

Interactions between the parasitoid wasp *Dinocampus coccinellae* and two species of coccinellid from Japan and Britain

Satoshi Koyama · Michael E. N. Majerus

Received: 4 September 2007 / Accepted: 30 October 2007 / Published online: 15 November 2007
© International Organization for Biological Control (IOBC) 2007

Abstract The establishment, spread and increase of the invasive coccinellid *Harmonia axyridis* Pallas in North America and Britain have coincided with declines in native ladybirds. In Britain, this pattern was predicted soon after *H. axyridis* was first recorded. However, predictions of the impact that *H. axyridis* may have on the parasites, parasitoids and pathogens of native coccinellids have been less certain, largely because of uncertainty over whether *H. axyridis* would become an alternate host for such agents. *Dinocampus coccinellae* (Schrank) is a braconid parasitoid of ladybirds of the sub-family Coccinellinae. In Japan, this wasp uses both *Coccinella septempunctata brucki* Mulsant and *H. axyridis* as hosts, but successfully parasitizes a higher proportion of the former species. Data are here presented that indicate the same is true in Britain, but to a greater extent. This study investigates the interactions of *D. coccinellae* with *C. septempunctata* L. and *H. axyridis* from Japan and Britain. We show that coccinellid activity affects encounter rates with the wasp, with *H. axyridis* being more active than *C. septempunctata* in the tests. Escalation rates from investigation to ovipositional attack were higher for *C. septempunctata* than *H. axyridis* for both Japanese and British wasps. Wasp emergence was higher, relative to ovipositional attack, from *C. septempunctata* than from *H. axyridis*. Wasps did not discriminate between Japanese and British ladybirds of the same species. British wasp eclosion rates were higher from Japanese than from British *H. axyridis*. We consider both the effect *D. coccinellae* may have on *H. axyridis* population demography, and the effect *H. axyridis* may have on *D. coccinellae* in Europe.

S. Koyama · M. E. N. Majerus (✉)
Department of Genetics, Downing Street, Cambridge CB2 3EH, UK
e-mail: m.majerus@gen.cam.ac.uk

Present Address:

S. Koyama
Department of Veterinary Medicine, Faculty of Agriculture, Tokyo University of Agriculture and Technology, 3-5-8 Saiwai, Fuchu, Tokyo, Japan
e-mail: skoyama@cc.tuat.ac.jp

Keywords Braconidae · *Coccinella septempunctata* · Coccinellidae · *Dinocampus coccinellae* · Harlequin ladybird · *Harmonia axyridis* · Host favourability · Host selection · Parasitoid wasp

Introduction

The invasive alien coccinellid, *Harmonia axyridis* Pallas, has been widely released as a biological control agent of aphids (Gordon 1985; Koch 2003). It became established in North America in the mid 1980s (Chapin and Brou 1991) and in mainland Europe in 1999 (Brown et al. 2007a). More recently, it has spread to Britain, where it became established in 2004 (Majerus et al. 2006a). In all three regions it has subsequently spread and increased rapidly (Koch 2003; Brown et al. 2007a, b), threatening biodiversity through predation of non-target aphids and coccids (Majerus and Roy 2005), intra-guild competition and intra-guild predation (Majerus et al. 2006b; Roy et al. 2006; Ware and Majerus 2007). One intriguing and unanswered question is why *H. axyridis* appears to co-exist with other coccinellids in its native range, but has a tendency to displace other coccinellids in parts of its introduced range (Majerus et al. 2006b; Roy et al. 2006; Snyder and Evans 2006). Although a variety of possible reasons for this situation have been suggested (large size, better attack capabilities, shape of mandibles, better chemical and physical defences of immature stages, phenotypic plasticity, multivoltinism, high fecundity, low prevalence of parasitoids, parasites and pathogens in introduced range) (Koch 2003; Pell et al. 2007 for reviews), experimental evidence in support of any of these speculations is lacking.

Dinocampus coccinellae (Schrank) (Hymenoptera: Braconidae) is a solitary parasitoid of coccinellids of the sub-family Coccinellinae (Ceryngier and Hodek 1996). Within this sub-family, it has been reported from over 50 species. The species reproduces by thelytokous parthenogenesis. Female wasps lay a single egg into hosts during an oviposition attack (Balduf 1926). However, females do not discriminate against hosts into which they or other females have already oviposited (Ceryngier and Hodek 1996; Geoghegan et al. 1998; Majerus et al. 2000). This is despite the fact that only a single wasp can develop in a host (Ceryngier and Hodek 1996; Geoghegan et al. 2000).

