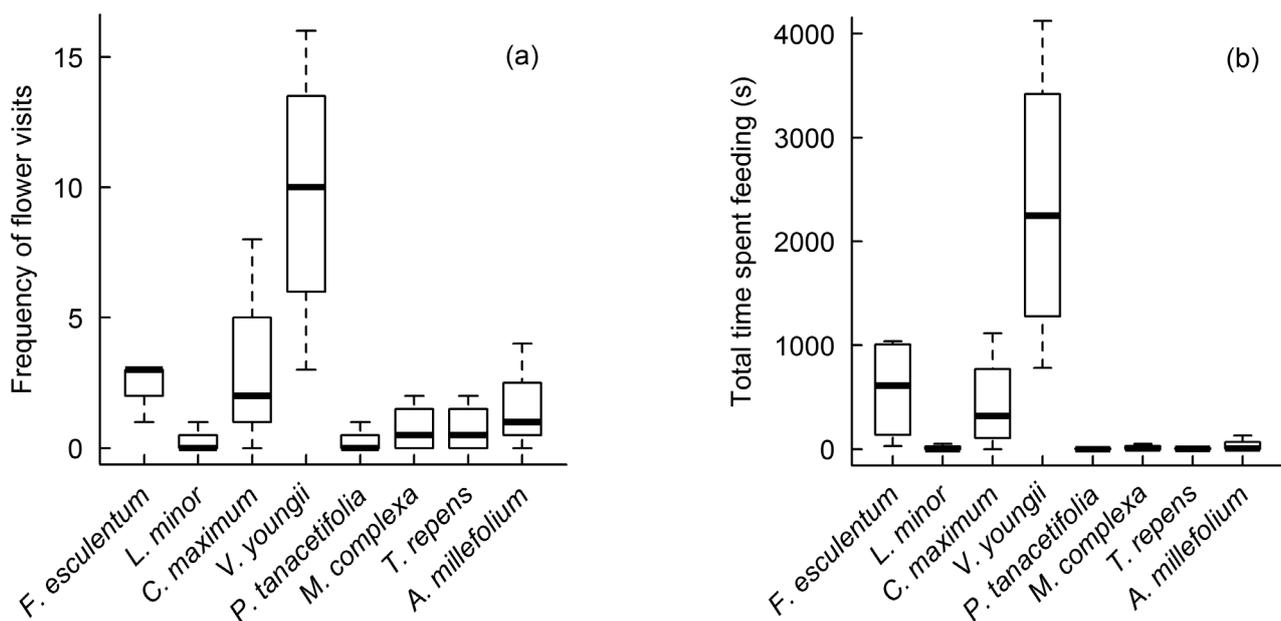
## Results

### Field observations

Feeding records from the 2008/09 summer season are summarised in Table 1. The most frequented species was *Cirsium arvense*, with *Achillea millefolium*, *Geranium* spp. and *Muehlenbeckia axillaris* also visited a large number of times. However, when feeding observations were divided by the abundance of flowers, the *Geranium* spp. were the most visited species, followed by *Muehlenbeckia axillaris*, *Chenopodium* sp., *Cirsium arvense* and *Achillea millefolium*. In total, *L. salustius* was observed visiting 15 different species.

### Field experiment

The explanatory variable 'flower species' was significant for both the frequency of visits (Friedman  $\chi^2 = 16.92$ , d.f. = 7,  $P = 0.018$ ) and total time spent feeding (Friedman  $\chi^2 = 17.59$ , d.f. = 7,  $P = 0.014$ ). Performing the same analysis with the explanatory variables reversed (i.e. fitting 'row' as grouping variable and 'flower species' as the blocking variable) showed that the effect of row was not significant (Frequency: Friedman  $\chi^2 = 1.7344$ , d.f. = 3,  $P = 0.6293$ ; Time: Friedman  $\chi^2 = 1.0714$ , d.f. = 3,  $P = 0.784$ ). Plots of the effects of flower species (Fig. 1) show that *Veronica* 'Youngii' was the most visited flower species, followed by *Fagopyrum esculentum*, *Chrysanthemum maximum* and *Achillea millefolium*. All species were visited at least once, but not all individual plants were visited.

