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## The biological control of *Aulacorthum solani* (Kaltenbach) (Homoptera: Aphididae) in greenhouse grown pepper; research on a tri-trophic system

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THE BIOLOGICAL CONTROL OF *Aulacorthum solani* (KALTENBACH) (HOMOPTERA: APHIDIDAE) IN GREENHOUSE GROWN PEPPER; RESEARCH ON A TRI-TROPHIC SYSTEM

J.V. SCHELT & F. WÄCKERS



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To improve the control of the foxglove aphid (*Aulacorthum solani*) in greenhouse sweet pepper, several trials on three trophic levels have been conducted. Two different lines of *Aphelinus abdominalis* were compared on flight capacity, parasitism and predation. One line was significantly better in parasitism of *A. solani* (6.3 vs 0.2 mummies/female/day). This line performed also better at lower temperatures (15-18° C.) in flight capacity. Predation was the same for both lines (2 aphids/female/day).

The honeydew of *A. solani* was tested as a food source on *Aphidius* spp. The life span of *Aphidius* on this honeydew was 4 days which was equal to water and half of sucrose. On 100 ha. of commercially grown sweet pepper a banker plant system (wheat, *Sitobion avena*, *A. abdominalis*) was used. In general growers were able to reduce the number of chemical corrections from 7 to around 3.

In sweet peppers grown under glass there is a natural oscillation of 4-5 weeks in flowering and fruit set. We observed large differences in aphid growth depending on this vegetative or fruiting phase. The practical considerations how to adjust the biological control still has to be investigated.

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## INTRODUCTION

The integrated control of aphids in glasshouses has become common practice in tomatoes, peppers, cucumbers, and aubergines. (VAN SCHELT 1999; KLAPWIJK 1999; MULDER et al. 1999). *Aphis gossypii* and *Myzus* spp. are controlled with *Aphidius colemani*; *Macrosiphum euphorbiae* with *Aphidius ervi*. Often generalist predators like *Aphidoletes aphidimyza* and lady beetles are used in conjunction with the parasitoids to control high aphid numbers. However the control of *Aulacorthum solani* in peppers is still very problematic.

*A. solani* has become a very common pest in sweet pepper in greenhouses over the last three

years. Because *A. solani* is a very polyphagous and a cosmopolitan species, it has become a problem in most pepper growing regions in North West Europe and Canada. The aphids are generally found in the lower parts of the plant and are easily overlooked at the beginning of an infestation. Already at low densities the plants can react very strongly to the saliva of the aphid. On the leaves yellow necrotic spots can be found and often the top is showing malformations. In the period 1997-2000 biological control with *A. ervi* and the gall midge *A. aphidimyza* was practised on several hundreds of hectares by commercial growers in the Netherlands. Results were unsatisfactory because too often chemical corrections with Pirimicarb had to be applied.

The low percentage parasitism may be explained by the foraging behaviour of this parasite (SCHWORER & VÖLKL 2001). If within a certain time no aphids are found than *A. ervi* has the tendency to disperse over large distances.

The potential use of another parasite, *Aphelinus abdominalis*, was explored. Two lines were compared on their dispersal capacity and fecundity. The use of banker plant systems (winter wheat with *Sitobion avenae*) was developed to enhance the numbers of *A. abdominalis* before aphid growth in the crop.

Research on the aphid itself was carried out by NIOO-CTE. We speculated that the aphid's honeydew could be toxic or unsuitable as a food source for the beneficials used. Finally we looked more in detail at the physiological status of the plant and its influence on aphid growth.

#### SELECTING LINES OF *A. abdominalis*

##### Introduction

Selecting of lines within a species can be an option to improve the control capacity of beneficials.

*Aphelinus abdominalis* is normally associated with *Macrosiphum euphorbiae* and cereal aphids as *Sitobion avenae*. HÖLLER & HAARDT (1993) compared a uniparental German line and a biparental French line. Both lines showed a high fecundity on *S. avenae* in the laboratory but failed in the field. Both lines however had similar biological characteristics, but were only tested on *S. avenae*.

In this study we compared two lines of *A. abdominalis*. One line came from a German producer, the other line was provided by INRA/Antibes (France). Both lines were reared separately on *Macrosiphum euphorbiae*.

They were compared with respect to their host feeding behaviour and their fecundity with *A. solani* as a host.

Because there is a tendency to save energy in greenhouse systems by reducing temperature in the winter, the dispersal capacity of beneficials under cool conditions (15-18 °C) is important. Moreover if the leaves of the plants are not touching each other in the beginning of the season

the beneficials have to fly for an optimal dispersal in the greenhouse. Flight propensity was assessed at different temperatures for both lines.