Various studies have shown that *D. coccinellae* females have a range of host preferences. Richerson and DeLoach (1972) noted that the wasp usually prefers larger coccinellid species, with recorded prevalences (proportion infected) being highest in *Coleomegilla maculata* (DeGeer) and *Hippodamia convergens* (Guérin) in North America (Balduf 1926; Obrycki 1989), *Coccinella septempunctata* L. in continental Europe (Ipert 1964), *C. septempunctata brucki* Mulsant in Japan (Koide 1961; Maeta 1969; Kawachi 1984) and *Coccinella transversalis* (Mulsant) in Australia (Anderson et al. 1986). Within a host species, field prevalence levels are higher in females than males (Maeta 1969; Parker et al. 1977; Cartwright et al. 1982). In laboratory tests, Davis et al. (2006) showed that female *D. coccinellae* have a preference to oviposit into female rather than male adults of *C. septempunctata*, and that this preference was in addition to the preference the wasp shows for larger hosts in choice tests. As female coccinellids eat more than males (Hodek 1996; Dixon 2000), Davis et al. (2006) argue that this preference is adaptive, providing developing wasp larvae with increased resources. *Dinocampus coccinellae* also has a preference to oviposit into adult compared to immature coccinellids (second, third and fourth instar larvae or pupae), in both choice and no choice tests (Geoghegan et al. 1998). This preference is also adaptive, as the level of successful wasp emergence was higher from adult coccinellids than from fourth instar larvae (Geoghegan et al. 1998). Finally,

D. coccinellae oviposits preferentially into young (pre-winter) rather than old (overwintered) hosts of the same sex and species, conferring increased fitness on offspring because recently eclosed hosts are more likely to survive to allow full parasitoid development than are old adults (Majerus et al. 2000).

In Britain, highest prevalence of *D. coccinellae* has been recorded in *C. septempunctata*, *Coccinella undecimpunctata* L. and *Harmonia quadripunctata* (Pontoppidan) (Geoghegan et al. 1997; Majerus 1997). We here give comparative data on the prevalence of *D. coccinellae* from wild, British samples of *H. axyridis* and *C. septempunctata*. Further, we present findings of choice tests to determine whether *D. coccinellae* shows any preference to attack *C. septempunctata* or *H. axyridis* in a choice situation, and whether the origin of wasps used (Japanese or British) had an effect on the outcome.

Materials and methods

Collection and treatment of field prevalence samples

Samples of *C. septempunctata* and *H. axyridis* were collected in south-west London (Battersea and Clapham districts), in March 2007. The samples were fed on an artificial diet (Majerus et al. 1989) for 1 day, and thereafter on *Acyrtosiphon pisum* (Harris) (pea aphids), at 21°C. The samples of both species were sexed, and then divided into two sub-samples, A and B, at a ratio of 2:1 for each sex. Sub-sample A beetles were retained on a diet of excess *A. pisum*, daily for 45 days. All ladybirds were dissected after 45 days, or when they died if earlier, and were examined for evidence of parasitization. Larval emergence, pupation and adult eclosion of *D. coccinellae* were recorded. Sub-sample B beetles were anaesthetized with CO₂, killed and dissected within a week of collection. Immature stages of *D. coccinellae* that were found were recorded. The two treatments, A and B, were undertaken to test whether prevalence levels in adults retained under laboratory conditions were not underestimated as a result of incomplete ascertainment when parasites failed to complete development.

Experimental stocks

Ladybirds used in choice tests were of four types, British *H. axyridis*, Japanese *H. axyridis*, *C. septempunctata septempunctata* L. and *C. septempunctata brucki*. British *H. axyridis* were either field-collected or F1 adults from samples collected in London in November 2005, May 2006 or September 2006. Japanese *H. axyridis* were F1 adults from samples collected in Fuchu, Honshu, in September 2005 or May 2006. *Coccinella septempunctata* were taken from overwintering sites around Cambridge and Thetford Forest, in February, March and October 2006. *Coccinella septempunctata brucki* were either field-collected or F1 adults from samples collected in Fuchu, Honshu, in May 2006. Field-collected ladybirds were kept in culture on *A. pisum*, at 21°C, for a minimum of 28 days before use to ensure that they were not previously infected with *D. coccinellae*. F1 *H. axyridis* were between 21 and 56 days old. F1 *C. septempunctata* and *C. septempunctata brucki* were between 21 and 56 days old and had not experienced a diapause. Ladybirds were sexed under CO₂ anaesthetic using previously described criteria (for *C. septempunctata* see Randall et al. 1992; for *H. axyridis* see Majerus et al. 1998). All ladybird stocks were maintained at 21°C, 16L:8D, on *A. pisum*, with a once weekly provision of artificial food as described by Geoghegan et al. (2000).

