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Ladybird population dynamics in potato: comparison of native species with an invasive species, *Harmonia axyridis*

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Abstract Following the detection of the harlequin ladybird, *Harmonia axyridis*, in 2003 in potato crops in Belgium, a study was carried out between 2004 and 2006 on the phenology of this species compared to native species in potato. The results confirmed the success of *H. axyridis*, with high population levels in 2004 and 2005. In 2006, aphid populations were very low and no *H. axyridis* larvae were sampled in potato, but the indigenous species *Coccinella septempunctata* and *Propylea quatuordecimpunctata* were detected. A species by species comparison of the date of first larvae detection, the larvae population peak, and the difference between this peak and the aphid population peak was performed. Results showed a clear correlation between *C. septempunctata* and *P. quatuordecimpunctata* and potato aphids, with a delay of 3.5 and 6.5 days between the aphid and ladybird population peaks for the two native species. *H. axyridis* arrived 7–8 days after the two indigenous species and the larval peak population occurred 15.8 days after the aphid population peak. This meant that *H. axyridis* had to complete its larval development with very low aphid populations or even with no aphids at all. The reason for its late arrival and the possible food resources used by *H. axyridis* larvae are discussed.

Keywords *Adalia bipunctata* · *Coccinella septempunctata* · *Harmonia axyridis* · Invasive species · Intraguild predation · *Propylea quatuordecimpunctata*

Introduction

Despite the reported impact of the harlequin ladybird, *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae), in the USA on indigenous ladybird populations (LaMana and Miller 1998; Colunga-Garcia and Gage 1998; Michaud 2002; Brown 2003; Alyokhin and Sewell 2004), this species was commercially introduced in large numbers into Belgium between 1997 and 2003 for aphid biological control in glasshouses. This

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ladybird species rapidly became invasive and was first reported outdoors in 2001 in northern Belgium, where glasshouse production is widespread. After this first report, *H. axyridis* populations gradually increased and they were found in spring and summer 2003 on *Acer* and *Tilia* tree species and in large numbers during winter in 2003 (Adriaens et al. 2003). In parks and gardens in Brussels, *H. axyridis* soon became the dominant species on *Tilia*, *Acer* and *Pinus* trees (San Martin 2003; Ottart 2005). In 2003 it was first recorded on arable crops in Belgium, on potato (Jansen and Warnier 2004). Eggs, larvae, nymphs and adults were found at various sites, indicating that the species could complete its development on potato, as it does in the USA (Nault and Kennedy 2003; Alyokhin and Sewell 2004).

Several authors have emphasised the risk of introducing exotic species for indigenous biodiversity (van Lenteren et al. 2003). In this context, *H. axyridis* appears to be a great threat for indigenous aphid predator species. Studies have shown that it is very aggressive and able to dominate *Coccinella septempunctata* L. and *Adalia bipunctata* (L.) (Coleoptera: Coccinellidae) in terms of competition for food resources and direct predation (Yasuda et al. 2001; Kajita et al. 2000; Hautier 2003; Hokkanen et al. 2003). This could explain why this species rapidly became dominant in several habitats when it was introduced in the USA, leading to major changes in aphidophagous ladybird diversity in natural and agricultural ecosystems.

In potato, in particular, change in the ladybird populations could disrupt aphid biological control and lead to an increase in insecticide use. Aphids can sometimes be a problem in potato for markets and industry, but aphid surveys in Belgium showed that populations rarely reached high levels, due mainly to the high activity of beneficial arthropods (Jansen 2002). In the 1994–2001 period, insecticide applications were required only in one field out of eight, on average. A dramatic decrease in aphid populations is regularly observed in mid-July, due to plant maturation and the activity of aphid-specific predators and parasitoids (Karley et al. 2003). Potato aphids are an important food source for at least three species: *C. septempunctata*, *Propylea quatuordecimpunctata* (L.) and *A. bipunctata*. These species completed their development in the potato crop between the end of June and mid-July (Jansen and Warnier 2004).

The aim of this study was to examine the establishment and phenology of *H. axyridis* in potato compared with native species, and to detect possible changes in aphidophagous ladybird populations in the crop, in order to be able to anticipate major changes in aphid control by beneficial arthropods and retain the biological control of potato aphids.