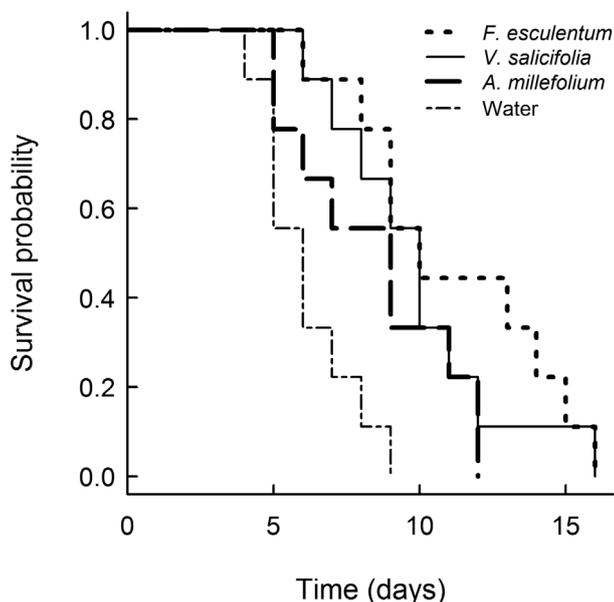


**Figure 1.** Field choice experiment results showing (a) the frequency of feeding visits of adult *Lycaena salustius* to different flowering species and (b) the total time spent by adult *L. salustius* feeding from the flowering species (*Fagopyrum esculentum*, *Leptinella minor*, *Chrysanthemum maximum*, *Veronica* 'Youngii', *Phacelia tanacetifolia*, *Muehlenbeckia complexa*, *Trifolium repens* and *Achillea millefolium*). The solid line in each box indicates the median, and the lower and upper edges of the box are the first and third quartiles. The whisker of the box reaches to the largest or smallest value.

### Adult longevity and fecundity

In all longevity and fecundity analyses the covariates ‘run’ and ‘cage’ were not significant and were removed as factors. Survival curves were significantly different between treatments (log-rank:  $P < 0.001$ , Wald test:  $P < 0.01$ ) (Fig. 2). Bonferroni-corrected paired comparisons using Cox’s proportional hazard model showed that *Fagopyrum esculentum* and *Veronica salicifolia* significantly ( $P < 0.01$ ) enhanced longevity of *L. salustius* females compared with water. There were no significant differences in longevity between the three flower species, but the longevity of females fed *Achillea millefolium* was not significantly different from those fed only water ( $P = 0.10$ ). Mean longevity and standard errors are shown in Table 2.

There was a significant effect of treatment on all measures of fecundity: eggs laid ( $F = 8.13$ , d.f. = 3,  $P < 0.001$ ; Fig. 3a), potential fecundity ( $F = 12.07$ , d.f. = 3,  $P < 0.0001$ ; Fig. 3b), reproductive period ( $F = 4.93$ , d.f. = 3,  $P < 0.01$ ; Fig. 3c) and eggs laid per day ( $F = 5.35$ , d.f. = 3,  $P < 0.01$ ; Fig. 3d). The number of eggs laid by females fed *Veronica salicifolia* did not differ from those fed *Fagopyrum esculentum*, but both laid significantly more eggs than those fed only water ( $P < 0.001$  and  $P < 0.01$  respectively) (Fig. 3a). In addition, *Veronica salicifolia* was superior to *Achillea millefolium* in enhancing the egg-laying capacity of females ( $P < 0.05$ ). The number of eggs laid by females feeding on *Achillea millefolium* did not differ from those fed only water ( $P = 0.35$ ) (Fig. 3a).



**Figure 2.** Kaplan-Meier estimates of the survivorship function of female *Lycaena salustius* when given different flower species (*Fagopyrum esculentum*, *Veronica salicifolia*, *Achillea millefolium*) or water.

**Table 2.** Mean longevity in days, and standard error of *Lycaena salustius* females given *Fagopyrum esculentum*, *Veronica salicifolia* and *Achillea millefolium* flowers or water in a greenhouse no-choice experiment. Treatments with the same letter did not differ significantly with Bonferroni-corrected paired comparisons using Cox’s proportional hazard model ( $P < 0.01$ ).

