## Aggregation behaviour of Harmonia axyridis

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The multicoloured Asian ladybeetle, *Harmonia axyridis* (Coleoptera: Coccinellidae), forms large aggregations inside dwellings to survive cold winters. The species' migratory flight is well documented. *Harmonia axyridis* preferentially moves towards prominent and visually contrasting elements (Obata, 1986; Nalepa *et al.*, 2005). However, the mechanisms involved in the selection of aggregation sites are misunderstood. The work presented here was devoted to the study of chemicals used by *H. axyridis* during its aggregation process.

During sampling of infested dwellings, non-volatile compounds were collected from aggregation sites. Gas chromatrography-mass spectometry (GC-MS) analyses showed that the chemicals found on the substrate where the beetles aggregate were different from the ones collected around these aggregation sites. These two blends are made up of the same long-chain hydrocarbons, comprising saturated and unsaturated homologues, but they are quantitatively different, the blend collected directly on the overwintering sites containing a higher proportion of saturated compounds than the other one.

Behavioural experiments, involving overwintering *H. axyridis* individuals, were then conducted in the laboratory to understand the roles of these chemicals in aggregation. Firstly, an aggregation assay using the blend collected inside overwintering sites showed a clear preference of ladybeetles for areas containing these compounds, highlighting the retention capacity of the blend on *H. axyridis*. On the other hand, a Y-shaped tube assay, using the chemical blend found around the sites, showed that those compounds are used by male and female congeners as cues, allowing individuals to orientate towards the side of the set-up containing the tested chemicals. These results suggest the use of two different area markings by *H. axyridis* during its aggregation: the first one to lead congeners towards aggregation sites, and the second to ensure the cohesion of the cluster.

Additional investigations were conducted to study the influence of (1) the presence of congeners and (2) the shelters' luminosity on the *H. axyridis* decision to settle and aggregate under shelters. A binary choice experiment conducted in the laboratory under non-wintering conditions showed that the multicoloured Asian ladybeetles present a permanent aggregative behaviour, as a result of the existence of social interactions. These experiments also highlighted the clear preference of *H. axyridis* for dark shelters.

## References

- Nalepa, C. A., Kennedy, G. G. & Brownie, C. 2005: Role of visual contrast in the alighting behavior of *Harmonia axyridis* (Coleoptera: Coccinellidae) at overwintering sites. Environ. Entomol. 34: 425-431.
- Obata, S. 1986: Determination of hibernation site in the ladybird beetle, *Harmonia axyridis* Pallas (Coleoptera, Coccinellidae). Kontyu 54: 218-223.