Material and methods

Field sites

Ladybirds and aphids were sampled in the 2004–2006 period in commercial potato fields at Corroy-le-Château (2004, cv Asterix), Florennes (2005 and 2006, cv Bintje), Gembloux (2004 and 2005, cv Bintje, 2006, cv Nicola) and Nivelles (2004, 2005 and 2006, cv Bintje). Potato fields were all located in typically intensive agriculture production area in Belgium. Neighbouring crops were mainly cereals (winter wheat and barley) and sugar beet. Field margins, hedgerows, tree rows, grasslands or fallows were nearly absent in these landscapes. Potato fields were at least 4 ha large and regularly received fungicide applications at intervals of 7–15 days, depending on disease pressure, humidity and temperature. In 2005, the Nivelles and Florennes fields received one insecticide application (Plenum

0.3 kg/ha, WG 50% pymetrozine) at the beginning of August to control the buckthorn aphid, *Aphis nasturtii* Kaltenbach (Homoptera; Aphididae). No other insecticide treatments were applied.

Aphid and ladybird sampling

Sampling was performed weekly, from mid-June, when the first aphids were observed, until mid-August, when no aphids or ladybirds were observed. The aphids were counted in situ on four samples of 50 leaves on each occasion. The samples were taken randomly along four transect lines in the field, using 25 leaves in lower part of the plants and 25 in the upper part.

The ladybirds were sampled using a beating method. Plants were shaken 30 s above a plastic tray (40 × 50 × 18 cm) and the insects collected were transferred to plastic vials and taken to the laboratory for counting and identification. All the samples were kept at 5–10°C during transportation to limit insect activity and avoid ladybird larvae cannibalism. Each week, ladybirds were sampled by shaking 4 × 10 plants in each field, taken randomly along four transects, as for the aphid counting. In 2006, due to the very low aphids and ladybird population levels, the sample size was increased from 10 plants to 15 per transect. The results obtained in 2006 were divided by 1.5 so that a comparison between years could be made.

Statistical analysis

The dates of the first larvae and adult detections and of the peak larvae and adult populations, and the differences between the peak larvae and adult populations and the peak aphid numbers, were compared by species and by field, using a Generalised Linear Model (GLM) with species ($p = 4$) and field × year ($q = 8$) as variables. The dates were compared two by two with the help of a Student t test based on Least Square Means at $P = 0.05$ level, using SAS software. Before analysis, the dates were converted into numerical values, taking 30 June as 0 for larvae data and 31 May as 0 for adult data.

Results

The results of the aphid population counts and the ladybird larvae and adult samplings are presented in Fig. 1 (2004), Fig. 2 (2005) and Fig. 3 (2006). The mean of the three sites is given, except for 2006 when it was calculated for two of the three sites, as no ladybirds were caught at the third site (Gembloux). Only species found at both larval and adult stages, and considered relevant to potato, were taken into account in the figures: the three native species *C. septempunctata, P. quatuordecimpunctata* and *A. bipunctata*, and the invasive species *H. axyridis.* The other species collected from time to time only at adult instar stage were the indigenous species *Hippodamia variegata* (Goeze) (1 specimen in 2004, 8 in 2005, none in 2006) and *Adalia decimpunctata* (L.) (1 specimen in 2004).

The aphid populations in 2004 and 2006 followed a typical pattern observed in potato in Belgium, with a progressive increase in populations towards the end of June, a peak between 6 and 12 July and then a rapid decline due to the activity of natural enemies of aphids and to plant physiology changes which slow down aphid population growth. The peak aphid populations in 2004 and 2006 were below the economic threshold of 10 aphids

