Egg laying site preferences in *Pterostichus melanarius* Illiger (Coleoptera: Carabidae)

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In the case of ground beetles, the number of studies investigating the effects plant-related structure and microclimate on the selection of egg laying sites is very limited. The egg laying site preference of *Pterostichus melanarius*, an important carabid beetle in agricultural fields, was studied under laboratory conditions. The effects of wet/dry substrate, light/shadow and structured/unstructured environment on the number of eggs laid were investigated, as well as the influence of the presence of barley and Brussels sprout plants. We found that there was a strong influence of plant structure and microclimate on the selection of egg laying sites. Results showed a higher numbers of eggs laid in barley compared to Brussels sprouts. These results were supported by a significant preference for moist, shadowed, structurally complex environments as egg laying sites. Results indicate that vegetation characteristics in themselves may influence egg laying site preferences, beside the availability of prey for the adults and larvae, in the different plant systems.

Keywords: egg laying, intercropping, plant structure, *Pterostichus melanarius*

Studying the stimuli that modify oviposition might enable us to manipulate the distribution of the eggs of beneficial predators (Boldyrev *et al.* 1969). Carabid beetles are considered to be an important group of generalist predators in agricultural fields. Their biology and distribution are widely studied but surprisingly little is known about their reproduction, with regards to the selection of egg laying sites.

The most studied aspects of the reproduction of beneficial insects are the effects of food quantity and quality on fecundity (Sunderland *et al.* 1996). In the case of carabids, some information is available also on the relationship between the characteristics of the substrate and oviposition (Thiele 1977, Van Dijk & Den Boer 1992). Generally, however, the reasons why a given site is preferred for oviposition by carabids are not understood (Powell & Ashby 1995).

The results presented here are parts of a series of experiments that are investigating the relationship between intercropping and an important predatory carabid beetle, *Pterostichus melanarius*. In preliminary studies more *P. melanarius* larvae were trapped in Brussels sprout intercropped with barley than in Brussels sprout alone. To explain these results, experiments were designed to investigate egg laying site preferences of the species. Throughout these experiments, food quantity and quality in the choice situations were kept equal so that site selection depended only on microclimatic and structural conditions.

MATERIAL AND METHODS

Two sets of experiments were carried out to investigate oviposition site preferences. In the first set of experiments, different environmental variables were tested, in the second set of experiments live plants were used. Both sets of experiments were realised at the Department of Crop and Weed Ecology, Wageningen University, The Netherlands, in 2002 (first set) and 2003 (second set).

The first set of experiments was carried out in 1-m^2 arenas, in a climate chamber. Climate chamber settings followed the environmental conditions of mid-August in The Netherlands, the peak reproduction period of the species in this region (L:D = 14.5:9.5, 20°C in the light period, 16°C in the dark period, 70% RH in the light period, 95% RH in the dark period). Arenas contained sand, in an evenly laid, 1.5 cm layer.

Arenas were divided into two equal areas. Treatments were applied on these areas as follows:

- dry substrate (no water) against wet substrate (watered twice a day, each time with 1 litre of water sprayed evenly over the treatment area);

- shadow (a cardboard sheet placed in the way of light, 0.5 m above the area) against no shadow;
- structure (all together 200 pieces of wire fixed to the bottom of the area in a uniform distribution) against no structure.

For each of these pairs, two arenas were assigned (two replicates in space). Two arenas were assigned for procedure control, these arenas received 2 litre water twice a day, were not shadowed and contained no wires. 5 female and 5 male beetles were released in the middle of each arena, and left in the arena for a week. Beetles were fed daily with *ad libitum* amount of cat food placed in the middle point of the arena on a Petri dish lid, and were watered twice a day with 2 litre water sprayed evenly over the whole arena, unless specified otherwise in the treatments. After one week the beetles were removed and the eggs laid were washed out of the sand. The procedure was repeated over 5 weeks (five replicates in time). Each beetle was used only once. Beetles were trapped in an alfalfa field in dry pitfall traps and were kept in captivity, with *ad libitum* food supply of catfood, for a minimum of 2 weeks prior to the experiments.

The second set of experiments was carried out in 40 x 70 x 90 cm plastic boxes, in a climate chamber with the same settings as in the first set of experiments. The bottom of the boxes was lined with potting compost. Boxes were divided into 2 equal areas. Into one area, one 6-week-old Brussels sprout plant was planted, in the middle. Into the other area, 20 barley plants, 12 weeks old, were planted in a 4 x 5 grid. Twelve-week-old barley plants represented the age of plants in the field in mid-August. Twelve-week-old Brussels sprout plants, on the other hand, were too big for experimental purposes. Two days after planting, the compost was sealed with a sheet of textile, in which holes were made for the plants, the digging of beetles by plant stems was prevented by adhesive tape. On the surface of the textile, sand was distributed in an even, 1.5 cm layer.