The *D. coccinellae* adults used in tests were either of British origin from *C. septempunctata* from Cambridgeshire or Suffolk, or of Japanese origin from *C. septempunctata brucki* from the Fuchu stock. They were fed on an agar based artificial diet (Geoghegan et al. 2000).

Test procedures

Experiment 1

Does *D. coccinellae* prefer hosts from its own region?

Two female ladybirds of the same species (either *H. axyridis* or *C. septempunctata*), but from different countries, were placed in a 9 cm Petri dish. A single *D. coccinellae* of known origin (Japanese or British) was introduced into the dish at least 10 min after the ladybirds. The ladybirds used in each test were chosen randomly from a sample: they were not match-paired for size or other phenotypic traits apart from sex. All *H. axyridis* used were f. *succinea*. The two sub-species of *C. septempunctata* were easily distinguishable. When Japanese and British *H. axyridis* were used in the same test, patterning details were used to distinguish individual origin. Each ladybird was used only in a single test. Each wasp was used for a single test only. All four possible combinations of wasp and ladybird species were tested. Each type of test was replicated 20 times.

All interactions between wasp and ladybirds were recorded over the 20-min period immediately following the introduction of the wasp. Behavioural observations were categorized as: pursuance and investigation of the host (PI); ovipositional stance (OS); or ovipositional attack (OA) (after Richerson and DeLoach 1972). These categories were invariably consequent on one another, with OA following from OS and OS following from PI (Richerson and DeLoach 1972; Davis et al. 2006; this study). The origin of the ladybird involved in each interaction with a wasp was recorded. Whether a coccinellid involved in an interaction with *D. coccinellae* was moving at the moment that the interaction was initiated was recorded. As *D. coccinellae* does not avoid superparasitism (Ceryngier and Hodek 1996; Geoghegan et al. 1998; Majerus et al. 2000), comparative tests of total number of OAs on ladybirds could be used.

Experiment 2

Do Japanese *D. coccinellae* discriminate between *H. axyridis* and *C. septempunctata*?

The protocol used was broadly as that described for experiment 1. However, only Japanese wasps were used, and the two ladybirds used in a choice test were one *H. axyridis* female and one *C. septempunctata* female, with both ladybirds originating from the same country (Japan or Britain). Each type of test was replicated ten times.

Experiment 3

Do British *D. coccinellae* discriminate between *H. axyridis* and *C. septempunctata*?

The protocol was as used for experiment 2, but with British wasps used in place of Japanese wasps. Each type of test was replicated ten times.

When possible, ladybirds that were used in tests and had been the subject of OA were thereafter maintained singly in culture on a diet of *A. pisum* for 5 weeks, and wasp cocoons resulting were recorded.

Statistical analysis

For the field samples, an orthogonal analysis of *G*-test for goodness-of-fit was used to compare numbers recorded, with the level of prevalence of the parasite being considered against the variables: host species, host sex and ladybird sub-sample (A or B). Interaction between these factors was obtained from the residual *G*-value. In addition, the *G* statistic was calculated to compare the success of parasitism between host species, using the sub-set comprising only those ladybirds that were infected.

In the preference experiments, *G*-test goodness-of-fit tests were used to compare results. Where a series of comparisons were made, a fully orthogonal analysis of *G* was used to avoid repeat use of numerical data. A similar approach was used to analyse data on the development of *D. coccinellae* in different hosts following observed OA.

Results

Field prevalences

Details of the samples collected to assess field prevalence levels are given in Table 1. All larvae that emerged from both coccinellid species in the A sub-samples successfully pupated and eclosed. Total *G*-test for goodness-of-fit, of parasitized or not, with variables host species, host sex, and sub-sample, was highly significant ($G = 31.16$, $df = 9$, $p < 0.001$). The overall prevalences of *D. coccinellae* in the two species were 0.122 ± 0.018 for *C. septempunctata* and 0.055 ± 0.009 for *H. axyridis*, and were significantly different from one another ($G = 11.79$, $df = 1$, $p < 0.001$). There was no significant difference in the prevalence levels in male compared to female hosts, either in the two species combined ($G = 1.4$, $df = 1$, $p > 0.05$), or either species individually (for *C. septempunctata*, $G = 2.62$, $df = 1$, $p > 0.05$; for *H. axyridis*, $G = 0.02$, $df = 1$, $p > 0.05$). There was no significant difference in prevalence between sub-samples A and B for the two species combined ($G = 0.44$, $df = 1$, $p > 0.05$) or for either species individually (for *C. septempunctata*, $G = 0.72$, $df = 1$, $p > 0.05$; for *H. axyridis*, $G = 2.92$, $df = 1$, $p > 0.05$). The residual *G*-value representing the interaction between variables with respect to the numbers parasitized or not was significant ($G = 11.25$, $df = 2$, $p < 0.05$).