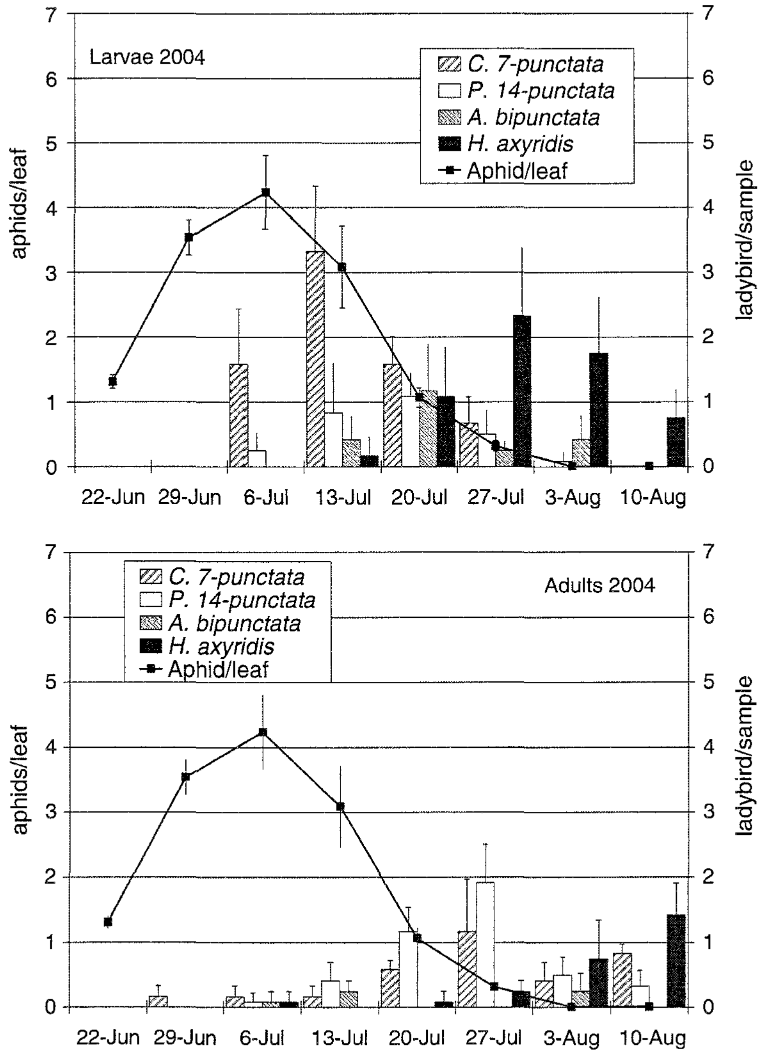

Fig. 1 Seasonal abundance of aphids and ladybird larvae and adult ladybird populations in potato in Belgium in 2004 (Corroy-le-Chateau, Gembloux and Nivelles). Mean aphids per leaf ± sd and mean ladybird per sample ± sd

per leaf, with moderate aphid populations in 2004 (a mean of 4.2 aphids per leaf, range 3.3–5.6) and low levels in 2006 (a mean of 0.7 aphids per leaf, range 0.1–1.3). The aphids observed belonged to three species of equal importance numerically: *Myzus persicae* Sulzer, *Macrosiphum euphorbiae* (Thomas) and *A. nasturtii*. Both *Aulacorthum solani* (Kaltenbach) and *Aphis fabae* (Scopoli) were also regularly found, but not in large colonies. In 2005, the aphid populations were much higher, with a mean of 18.0 aphids per leaf (range 1.3–43.2) and it was necessary to apply insecticide in two of the three fields. These high levels resulted from an *A. nasturtii* outbreak, with the populations of other aphid species being similar to the 2004 and 2006 counts. The aphid population peak in 2005 also occurred later than normal because of a second and late growing period at the beginning of