##### Material and Methods

The parasitism and predation rate was determined by introducing individual females of *A. abdominalis* (n=30) for 24 hours on a sweet pepper leaf disc on agar with 20 first and second instar *A. solani*. After 24 hours the female was removed and the number of aphids that were fed upon by the female, was determined. Because of slow deterioration of the leaf, the aphids were transferred to a fresh leaf after 8 days. After fourteen days the number of parasitized aphids (mummies) was assessed.

The French line was tested again on *A. solani* after being reared for one generation on this host.

The experiments were conducted in a climate cell at 21 ± 1°C, 70% RH, 16L:8D.

Flight propensity was determined by putting 100 adult wasps, less than 24 hrs old, in an open 50 ml. plastic bottle. The bottle was placed on a small concrete platform in the middle of a bowl (Ø 25 cm.) with water containing a drop of detergent.

This set up was put in a cage of 30 by 30 cm. and 50 cm. in height. The opening of the bottle was 6.5 cm. above the surface of the water. The cage was put in a climate box with fluorescent tubes at all sides. Experiments were done at 15, 18, 21, 24 and 27 °C and 75% RH. After 24 hours the number of parasitic wasps that remained in the bottle, that had drowned, and that had crossed the water were counted. Before releasing the parasitic wasps, they were acclimatised for one hour to the ambient temperature. All tests were done twice.

##### Results

###### *Parasitism and Predation*

Amount of host feeding was 2.33 aphid/day for the French line, 2.03 for the German line (n.s. MWU-test).

Parasitization was significantly different: 6.3 mummie/female for the French line, 0.23 for the German line (MWU, P<0.001)

No differences could be seen between the parasitisation capacity of the French line that had been reared on *M. euphorbiae* (6.3 mummie/female) and parasites of the same line that had been reared for one generation on *A. solani* (7.7 mummie/female) (n.s. MWU).

#### Flight capacity

The lines also differed clearly with respect to their flight capacity. The number of wasps from the German line, that left the bottle was significantly lower at 15 and 18°C. (MWU-test) (Fig. 1) In the numbers that actually flew across the water the same pattern was visible (Fig. 2).

The flight-pattern could be well observed in this set-up. When *A. abdominalis* are taking off, they jump up a little, and subsequently lose altitude very quickly before regaining height again. Only when they did not hit the water at the lowest point, could they reach the other side of the barrier.

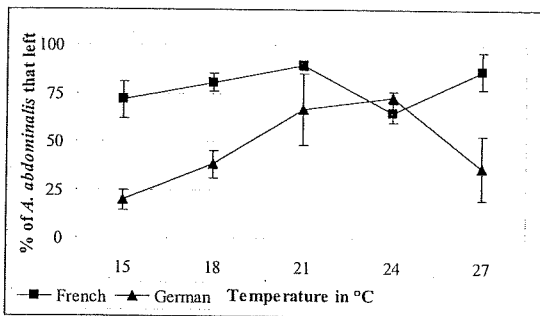


Fig. 1. Percentage adult *A. abdominalis* that left the the bottle at different temperatures.

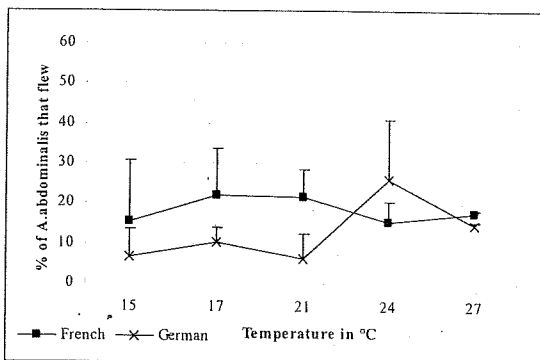


Fig. 2. Percentage adult *A. abdominalis* that flew across the waterbarrier.

#### Discussion

From these results it was concluded to use the French strain of *A. abdominalis* in the mass rearing and for commercial use. In theory the German line could be improved by rearing them for several generations on *A. solani*, but the offspring of the first experiment was so low, that further efforts in this direction were stopped. Also *M. euphorbiae* and not *A. solani* is the preferred aphid for a mass rearing system.

It can be speculated that a further exploration of *A. abdominalis* coming from other parts of its distribution area can be interesting.