	<i>F. esculentum</i>	<i>V. salicifolia</i>	<i>A. millefolium</i>	Water
Mean time (days)	11.11 <sup>a</sup>	9.89 <sup>a</sup>	8.44 <sup>ab</sup>	6.11 <sup>b</sup>
Standard error	1.16	0.99	0.94	0.54

Potential fecundity (number of eggs laid + eggs in abdomen after death; Fig. 3b) showed the same patterns: females fed *Veronica salicifolia* had significantly higher potential fecundity than those fed water ( $P < 0.0001$ ) and *Achillea millefolium* ( $P < 0.01$ ) while adults fed *Fagopyrum esculentum* also had a higher potential fecundity than those fed water ( $P < 0.01$ ). For eggs laid per day, females fed *Veronica salicifolia* also differed significantly from those provided with water ( $P < 0.01$ ) and *Achillea millefolium* ( $P < 0.05$ ) (Fig. 3d). Only females fed *Fagopyrum esculentum* had a longer reproductive period than those fed water ( $P < 0.01$ ).

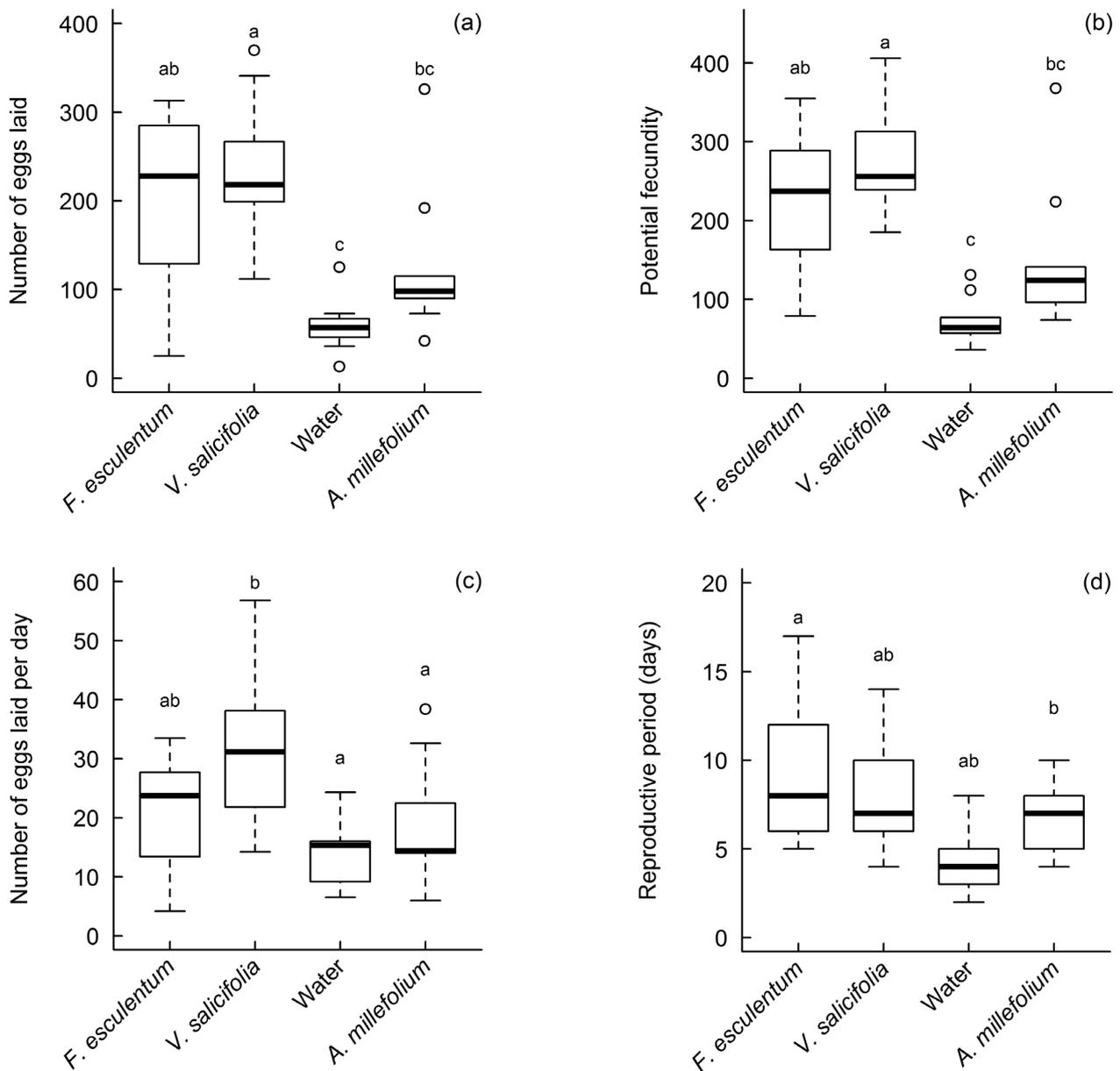
## Discussion

### Nectar availability and choice in Waipara vineyards

*Lycaena salustius* adults used a range of 15 flower species in vineyards, although only half of these were used 20 or more times. Only two species were native, *Muehlenbeckia axillaris* and *M. complexa*, with the remainder consisting of common introduced perennial ‘weed’ species as expected. The list in Table 1 is not an exhaustive account of the butterfly’s potential resources: Craw (1975) and Flux (1968) observed *L. salustius* feeding from a range of other species (e.g. *Parsonsia heterophylla*, *Metrosideros perforata*, *Nasturtium officinale*, *Rubus fruticosus*). Such a range of preferences suggests that *L. salustius* is an opportunist with respect to adult food sources, opting for those species that are abundant and accessible and feeding on different flower species at different sites (Shreeve 1992).