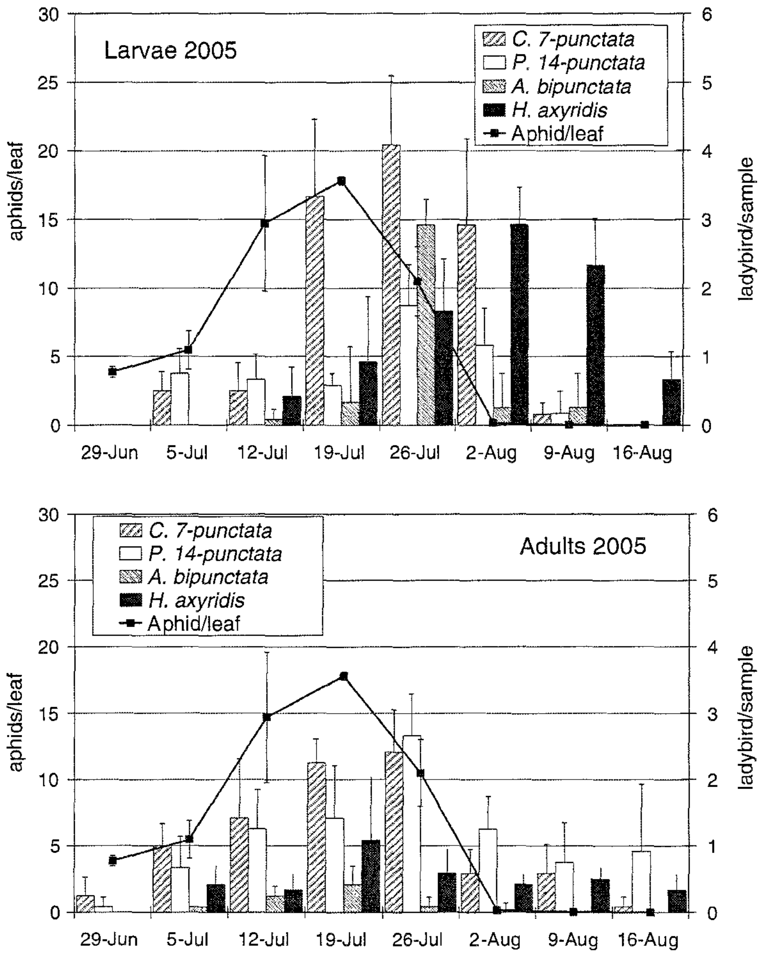

Fig. 2 Seasonal abundance of aphids and ladybird larvae and adult ladybird populations in potato in Belgium in 2005 (Florennes, Gembloux, Nivelles). Mean aphids per leaf ± sd and mean ladybird per sample ± sd

July due to abundant rainfall after 3–4 weeks of drought that stopped foliage development completely. The decline in aphid numbers observed after mid-July was due to the use of pymetrozine in two of the three fields. This insecticide is know to be selective for ladybirds and has no acute toxicity on ladybird populations (Hautier et al. 2006). However, the drastic reduction in aphid populations probably affected the ladybirds by reducing food availability and increasing cannibalism and intraguild predation.

The ladybird sampling results showed a clear population trend in 2004 and 2005 for larvae and adults. Two species, *C. septempunctata* and *P. quatuordecimpunctata*, arrived significantly earlier than *H. axyridis* and *A. bipunctata* and their population dynamics were more closely related to aphid population abundance (Table 1). The ladybird populations could be split into two groups: (1) *C. septempunctata* and *P. quatuordecimpunctata* and (2) *A. bipunctata* and *H. axyridis* which arrived 7–8 days later. The same delay was observed for the larva population peak, with a difference of 13 days between

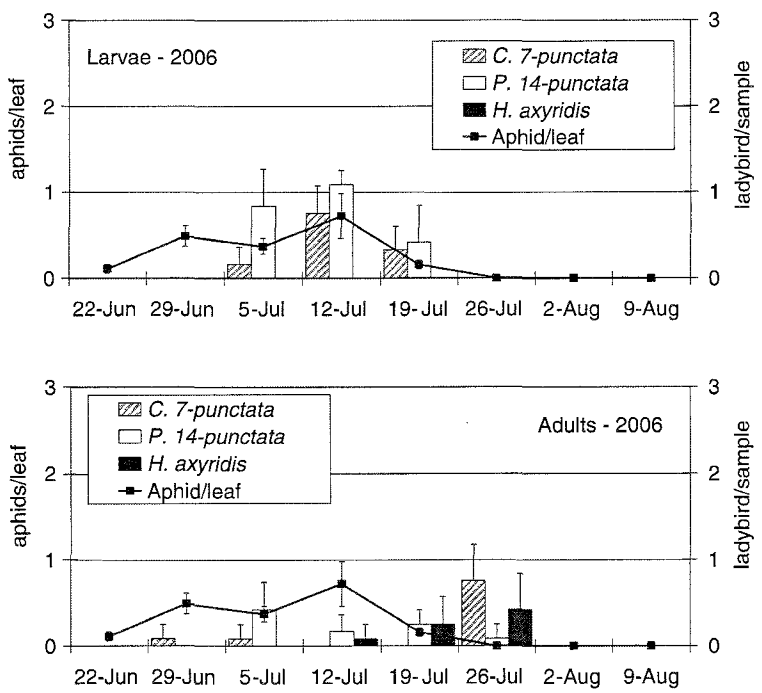

Fig. 3 Seasonal abundance of aphids and ladybird larvae and adult ladybird populations in potato in Belgium in 2006 (Florennes, Nivelles). Mean aphids per leaf ± sd and mean ladybird per sample ± sd

C. septempunctata and *H. axyridis*. This delay was also observed with regard to the date of the aphid population peak, with a delay of only 3.5 days for *C. septempunctata* but 15.8 days for *H. axyridis*.