Within the sub-set of parasitized ladybirds, the proportion of infected individuals in the A sub-samples that produced adult wasps was significantly greater for *C. septempunctata* than for *H. axyridis* ($G = 41.66$, $df = 1$, $p < 0.001$).

Experiment 1

A summary of the interactions is given in Table 2. The numbers of OA did not differ significantly between test types ($G = 4.12$, $df = 3$, $p < 0.05$). There was no significant difference in the level of escalation from PI through OS to OA for wasps of either origin

Table 1 Details of field samples of *C. septempunctata* and *H. axyridis* from South-west London

<i>Coccinella septempunctata</i> Sub-sample A					
Males			Females		
Uninfected	Produced adult wasp	Dissection revealed infection	Uninfected	Produced adult wasp	Dissection revealed infection
102	10	2	88	16	1
<i>Coccinella septempunctata</i> Sub-sample B					
Males			Females		
Uninfected	Produced adult wasp	Dissection revealed infection	Uninfected	Produced adult wasp	Dissection revealed infection
53	N/A	4	46	N/A	7
<i>Harmonia axyridis</i> Sub-sample A					
Males			Females		
Uninfected	Produced adult wasp	Dissection revealed infection	Uninfected	Produced adult wasp	Dissection revealed infection
189	1	11	264	0	9
<i>Harmonia axyridis</i> Sub-sample B					
Males			Females		
Uninfected	Produced adult wasp	Dissection revealed infection	Uninfected	Produced adult wasp	Dissection revealed infection
96	N/A	5	123	N/A	13

The numbers of uninfected and infected males and females of each host species are given. Infected hosts are sub-divided into those in which wasps emerged and those revealed by dissection. For differences in treatments of sub-samples A and B, see text

Table 2 Results of experiment 1, in which the interactions of both Japanese and British *D. coccinellae* to sympatric and allopatric *C. septempunctata* and *H. axyridis* were tested

Origin of wasps	Host species	Origin of hosts	Interactions observed		
			PI	OS	OA
Japan	<i>C. septempunctata</i>	Japan	137	74	49
	<i>C. septempunctata</i>	UK	119	57	47
Japan	<i>H. axyridis</i>	Japan	128	72	34
	<i>H. axyridis</i>	UK	145	82	27
British	<i>C. septempunctata</i>	Japan	85	52	28
	<i>C. septempunctata</i>	UK	88	60	35
British	<i>H. axyridis</i>	Japan	205	96	32
	<i>H. axyridis</i>	UK	198	75	30

Interactions: PI pursuance and investigation, OS ovipositional stance, OA ovipositional attack, after Richerson and DeLoach 1972

towards allopatric compared with sympatric ladybirds for either *C. septempunctata* or *H. axyridis* (for Japanese wasp with *C. septempunctata* $G = 0.66$, $df = 2$, $p > 0.05$; with *H. axyridis* $G = 1.68$, $df = 2$, $p > 0.05$; for UK wasps with *C. septempunctata* $G = 0.46$, $df = 2$, $p > 0.05$; with *H. axyridis* $G = 3.4$, $df = 2$, $p > 0.05$).

Experiment 2

The results (Table 3) show that Japanese wasps did not differ in the levels of OA in the two tests ($G = 3.24$, $df = 1$, $p > 0.05$). It was noted, however, that the wasps rarely investigated (PI) non-active *C. septempunctata*, while they often investigated non-active *H. axyridis*, the difference being significant with both Japanese ($G = 24.85$, $df = 1$, $p < 0.001$), and UK ($G = 6.92$, $df = 1$, $p < 0.001$) ladybirds. The rate of escalation from PI to OA was not significantly different between the two species for Japanese ladybirds ($G = 0.3$, $df = 2$, $p > 0.05$), but was for UK ladybirds ($G = 4.58$, $df = 1$, $p < 0.05$), with greater escalation being observed towards *C. septempunctata*.

Experiment 3

The results (Table 4) show that British wasps have a significant preference for PI towards Japanese *H. axyridis* over *C. septempunctata brucki* ($G = 17.25$, $df = 1$, $p < 0.001$). This is largely a product of a much higher level of activity by the *H. axyridis* in these tests compared to *C. septempunctata brucki*. When the level of PI towards stationary *H. axyridis* and *C. septempunctata brucki* are compared, the level of PI is greater towards the latter, and the difference is not significant ($G = 1.88$, $df = 1$, n.s.). Overall, the rate of escalation from PI through OS to OA was significantly greater for *C. septempunctata brucki* than for *H. axyridis* ($G = 8.95$, $df = 2$, $p < 0.05$). Analysis of G showed this to be due to a higher rate of escalation from OS to OA in *C. septempunctata brucki* than in *H. axyridis*.