The preference for alternative nectar sources in the field experiment also suggests that if *L. salustius* adults always choose the most profitable source of nectar, high quality resources may not currently be present in some farmland habitats. While these results are not directly comparable to the laboratory results, similar findings from the no-choice experiment provide support to this suggestion. However, the field study results rely on a number of assumptions regarding the nature of the field choice experiment. Records of feeding in the plot are a ‘snapshot’ of a butterfly individual’s feeding history. A true, but impractical, assessment of preference would record feeding for the life of a number of butterfly individuals and include other aspects of nutrition such as larval feeding.

The preference for *Veronica* ‘Youngii’ may not be surprising though. *Veronica* species have been suggested to be favoured by native butterflies (G. W. Gibbs, pers. comm.) and a number of butterfly species have been seen feeding voraciously on *Veronica* spp. nectar in garden settings (M. Gillespie, pers. obs.), but this ‘attraction’ has never been quantified. *Fagopyrum esculentum*, also used in the trial plot, is a useful nectar source for beneficial insects such as the parasitoids and predators of agricultural invertebrate pests (Landis et al. 2000; Berndt & Wratten 2005; Berndt et al. 2006). The clear value of these



**Figure 3.** (a) Fecundity, (b) potential fecundity (number of eggs laid + unlaied eggs after death), (c) number of eggs laid per day, and (d) reproductive period of laboratory-reared female *Lycaena salustius* adults given different nectar sources (*Fagopyrum esculentum*, *Veronica salicifolia*, *Achillea millefolium*) and water. The solid line in each box indicates the median, and the lower and upper edges of the box are the first and third quartiles. The open circles refer to outliers, which are points greater than 1.5 times the interquartile range. The whisker of the box reaches to the largest or smallest value that is not an outlier. Treatments with the same letter did not differ significantly with Tukey's HSD ( $P < 0.05$ ).

species highlights the need to consider the possibility that high quality flower species may not be present, abundant or accessible in a habitat, despite observed patterns of preferential feeding on potentially substandard species.