The sampling method used did not allow data on ladybird pupae to be collected but visual observations and collecting pupae manually in order to study the parasitism of *C. septempunctata* and *H. axyridis* in potato were performed at the same time as the beating sampling in 2005. Pupae were identified by the presence or absence of spines at the base and identification was confirmed when the adults emerged. There was evidence that

Table 1 Date of first larva detection, peak larval population and delay between aphid and ladybird larval population peaks in 8 potato in Belgium (3 in 2004, 3 in 2005 and 2 in 2006). Numbers in the same columns followed by different letters are different at $P = 0.05$ level

Ladybird species	Occurence (2004, 2005, 2006)	First larval detection	Peak larval population	Peak larval – Peak aphid population
C. 7-punctata	3, 3, 2	7.1 July (a)	16.4 July (a)	3.5 days (a)
P. quaturodecimpunctata	3, 3, 2	8.0 July (a)	19.4 July (ab)	6.5 days (ab)
A. bipunctata	3, 2, 0	15.7 July (b)	22.5 July (bc)	9.6 days (bc)
H. axyridis	3, 3, 0	15.1 July (b)	28.7 July (c)	15.8 days (c)
F		7.62	7.42	7.42
df		3, 7	3, 7	3, 7
P		0.0022	0.0025	0.0025

Table 2 Date of first adult detection, peak adult population and delay between peak aphid and ladybird adult population in potato in 8 potato in Belgium (3 in 2004, 3 in 2005 and 2 in 2006). Numbers in the same columns followed by different letters are different at $P = 0.05$ level

Ladybird species	Occurence (2004, 2005, 2006)	First adult detection	Peak adult population	Peak adult—Peak aphid population
C. 7-punctata	3, 3, 2	8.4 July (a)	18.8 July (a)	24.3 days (a)
P. quatuordecimpunctata	3, 3, 2	8.1 July (a)	19.4 July (a)	24.9 days (a)
A. bipunctata	3, 2, 0	15.3 July (a)	24.6 July (a)	30.0 days (a)
H. axyridis	3, 3, 2	15.9 July (a)	29.0 July (a)	32.0 days (a)
F		2.30	2.14	1.62
df		3, 7	3, 7	3, 7
P		0.1115	0.1306	0.2198

H. axyridis larvae reached the pupa stage in a same extend as *C. septempunctata*, with 134 *C. septempunctata* and 83 *H. axyridis* pupae collected at Nivelles on 3 August 2005, and 36 *C. septempunctata* and 14 *H. axyridis* larvae collected by beating in the same field one week earlier.

The same trends as for larvae results were observed for the adult ladybird samplings, with an apparent late detection date of *H. axyridis* and a longer delay between peak aphid and adult ladybird populations compared to *C. septempunctata* and *P. quatuordecimpunctata* (Table 2). However, the statistical analysis conclude that they were no differences concerning adult first detection date, adult peak population date and delay between peak aphid and adult ladybird population (see Table 1 for details). This can be explain by the fact that several adult catches did not correlate with the larva data in terms of dates and probably related to adult ladybirds searching for food and not becoming successfully established in the visited field. The direct consequence of this is an increase of the data variability that made the statistical analysis of the adult observations more difficult than for larvae.

The relative abundance of the different species is illustrated in Fig. 4 (2004) and Fig. 5 (2005). The minimum and maximum records of larvae were 9 (10 August) and 59 (20 July) in 2004 and 8 (16 August) and 125 (26 July) in 2005. The minimum and maximum records of adults were 5 (6 July) and 40 (27 July) in 2005 and 16 (16 August) and 69 (26 July) in 2005. At the beginning of July, only *C. septempunctata* larvae and *P. quatuordecimpunctata* larvae were found. Subsequently, *A. bipunctata* and *H. axyridis* arrived. The relative abundance of *H. axyridis* increased to a point where it completely dominated the other species by the end of the aphid potato season in August, when no aphids were observed. The results with the adults were less clear, probably because of the possible 'background noise' in adult catches and the low adult densities in August.