Three boxes of the given parameters were used (three replicates in time). Five female and five male beetles were released in the middle of each arena, and left in the arena for a week. After one week the beetles were removed and the eggs laid were washed out of the sand. After each experimental week, new plants were planted in the boxes. The experiment was repeated three times (three replicates in time), one beetle was used only once. Beetles were trapped in winter wheat, in dry pitfall traps and were kept in captivity, with *ad libitum* food supply of cat food, for a minimum of two weeks prior to experiments. Feeding in the boxes was the same as in the first set of experiments, the boxes were watered twice a day, with 1.5 litre water sprayed evenly over the surface.

After counting the eggs, egg numbers laid within treatments were summarised, the percentages of eggs laid in the different treatments were calculated and the Chi^2 test was applied.

RESULTS

In the first set of experiments, significantly more eggs were laid in wet substrate than in dry substrate (DF=1, $p<0.05$, $\chi^2=59.063$) (Fig. 1a.); in shadow than in normal light (DF=1, $p<0.05$, $\chi^2=49.648$) (Fig. 1b.); and in structured environment than in unstructured environment ($p<0.05$, $\chi^2=75.313$) (Fig. 1c.). In the procedure control beetles laid similar numbers of eggs in the two areas of the arenas (DF=1, $p>0.05$, $\chi^2=4.84$).

In the second set of experiments, beetles laid significantly more eggs in areas planted with barley than in areas planted with Brussels sprouts ($p<0.05$, $\chi^2=75.469$) (Fig. 2.)

DISCUSSION

When vegetation diversity is increased in an agricultural area, usually the number of species/individuals of trapped ground beetles also increases (Kromp 1999). Some authors suspect the beneficial effect of increased plant diversity on the reproduction of beetles; papers usually suggest higher fecundity of beetles due to better food quality (Lys & Nentwig 1992, Zangger *et al.* 1994, Bommarco 1998).

According to the results presented here, plants may influence the choice for egg laying sites in the case of *P. melanarius*, through defining moisture levels of the substrate, light conditions and the structure of the environment.

The influence of the soil moisture on the number of eggs laid by *P. versicolor* and *Calathus melanocephalus* was mentioned by Van Dijk & Den Boer (1992), Aukema (1991) also suspected an effect of substrate moisture on egg laying in *C. melanocephalus*. Wet soil may not only prevent the desiccation of eggs and young larvae but may also indicate favourable conditions for prey items for larvae.