#### THE SUITABILITY OF *A. solani* HONEYDEW AS A FOOD SOURCE FOR *Aphidius* spp.

##### Introduction

The great majority of parasitoids and many arthropod predators depend on sugar sources to cover their energetic needs. There is strong theoretical as well as empirical evidence that the availability of suitable sugar sources can be a key factor determining the population dynamics of predator-prey and parasitoid-host systems (KRIVAN & SIROT 1997; WÄCKERS 2003). Besides (extra) floral nectar, honeydew is the most prevalent source of exogenous sugars in nature. Due to the fact that agricultural ecosystems often lack flowering plants, honeydew is likely of particular importance in agriculture. A recent study by Wäckers and Steppuhn (unpublished) demonstrated that 80% of the larval parasitoid *Cotesia glomerata* collected in a cabbage field contained honeydew specific sugars, indicating a high incidence of honeydew feeding by this parasitoid. Parasitoids of honeydew-producing insects are believed to be even more intimately linked to this food source.

Despite this intimate link, honeydew can vary considerably with respect to its nutritional quality. Whilst certain types of honeydew can be equally suitable as nectar or sugar solutions, others are clearly inferior or even toxic (WÄCKERS 2000). To test the suitability of *A. solani* honeydew from various *A. solani*-plant

combinations for aphid parasitoids, we compared the longevity of honeydew-fed *A. colemani*, to parasitoids fed with a sucrose solution. Though *A. colemani* is not a natural parasite of *A. solani* it served as a model for aphid parasitoids in general and *Aphidius* spp. in particular.

#### Materials and method

*Aulacorthum solani* honeydew was collected from aphid colonies feeding on the following plant species: *Brassica nigra*, *Capsicum frutescens*, *Gossypium herbaceum*, and *Vicia sativa*. The plants had been grown in 1l. potting soil in greenhouses at the Dutch Institute for Ecology (NIOO-CTE) in Heteren. Growing conditions were T=20°C, RH=50-80%, 16L8D.

The homopteran-plant combinations were kept in fine-mesh screen cages to prevent contamination. To collect the honeydew, a glass plate was placed underneath the plant. After 24 hours the honeydew was collected using a glass micropipette and subsequently stored in a freezer at -15°C.

Newly emerged parasitoids were placed in a petridish and provided with a piece of water-soaked cotton wool as well as a 1µl droplet of one of the honeydew types placed on the lid. As

controls, separate *A. colemani* cohorts were subjected to water only and a 1µl droplet of a 1M sucrose solution respectively. The petridishes were kept at T=20°C, RH=95-100%, 16L8D. The high humidity prevented the honeydew and sucrose solution from becoming too viscous.

Survival of wasps was scored daily. Every second day the food droplets were renewed. For each treatment we tested 50-60 individuals. Parasitoids that were found dead in the honeydew droplets were discarded.

We used HPLC (High Performance Liquid Chromatography) to analyse the sugar composition of the honeydew types used in the longevity experiment

#### Results

##### Longevity

Parasitoids provided with water only (control) lived 4.0 days on average. Access to the sucrose solution increased parasitoid lifespan more than twofold (8.4 days). The honeydew, on the other hand, had only a marginal effect on parasitoid longevity (Fig 3). Only honeydew from *A. solani* on *V. sativa* raised parasitoid longevity relative to the water control (Mann-Whitney U-test).

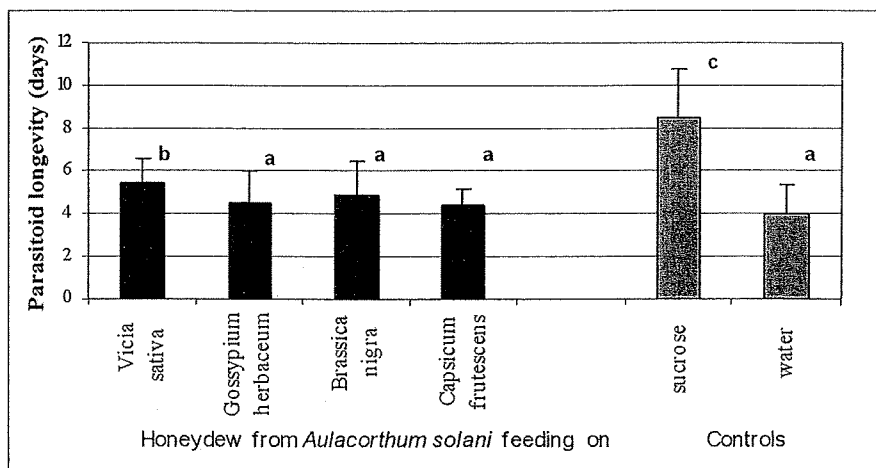


Fig. 3. Longevity of *A. colemani* when provided with various sugar sources, or water only (control). Different letters indicate significant differences among treatments (MWU-test).

