

Influence of refuge availability on the oviposition strategy of anachoretic and non-anachoretic aphidophagous ladybirds

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INTRODUCTION & AIM

In nature, insects display different oviposition strategies that evolved to increase the reproductive success of species (e.g. Kindman and Dixon 1993; Gonzalez et al. 2023). Usually, insect females lay their eggs in clusters, on the surface of the host plants (e.g. Hemptinne and Dixon 2000; Agarwala and Yasuda 2001; Timms and Leather 2007). However, other insect species lay their eggs singly, concealed in the habitat on the underside of leaves, in crevices of the substrate or beneath undigested remnants of eaten aphids (e.g. Hemptinne and Dixon 2000; Agarwala and Yasuda 2001; Brown et al. 2004).

Most of the ladybird species can be roughly divided into anachoretic (single egg laying) and non-anachoretic (cluster egg laying) strategists and it seems that body size mirror oviposition strategy: larger ladybirds tend to lay eggs in clusters whereas smaller species typically laying eggs singly.

The aim of this study was to characterize the components of the oviposition strategies of the anachoretic *S. nubilus* and the non-anachoretic *C. undecimpunctata*, corresponding to small and single egg laying strategist with a bigger and clustering egg laying strategist, respectively: 1) daily fecundity, 2) effect of refuge availability on females' fecundity (hide/not hide; far/close to aphid colonies) and 3) daily pace of oviposition.

MATERIAL AND METHODS

- Experiment 1** • Availability of refuge sites on fecundity of *S. nubilus* and *C. undecimpunctata* females: This experiment comprises five treatments in which five different experimental set-up devices were offered to ovipositing females of *S. nubilus* and *C. undecimpunctata* (Fig. 1).
- Experiment 2** • The effect of the availability of refuge (concealed sites) on females' fecundity: Three treatments corresponding to different conditions that females may find in nature: i) plant infested with fresh prey (IP); ii) plant infested with fresh prey and prey carcasses (IPC); iii) plant infested with fresh prey and three pieces of bark with 5 cm length (IPB) (Fig. 2).
- Experiment 3** • Daily pace of oviposition of *S. nubilus* vs *C. undecimpunctata* females: A 15-day old ovipositing female was released in a petri dish containing the preferred set-up device for oviposition. The laid eggs were counted hourly, from 7 a.m. to 6 p.m., and an additional count at 10 p.m. Egg counts carried out between 8 a.m. and 6 p.m. were labeled "early" and the remaining as "late".

Fig.1 Experimental set-up devices

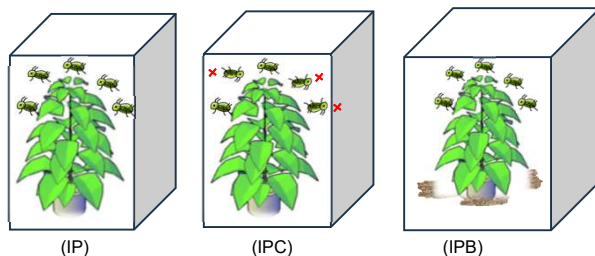
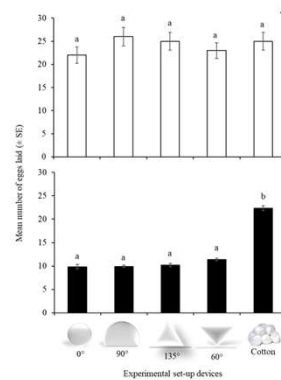


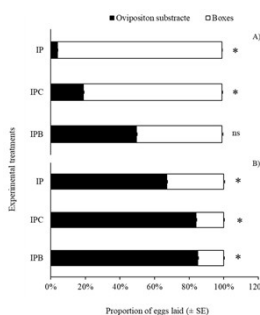
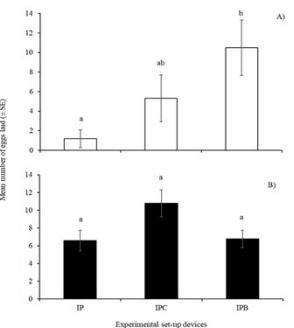
Fig.2

RESULTS & DISCUSSION



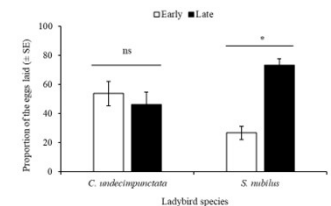
The average number of eggs laid by *C. undecimpunctata* females ranged from 21 to 26 and did not significantly differ according to the experimental set-up devices (Anova: $F(4, 295) = 1.226, p = 0.3$) (Fig. 1A). For *S. nubilus* females, daily fecundity ranged from 10 to 22 and only differed when the experimental set-up device was cotton ($\chi^2 = 134.278, df = 4, p \leq 0.001$) (Fig. 1B).

Fecundity by *C. undecimpunctata* significantly differed between treatments ($\chi^2 = 8.125, df = 2, p = 0.017$), being lower when the set-up device included a plant infested with fresh prey (IP) (Fig. 2A). For *S. nubilus*, no significant differences were found ($\chi^2 = 4.522, df = 2, p = 0.104$) (Fig. 2B).



Except for IPB device ($\chi^2 = 0, df = 1, p = 1$), *C. undecimpunctata* laid a higher proportion of eggs on the acrylic boxes, away from the aphid colony (IP: $\chi^2 = 53.6783, df = 1, p < 0.00001$; IPC: $\chi^2 = 21.2634, df = 1, p \leq 0.00001$; Fig. 3A). *Scymnus nubilus* laid a higher proportion of eggs close to or on the devices, near to the aphid colony (IP: $\chi^2 = 5.952, df = 1, p = 0.0147$; IPC: $\chi^2 = 27.9202, df = 1, p < 0.00001$; IPB: $\chi^2 = 27.9202, df = 1, p \leq 0.00001$; Fig. 3B).

We found a tendency for oviposition events to occur during scotophase, significantly for *S. nubilus* ($\chi^2 = 61.2117, df = 1, p < 0.00001$), but not for *C. undecimpunctata* ($\chi^2 = 3.3543, df = 1, p = 0.067$) (Fig. 4).



CONCLUSION

Our results showed that the smaller anachoretic ladybird (*S. nubilus*) is more dependent than the larger non-anachoretic ladybird (*C. undecimpunctata*) on habitat structural complexity to maximize its fitness and that laying a single hidden egg mainly during the scotophase confers a potential adaptive advantage to increase fitness.