Discussion

This 3-year study showed that the harlequin ladybird, *H. axyridis*, has become successfully established in potato fields in Belgium and that the first detection in 2003 (Jansen and Warnier 2004) was not accidental. The first *H. axyridis* larvae were detected, on average, 7–8 days later than *C. septempunctata* and *P. quatuordecimpunctata*, the two most important species found in those fields. Compared to these two native species, the

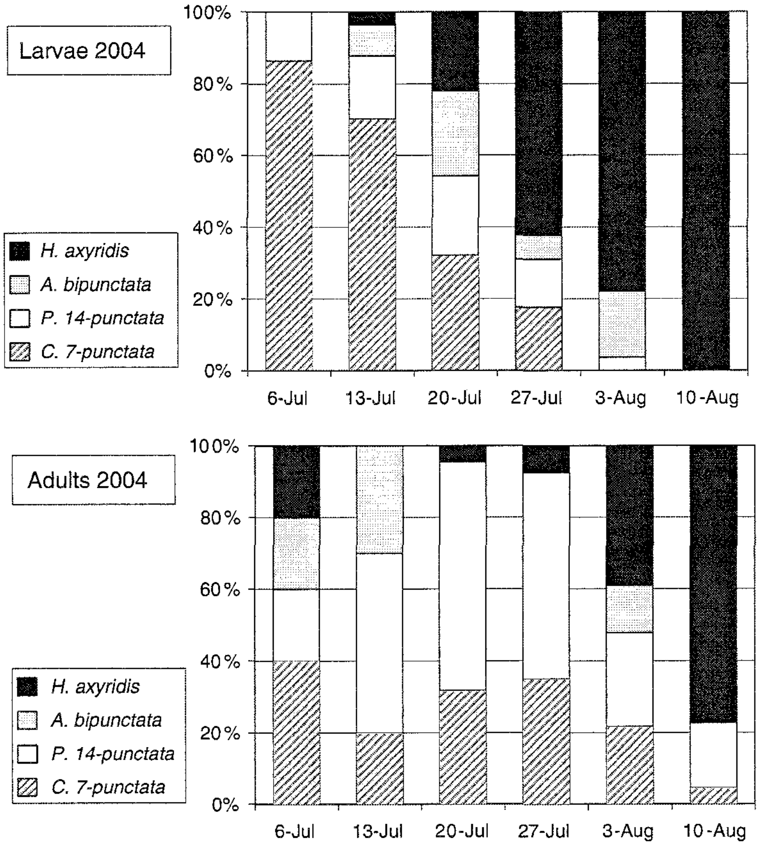

Fig. 4 Relative abundance of ladybird larvae and adults sampled in potato fields in 2004 (sum of Corroy-le-Chateau, Gembloux and Nivelles)

H. axyridis larval population peak occurred 12 and 9 days later, respectively. As aphid resources are very limited in time (Jansen 2005), the harlequin ladybird is not at all synchronised with aphids and that the main preimaginal population development occurred with very few aphids. This suggests that *H. axyridis* is able to complete its larval development feeding on prey other than aphids. This has been observed in the USA where *H. axyridis* larvae were found in great numbers in potato fields where they were supposed to feed on Colorado beetle larvae (Nault and Kennedy 2003), and in Japan on *Hibiscus* plants (Hironori and Katsuhiro 1997). In terms of the phenology of *H. axyridis*, *C. septempunctata* and *P. quatuordecimpunctata* appear to be the prey and *H. axyridis* the predator, with a delay in arrival times and peak population levels between prey and predator. This hypothesis is also supported by the fact that no *H. axyridis* larvae were collected in 2006, when the population levels of European species were much lower than in 2004 and 2005, due to low aphid populations.