##### Honeydew Sugar Composition

The HPLC sugar analysis showed that the composition of *A. solani* honeydew depends on

the aphid's host plant (Table 1). Most notably, the aphid synthesized sugar erlose was absent in honeydew from *G. herbaceum*, while occurring in

substantial amounts in honeydew collected from the other three plants. Maltose was only found in the honeydew collected from *Vicia faba*. Overall, however, the honeydew was dominated by sucrose and its hexose components glucose and fructose.

Table 1  
Sugar composition of honeydew collected from various *Aulacorthum solani*-plant combinations. Numbers represent percentage of total sugar content (weight/weight).

	Sucrose	Glucose	Fructose	Unidentified	Erllose	Trehalose	Maltose	Sorbitol	Mannitol
<i>Capsicum frutescens</i>	46.9	12.0	22.5	1.4	15.6	1.1	0.0	0.0	0.4
<i>Gossypium herbaceum</i>	35.5	19.4	39.5	2.0	0.0	0.0	0.0	1.3	2.3
<i>Brassica nigra</i>	35.0	15.2	27.7	3.3	16.7	0.8	0.0	0.0	1.4
<i>Vicia sativa</i>	26.8	19.7	30.5	2.1	11.5	0.0	7.7	0.8	0.9

## Discussion

Based on the fact that honeydew is primarily a sugar solution, it is often assumed that it makes for a suitable insect food source. However, the data presented here, as well as previous reported studies show that there can be a considerable variation in honeydew suitability. It is yet unclear which honeydew components are responsible for this variation, but two possibilities have been proposed: (i) secondary plant compounds and (ii) sap-feeder synthesized sugars. It has been reported that secondary plant compounds appear in honeydew (e.g. MALCOLM 1990). However, little is known about the effects of these compounds on honeydew-feeding insects. While secondary plant compounds in the honeydew might have been responsible for the poor suitability of *A. solani* honeydew, it would be remarkable that the effect on the parasitoid would be similar in the four plants tested, as they represent a wide variation in plant secondary chemistry.

The few studies addressing the effect of sap-feeder synthesized sugars indicate that these specific sugars might have a negative effect on insect longevity (WÄCKERS 2000). ZOEBELEIN (1956) reported that melezitose feeding reduced the life span of the parasitoid *Microplectron uscipennis* in comparison to food-deprived individuals, while sucrose, glucose or fructose increased *M. uscipennis* longevity by a factor of more than 2. In studies with *C. glomerata*, melezitose prolonged the parasitoid's life span relative to control individuals provided with water only, but reduced longevity by 44-47% in comparison to sucrose, fructose and glucose. Of

the other honeydew oligosaccharides studied, erlose was moderately suitable as a food source, while trehalose and raffinose were unsuitable (WÄCKERS 2001). Similar results were obtained in sugar longevity studies for *A. colemani* (Wäckers, unpublished). However, given the relatively low concentration of these sugars in the honeydew tested here, it is unlikely that the sugar composition is responsible for the poor survival of *A. colemani* in this study.

Irrespective of the underlying mechanism, the poor quality of *A. solani* honeydew may have considerable ecological and applied implications. The poor performance of *A. ervi* in controlling *A. solani* in the glasshouse may be partly due to the limited survival of *Aphidius* spp. on *A. solani* honeydew.

## THE USE OF *A. abdominalis* ON BANKER PLANTS

In order to obtain large numbers of *A. abdominalis* in the greenhouse at the right time, a banker plant system was developed. Winter wheat (cv. "Vivant") was sown in hanging baskets ( $\emptyset$  20 cm.) and infected with the cereal aphid *Sitobion avenae*. Small numbers of parasites (20-30 /banker) were introduced on the bankers. It took two generations before a large number of mummies (2-3000/banker) could be found. Most bankers were started in February (20/ha.) and peaked in parasite production at the beginning of April when first infections with *A. solani* could be expected.

By putting yellow sticky traps in the greenhouse it was possible to monitor the

production and dispersal of adult *A. abdominalis* over time. Peak production of the bankers occurred in week 15 (around 8-10 weeks after their introduction); bankers were removed at week 20, because all aphids were parasitised and all mummies were emerged. Until week 29 *A. abdominalis* was trapped on yellow sticky traps. The impact of *A. abdominalis* on the population of *A. solani* was hard to quantify. Marked colonies of *A. solani* often had disappeared when inspected again after one week. This can be partly explained by host feeding of the parasites. Black mummies however were found not only on leaves, but also at the base of the stem, on ropes, plastic of the rock wool pot, and even on the ground.