In interactions with British ladybirds, the British wasps showed no overall difference in levels of PI, OS or OA towards *C. septempunctata* compared with *H. axyridis* (G -test

Table 3 Results of experiment 2 in which Japanese wasps were offered a choice between *C. septempunctata* and *H. axyridis* from either Japan or UK

Ladybird activity	<i>C. septempunctata brucki</i> (Japan)			<i>H. axyridis</i> (Japan)		
	PI	OS	OA	PI	OS	OA
Moving	55	28	18	66	23	15
Not moving	6	4	2	37	27	13
Total	61	32	20	103	50	28
	<i>C. septempunctata</i> (UK)			<i>H. axyridis</i> (UK)		
	PI	OS	OA	PI	OS	OA
Moving	72	35	31	70	25	14
Not moving	10	6	3	34	16	7
Total	82	35	31	104	42	21

Whether ladybirds were moving or stationary when first contacted by a wasp is recorded

Table 4 Results of experiment 3 in which British wasps were offered a choice between *C. septempunctata* and *H. axyridis* from either Japan or UK

Ladybird activity	<i>C. septempunctata brucki</i> (Japan)			<i>H. axyridis</i> (Japan)		
	PI	OS	OA	PI	OS	OA
(a) Moving	11	7	3	83	46	9
Not moving	59	30	24	45	19	10
Total	70	37	27	128	65	19
	<i>C. septempunctata</i> (UK)			<i>H. axyridis</i> (UK)		
	PI	OS	OA	PI	OS	OA
(b) Moving	17	11	6	38	23	6
Not moving	34	32	4	18	10	3
Total	51	43	10	56	33	9

Whether ladybirds were moving or stationary when first contacted by a wasp is recorded

goodness-of-fit $p > 0.05$ for all comparisons). However, this lack of difference may be the result of differences in the activities of the two species of ladybird. *Harmonia axyridis* was more active than *C. septempunctata*, which is reflected in the higher level of wasp interactions with active *H. axyridis* and stationary *C. septempunctata*.

Successful development of *D. coccinellae* in different hosts

Table 5 gives the numbers of those ladybirds of the different species from different origins that were attacked by Japanese or British wasps and were then retained under suitable conditions that subsequently produced adult wasps. The origin of wasps did not affect the probability of successful development of their progeny in either species of ladybird (G -test $p > 0.05$ for all comparisons) (however, note the low successful development in UK *H. axyridis*). When considering wasps from both Japan and Britain together, significantly more wasps developed to adulthood in *C. septempunctata* than in *H. axyridis* ($G = 36.92$, $df = 1$, $p < 0.001$). Moreover, successful development was significantly higher in Japanese *H. axyridis* than in British *H. axyridis* ($G = 6.44$, $df = 1$, $p < 0.05$). Conversely, there was no significant difference in the rate of successful development in *C. septempunctata brucki* compared to *C. septempunctata* ($G = 1.04$, $df = 1$, $p > 0.05$).

Table 5 Number of wasps that emerged from Japanese and UK *H. axyridis* and *C. septempunctata* that were attacked (OA) during experiments

Host species	Number attacked (OA)		Number that produced adult wasps	
	Japanese wasps	UK wasps	Japanese wasps	UK wasps
<i>H. axyridis</i> Japan	36	34	9	9
<i>H. axyridis</i> UK	18	23	3	0
<i>C. septempunctata brucki</i>	38	45	21	22
<i>C. septempunctata</i>	30	33	17	21

Results are given separately for wasps of Japanese and UK origin

Discussion

In this paper, we have described initial tests to investigate interactions between the parasitoid wasp *D. coccinellae* and two coccinellids, *H. axyridis* and *C. septempunctata*.

Comparison of UK field collected sub-samples A and B showed that prevalence levels on hosts retained to allow wasp larvae to emerge, and those dissected soon after collection were similar, indicating that retention did not lead to incomplete ascertainment of unsuccessful parasitizations in the retained (A) sub-samples.