Many laboratory studies clearly show that *H. axyridis* larvae are able to reach the imago stage only when the ladybird larvae of other species are available as food, such as *C. septempunctata* and *A. bipunctata* (Yasuda and Ohnuma 1999; Yasuda et al. 2001; Sato and Dixon 2004). When placed in competition, *H. axyridis* easily dominates *A. bipunctata*

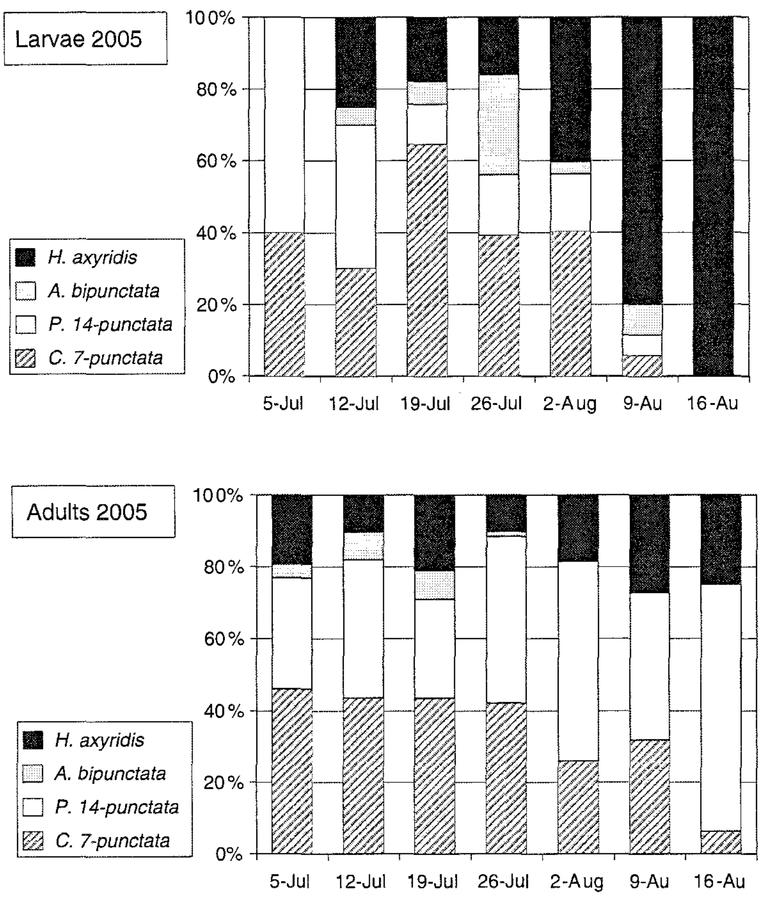

Fig. 5 Relative abundance of ladybird larvae and adults sampled in potato fields in 2005 (sum of Florennes, Gembloux and Nivelles)

in terms of direct predation and competing for food resources (Hautier 2003). The hypothesis that *H. axyridis* acts as an intraguild predator rather than an aphid predator has often been put forward but never completely verified in situ. Although intraguild predation was regularly observed in the field during this study and in previous studies (Hironori and Katsuhiro 1997; Nault and Kennedy 2003), no real assessment of the consequences of this predation was carried out because of the lack of quantification techniques in practical conditions.

Another explanation for the late arrival of *H. axyridis* is that it is known to be principally an arboreal species, at least in its area of origin (Iablokoff-Khnzorian 1982), and is commonly found on trees in Belgium, as indicated by the records on urban trees (San Marin 2003; Ottart 2005). One hypothesis is that *H. axyridis* colonizes herbaceous plants, as potato and wheat fields for example, only when it cannot find food resources on trees. This hypothesis is strengthened by the fact that *A. bipunctata*, generally considered as an arboreal species with the ability to extend to other ecosystems when there is a lack of food (Iperti 1965; Iablokoff-Khnzorian 1982), arrives at the same time as *H. axyridis*, as indicated by the results of this study. The great difference between *A. bipunctata* and

H. axyridis is that the developmental success of *A. bipunctata* when the aphid population is low or null is limited.

Whatever the reason for the late arrival of *H. axyridis* in potato fields compared with indigenous species, the direct consequence is the same: *H. axyridis* has to complete its development with very low aphid populations present, or even with no aphids at all, and it is able to do it successfully. The principal question, therefore, is what do *H. axyridis* larvae feed on? As very few insects other than ladybirds were observed in the potato fields at the end of July and the beginning of August and no alternative food resources such as pollen were available, conspecific and intraguild predation on the last indigenous ladybird larvae and pupae seems to be the most realistic hypothesis. The long-term consequences of this could be dramatic for the European species. However, no clear evolution of the dominance of *H. axyridis* was observed during this 3-year study.

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