In 2002 the banker plant system was used on approximately 100 ha. One grower (10 ha.) did not need to use any chemicals for aphid control, though several infections with *A. solani* were observed. Most other growers were able to reduce the number of Pirimicarb applications from around 7 to 2-3.

#### THE INFLUENCE OF TIMING OF FRUIT SET ON APHID CONTROL

An aspect which is easily overlooked is the influence of the physiology of the plant on aphid

growth and subsequently aphid control. We observed large differences in aphid growth between different greenhouses and we concluded that this could only be explained by differences in plant physiology. As an example in figure 4 the number of newly formed pepper fruits per week is plotted for the same greenhouse in two consecutive years. In 2001 fruit set occurred continuously during the year. In 2002 however, oscillations with a period of 4 weeks occurred. Especially in peppers fluctuations in fruit set is a "natural" phenomenon, though plant breeders are selecting for varieties with a more even fruit set. Also abiotic factors (like day/night temperature and feeding regime) can influence fruit set. Finally growers know that if fruit set is unequal at the beginning of the season it is almost impossible to go back to a more even fruit set later on. As a consequence it is observed that the aphid population growth is reflected in the fruit set pattern. In the vegetative phase the aphid population will grow much faster than in the fruiting phase. Small infections with *A. solani* will explode very fast in this period. Until now practical consequences for the release of beneficial insects have not been considered. However if the biological control is already poorly established and the crop is in a vegetative phase, an advice to use a chemical correction will be given sooner.

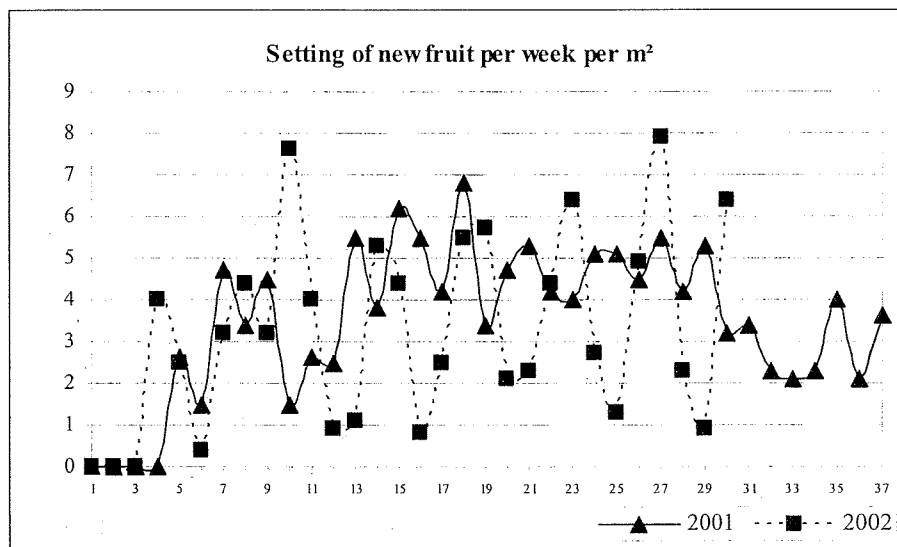


Fig. 4. Setting of new peppers per week/m<sup>2</sup> for 2 consecutive years.

## OVERALL CONCLUSIONS

The biological control of the foxglove aphid *A. solani* in peppers has been improved by several measurements. By comparing two lines of *A. abdominalis*, a line was chosen which performs better at lower temperatures. This is important because there is a tendency to lower the temperatures in greenhouses during winter and spring. Because the rate of increase of *A. abdominalis* is rather low, a banker plant system was developed to rear high numbers of parasites before *A. solani* can be expected in the greenhouse. With an average of 20 bankers per hectare and 3000 mummies per banker, around 6 parasites/m<sup>2</sup> were reared in the greenhouse. Numbers were sometimes that high that the number of predated aphids was even higher than the number of mummies found.

The hypothesis that the honeydew of the aphids was toxic for *Aphidius* spp. could not be confirmed. However the result that this honeydew had the same nutritional value as water was remarkable.

Future research will focus on further improvement of the banker plant system (optimal timing, quality of the banker plant), the screening of new parasites and predators. The influence of the plant physiology on the aphids and subsequently on biological control is challenging but can only be solved in cooperation with other partners.

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