Prevalence level of the wasp was more than twice as high in *C. septempunctata* than in *H. axyridis*. Moreover, the proportion of parasitized individuals in which *D. coccinellae* successfully completed development was significantly lower for *H. axyridis* than for *C. septempunctata*. This may be a result of the host specificity of UK *D. coccinellae* if these wasps are poorly adapted to *H. axyridis*, which only established in Britain in 2004 (Majerus et al. 2006a). However, tests in which UK and Japanese wasps were offered a choice of sympatric and allopatric *H. axyridis*, showed that wasps had no preference for sympatric ladybirds. The same result was found when wasps were tested against UK and Japanese sub-species of *C. septempunctata*. This suggests that the low prevalence of *D. coccinellae* in UK *H. axyridis* is not the result of the unfamiliarity of UK wasps with *H. axyridis*. That said, measures of the proportion of wasps that successfully developed in UK *H. axyridis* compared with Japanese *H. axyridis* showed that the former is a significantly poorer host, irrespective of whether ovipositing wasps were from Japan or the UK.

The reason for the low level of developmental success of *D. coccinellae* in UK *H. axyridis* is unknown and warrants further investigation. However, it is feasible that during the establishment of stocks by biocontrol companies, selection has inadvertently been imposed against *H. axyridis* individuals that have low resistance to *D. coccinellae* through guarantee procedures imposed on the stocks of *H. axyridis* obtained from its native range. Those *H. axyridis* that produced wasps, and those that failed to reproduce rapidly [*D. coccinellae* infection suppresses host ovarian maturation (e.g. Ceryngier and Hodek 1996)] would be discarded from stocks.

In tests where Japanese and British wasps were offered a choice between *H. axyridis* and *C. septempunctata*, interactions were affected by the levels of activity shown by the two species of ladybird. In general, *H. axyridis* were more active than *C. septempunctata*, with the result that wasp encounters with *H. axyridis* were more frequent than encounters with *C. septempunctata*. However, importantly, for both Japanese and UK wasps, rates of escalation from PI or OS to OA were higher towards *C. septempunctata* than *H. axyridis*. These higher escalation rates may explain the higher prevalences of *D. coccinellae* in *C. septempunctata* compared to *H. axyridis* that have been reported in both Japan (Maeta 1969; Kawauchi 1984) and Britain (this study).

Two cautionary notes should be borne in mind in further investigations of this type. First, all wasps used were from *C. septempunctata*. It is possible that natural populations of *D. coccinellae* are sub-divided by host, with matrilineal adaptations to, and preferences to oviposit into, the species of host that they developed in. Whether this is the case has not been tested. However, in tests in which *D. coccinellae* from *C. undecimpunctata* (a suitable host) were offered a choice between *C. undecimpunctata* and *Adalia bipunctata* (L.) (an unsuitable host) (Ipert 1964; Majerus 1994; Ceryngier and Hodek 1996), wasps attacked both species equally (S. Koyama and M. E. N. Majerus, unpublished data).

Second, the activity levels of the ladybirds used in choice tests seemed to affect the frequency of some interactions. It is likely that the lower activity of *C. septempunctata* compared to *H. axyridis* was a consequence of differences in the life-history strategies of

the two species. In appropriate conditions, *H. axyridis* will mate and oviposit following eclosion without requirement for a dormant period (Majerus and Roy 2005), while *C. septempunctata* usually requires such a period before becoming reproductively active (Dobrzanski 1922a, b). While it is difficult to control coccinellid activity levels, we suggest that in future tests, *C. septempunctata* that had undergone a period of dormancy, and so were reproductively active, should be used.

The initial aims of this study were to consider first, whether *D. coccinellae* is likely to have any significant impact on populations of *H. axyridis* in Europe, and second, whether the spread and increase of *H. axyridis* in Europe is likely to affect *D. coccinellae*. The wasps low prevalence in the field, the lower rate of escalation to OA in *H. axyridis* compared to *C. septempunctata* and the very low successful development of the wasp in UK *H. axyridis* all argue that *D. coccinellae* will have little impact on *H. axyridis* populations in Europe.

Conversely, the spread and increase of *H. axyridis* in Europe may have a detrimental impact on *D. coccinellae*. *Dinocampus coccinellae* attacks a wide range of Coccinellinae species (Ceryngier and Hodek 1996), preferring larger species (Richerson and DeLoach 1972). In locations where the commonest coccinellid encountered by *D. coccinellae* becomes *H. axyridis*, a host in which a high proportion of the parasitoid's eggs fail to develop to adulthood, much of the reproductive output of the *D. coccinellae* populations will be misdirected. For example, one might compare two sites, one containing 3,000 *C. septempunctata* and no *H. axyridis*, the other containing 1,000 *C. septempunctata* and 2,000 *H. axyridis*. Using the prevalence levels observed in UK field samples and the rate of successful development in the A sub-samples, the former site would result in 328 wasps (host population size \times prevalence level in *C. septempunctata* \times parasite success rate in this host), while the latter would result in 112 wasps (109 from *C. septempunctata* and just three from *H. axyridis*). It thus seems possible that populations of *D. coccinellae* will decline as *H. axyridis* increases and displaces native Coccinellinae. If this is the case, we predict that prevalences of *D. coccinellae* will be greater in suitable native host species, such as *C. septempunctata*, in parts of Europe where *H. axyridis* is not yet established, compared to parts of Europe where *H. axyridis* already predominates. Survey work across Europe should allow this prediction to be verified or refuted. An alternative to this scenario is that *D. coccinellae*, in areas where *H. axyridis* is abundant, will respond to selective pressure imposed by this abundance and become better adapted to this host. In this case, we predict that the proportion of infected *H. axyridis* that produce adult wasps will increase over time. This prediction can also be tested by field sampling over time, with assessment of both prevalence of parasitization and success of parasitization being assayed.