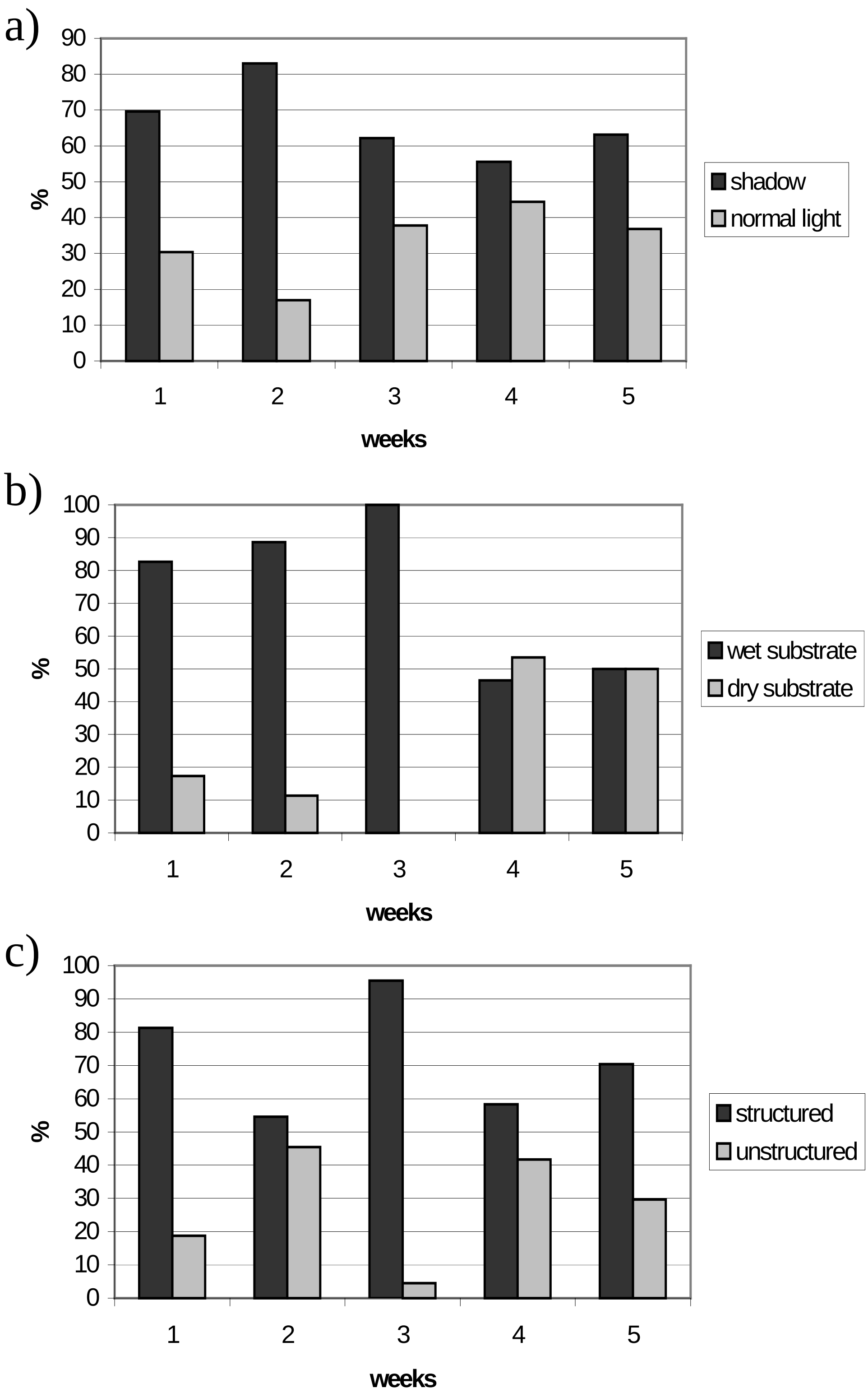

Figure 1. Percentages of eggs laid by *P. melanarius* in the first set of experiments in a) wet and dry substrate b) in shadow and normal light conditions and c) in structured and unstructured environment.

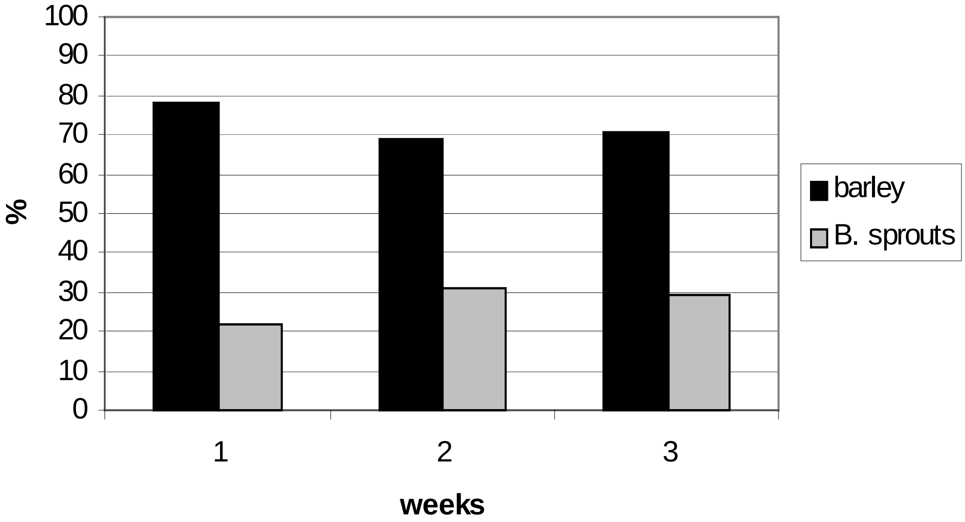

Figure 2. Percentages of eggs laid by *P. melanarius* in the different treatments in the second set of experiments.

The effect of shadow on egg laying site preference is surprising, because *P. melanarius* is mostly nocturnal and it might be presumed that it lays eggs during its main activity period, when light conditions should not influence egg laying site preferences. According to personal observations, beetles were not active during the light period throughout the experiments. Also, exposure to direct light in this case did not mean automatically the faster desiccation of the substrate.

Another surprising result is the beetles' choice for the sites 'structured' with wires. Wires did not prevent the beetles in moving freely, it is highly improbable that they laid eggs around wires because they were 'trapped' in the structured environment. Neither could provide the wires any chemical cues that might have directed the beetles' orientation. It is possible that the vertical structure of wires imitated plant stems for the beetles.

The results above suggest a preference for moist, dark and structurally complex sites for egg laying. Experiments with plants supported these findings as beetles preferred areas with barley plants to lay eggs to Brussels sprout plants that did not represent the same shadowed conditions and structural complexity as barley.

These experiments, though prove that *P. melanarius* can and will chose between sites for oviposition, give no information on how big a spatial scale this selection might work in the field. Nevertheless, results indicate that intercropping sparse vegetable cultures may improve the conditions for reproduction for this species. Results also suggest that beetles might leave vegetable cultures for oviposition if a more preferable environment is located nearby. This may have an influence on predator activity in a field for a given year as well as for the next.

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