#### Adult longevity and fecundity

The longevity bioassay results are comparable with those of mark–release–recapture field studies (Flux (1968): 8 days; Craw (1975): 10.4 days; this study: 6.1 to 11.1 days). However, the results of other fitness measures presented here should be considered with caution because the comparison of the different treatments relies on the assumption that the nectar quantity and quality of free-growing *Veronica salicifolia* were not affected

by cutting and presenting the excised inflorescences in water. While Wade and Wratten (2007) found that presentation method (excised or intact flowers) rarely affected the results of fitness assays, this was only for insect parasitoids and Begum et al. (2004) argued that direct comparison between the two methods was not strictly possible. Furthermore, fertility and fecundity among the Lepidoptera are known to be affected by morphological and phenological traits of both sexes such as body size, mating history, and age, and these effects vary with different mating systems (Fischer & Fiedler 2000; Gotthard et al. 2000; Jiménez-Pérez & Wang 2004; Calvo & Molina 2005; Trager & Daniels 2011). Therefore, as such variables were not recorded in our study, the results should be viewed

with caution, particularly without knowledge of the mating system of *L. salustius*.

Despite these shortcomings, it is possible to identify the clear benefit of both *Fagopyrum esculentum* and *Veronica salicifolia* to adult *L. salustius* compared with water. Due to the low fitness of *L. salustius* adults fed only water, the production of eggs in this species is evidently limited by a lack of nectar intake (Fischer & Fiedler 2001), and larval stores of these compounds were not enough to ensure optimal lifespan and high levels of egg production. Similarly, the composition of nectar of *A. millefolium* is apparently insufficient to overcome this limitation significantly, supporting the suggestion that this flower species may be a common and available food source used by *L. salustius* in the absence of superior resources.

Although the nectar composition is not known for the *Veronica* species used in this study, *V. stricta* produces large quantities of nectar (0.46 ul per day per flower for male flowers and 0.10 ul for female flowers) (Delph & Lively 1992). *Fagopyrum esculentum* is similarly a known source of high nectar quantities (0.17 ul per flower; Cawoy et al. 2008). Amino acid concentrations are not known for these two species, but *Achillea millefolium* does have a high concentration of these compounds (Baker & Baker 1973; Rathman et al. 1990). Flowers visited by dung- and carrion-feeding flies are the richest in amino acids (Baker & Baker 1973) and *Achillea millefolium* is particularly attractive to these fly species (Rathman et al. 1990). Plants primarily pollinated by butterflies are slightly less rich in amino acid sources than these fly-pollinated plants, however (Baker & Baker 1973). The amino acid concentrations of both *Veronica salicifolia* and *Fagopyrum esculentum* may therefore be more suited to *L. salustius* and butterflies in general, than that of *Achillea millefolium* although further study would help confirm this. Further study would similarly help to identify the importance of other flower selection criteria to *L. salustius* such as nectar quantity, flower colour, scent, size and morphology.

### Implications for conservation

A longer lifespan and a larger egg load resulting from feeding on *Veronica* species and *Fagopyrum esculentum* in the field may contribute to the conservation and enhancement of *L. salustius* by enhancing reproductive output considerably. The native *Veronica* spp. in particular may represent nectar sources with which the butterfly co-evolved, potentially highlighting its suitability as a group of plants to be added to farmland habitats in native planting programmes such as the Greening Waipara project. Additional ecosystem services such as the attraction of predators and parasitoids of pests may also result from such an addition.

Caution should be used in the interpretation of these results, as different plants were used in field and greenhouse trials, the number of replicates was low, and few of the plant species preferred by the butterfly in the field were subsequently used. However, it may be argued that even if *L. salustius* is not restricted by nectar provision per se in the field, it may be restricted by a lack of superior nectar sources in terms of fitness benefit. Many factors affect the choice of nectar source of butterflies in the field, such as weather and adult behaviour (Corbet 1978; May 1992), and may require further study in light of the current study. However, this work has a broader application. By investigating the effects of actual flower species on adult butterfly fitness, the results of the laboratory work suggest that the nectar sources selected by butterflies in a particular habitat may not be those that impart

the greatest fitness benefits. This is a finding that has also been reported for parasitoid wasps (Wäckers 2004). Similar work is therefore suggested for other species of butterfly in other modified habitats and such an approach would have potentially important connotations for habitat management.

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