Acknowledgements We thank Ian Wright for technical support, Remy Ware, Sherif El Nagdy, Laura Jane Michie and Sami Hassan for help with culture maintenance, and Yoshiaki Obara, Remy Ware and Christina Majerus for help in collecting samples.

References

- Anderson JME, Hales DF, van Brunschot KA (1986) Parasitisation of coccinellids in Australia. In: Hodek I (eds) Ecology of aphidophaga. Academia, Prague and Dr. W. Junk, Dordrecht
- Balduf WV (1926) The bionomics of *Dinocampus coccinellae* Schrank. Ann Entomol Soc Am 19:465–489
- Brown PMJ, Adriaens T, Bathon H, Cuppen J, Golarazena A, Hägg T, Kenis M, Klausnitzer BEM, Kovář I, Loomans AJM, Majerus MEN, Nedved O, Pedersen J, Rabitsch W, Roy HE, Ternois V, Zakharov IA, Roy DB (2007a) *Harmonia axyridis* in Europe: spread and distribution of a non-native coccinellid. BioControl (this issue). doi:10.1007/s10526-007-9132-y

- Brown PMJ, Roy HE, Rothery P, Roy DB, Ware RL, Majerus MEN (2007b) *Harmonia axyridis* in Great Britain: analysis of the spread and distribution of a non-native coccinellid. *BioControl* (this issue). doi: [10.1007/s10526-007-9124-y](https://doi.org/10.1007/s10526-007-9124-y)
- Cartwright B, Eikenbary RD, Angalet GW (1982) Parasitism by *Perilitus coccinellae* (Hym.: Braconidae) of indigenous coccinellid hosts and the introduced *Coccinella septempunctata* (Col.: Coccinellidae), with notes on winter mortality. *Entomophaga* 27:237–244
- Ceryngier P, Hodek I (1996) Enemies of Coccinellidae. In: Hodek I, Honek A (eds) *Ecology of Coccinellidae*. Kluwer, Dordrecht
- Chapin JB, Brou VA (1991) *Harmonia axyridis* (Pallas), the 3rd species of the genus to be found in the United States (Coleoptera, Coccinellidae). *Proc Entomol Soc Wash* 93:630–635
- Davis DS, Stewart SL, Manica A, Majerus MEN (2006) Adaptive preferential selection of female coccinellid hosts by the parasitoid wasp *Dinocampus coccinellae* (Hymenoptera: Braconidae). *Eur J Entomol* 103:41–45
- Dixon AGF (2000) *Insect predator-prey dynamics: ladybird beetles and biological control*. Cambridge University Press, Cambridge
- Dobrzanski FG (1922a) Imaginal diapause in Coccinellidae. *Izv Otdel Prikl Entomol* 2:103–124
- Dobrzanski FG (1922b) Mass aggregations and migrations in Coccinellidae. *Izv Otdel Prikl Entomol* 2:229–234
- Geoghegan IE, Thomas WP, Majerus MEN (1997) Notes on the coccinellid parasitoid *Dinocampus coccinellae* (Schrank) (Hymenoptera: Braconidae) in Scotland. *Entomologist* 116:179–184
- Geoghegan IE, Majerus TMO, Majerus MEN (1998) Differential parasitisation of adult and pre-imaginal *Coccinella septempunctata* (Coleoptera: Coccinellidae) by *Dinocampus coccinellae* (Hymenoptera: Braconidae). *Eur J Entomol* 95:571–579
- Geoghegan IE, Chudek JA, Mackay RL, Lowe C, Moritz S, McNicol R, Hunter G, Majerus MEN (2000) Study of the anatomical changes in *Coccinella septempunctata* (Coleoptera: Coccinellidae) induced by diet and by infection with the larvae of *Dinocampus coccinellae* (Hymenoptera: Braconidae) using magnetic resonance microimaging. *Eur J Entomol* 97:457–461
- Gordon RD (1985) The Coleoptera (Coccinellidae) of America north of Mexico. *J N Y Entomol Soc* 93: 1–912
- Hodek I (1996) Food relationships. In: Hodek I, Honek A (eds) *Ecology of Coccinellidae*. Kluwer, Dordrecht
- Iperti G (1964) Les parasites des Coccinelles aphidiphages dans les Basses-Alpes et les Alpes-Maritimes. *Entomophaga* 9:153–180
- Kawauchi S (1984) Ecological studies on the natural enemies of *Coccinella septempunctata brucki*, *Propylea japonica*, and *Scymnus (Neopullus) hoffmanni* (Coleoptera: Coccinellidae). *Kurume Univ J* 33:63–67
- Koch RL (2003) The multicoloured Asian lady beetle, *Harmonia axyridis*: a review of its biology, uses in biological control and non-target impacts. *J Insect Sci* 3:32
- Koide T (1961) Observations on *Perilitus coccinellae* (Schrank). *Gensei* 1961(11):1–5
- Maeta Y (1969) Biological studies on the natural enemies of some coccinellid beetles. I. On *Perilitus coccinellae* (Schrank). *Kontyu* 37:147–166
- Majerus MEN (1994) Ladybirds (New Naturalist 81). HarperCollins, London
- Majerus MEN (1997) Parasitization of British ladybirds by *Dinocampus coccinellae* (Schrank) (Hymenoptera: Braconidae). *Br J Entomol Nat Hist* 10:15–24
- Majerus MEN, Roy HE (2005) Scientific opportunities presented by the arrival of the harlequin ladybird, *Harmonia axyridis*, in Britain. *Antenna* 29:196–208
- Majerus TMO, Majerus MEN, Knowles B, Wheeler J, Bertrand D, Kuznetsov VN, Ueno H, Hurst GDD (1998) Extreme variation in the prevalence of inherited male-killing microorganisms between three populations of the ladybird *Harmonia axyridis* (Coleoptera: Coccinellidae). *Hereditas* 81:683–691
- Majerus MEN, Kearns PWE, Forge H, Ireland H (1989) Ladybirds as teaching aids: 1 Collecting and culturing. *J Biol Educ* 23:85–95
- Majerus MEN, Geoghegan IE, Majerus TMO (2000) Adaptive preferential selection of young coccinellid hosts by the parasitoid wasp *Dinocampus coccinellae* (Hymenoptera: Braconidae). *Eur J Entomol* 97:161–164
- Majerus MEN, Mabbott P, Rowland F, Roy HE (2006a) The harlequin ladybird, *Harmonia axyridis*, arrives in Britain. *Entomol Mon Mag* 142:87–92
- Majerus M, Strawson V, Roy H (2006b) The potential impacts of the arrival of the harlequin ladybird, *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae), in Britain. *Ecol Entomol* 31:207–215
- Obrycki JJ (1989) Parasitization of native and exotic coccinellids by *Dinocampus coccinellae* (Schrank) (Hymenoptera: Braconidae). *J Kans Entomol Soc* 62:211–218

- Parker BL, Whalon ME, Warshaw M (1977) Respiration and parasitism in *Coleomegilla maculata lengi* (Coleoptera: Coccinellidae). *Ann Entomol Soc Am* 70:984–987
- Pell JK, Baverstock J, Roy HE, Ware RL, Majerus MEN (2007) Intraguild predation involving *Harmonia axyridis*: a review of current knowledge and future perspectives. *BioControl* (this issue). doi: [10.1007/s10526-007-9125-x](https://doi.org/10.1007/s10526-007-9125-x)
- Randall K, Majerus MEN, Forge H (1992) Characteristics for sex determination in British ladybirds (Coleoptera: Coccinellidae). *Entomologist* 111:109–122
- Richerson JV, DeLoach CJ (1972) Some aspects of host selection by *Perilitus coccinellae*. *Ann Entomol Soc Am* 65:834–839
- Roy HE, Brown P, Majerus MEN (2006) *Harmonia axyridis*: a successful biocontrol agent or an invasive threat? In: Eilenberg J, Hokkanen H (eds) *An ecological and societal approach to biological control*. Kluwer, The Netherlands
- Snyder WE, Evans EW (2006) Ecological effects of invasive arthropod generalist predators. *Ann Rev Ecol Evol Syst* 37:95–122
- Ware RL, Majerus MEN (2007) Intraguild predation of immature stages of British and Japanese coccinellids by the invasive ladybird *Harmonia axyridis*. *BioControl* (this issue). doi:[10.1007/s10526-007-9135-8](https://doi.org/10.1007/s10526-007-9135-8)