Entomologia
Experimentalis et Applicata

DOI: 10.1111/j.1570-7458.2011.01141.x

Effect of prior diet on consumption and digestion of prey and non-prey food by adults of the generalist predator *Coleomegilla maculata*

Donald C. Weber[1]* & Jonathan G. Lundgren[2]

[1]*USDA Agricultural Research Service, Invasive Insect Biocontrol & Behavior Laboratory, BARC-West Building 011A, Room 107, Beltsville, MD 20705, USA, and* [2]*USDA Agricultural Research Service, North Central Agricultural Research Laboratory, 2923 Medary Avenue, Brookings, SD 57006, USA*

Accepted: 23 May 2011

Key words: biological control, diet switching, *Leptinotarsa decemlineata*, omnivory, pollen, predation, quantitative PCR, *Zea mays*, Coleoptera, Coccinellidae, Chrysomelidae, preconditioning

Abstract

Insect omnivores may vary in their diets and digestion based on extrinsic and intrinsic factors, including gender and nutritional history. Here, we test two hypotheses involving an insect omnivore, *Coleomegilla maculata* DeGeer (Coleoptera: Coccinellidae), feeding on prey [eggs of *Leptinotarsa decemlineata* (Say) (Coleoptera: Chrysomelidae)] and non-prey [pollen of maize, *Zea mays* L. (Poaceae)] foods: (1) males and females consume different quantities of prey and non-prey food within a set period of time, and they digest these two foods at varying rates; and (2) dietary experience (prey vs. non-prey) affects the subsequent digestion of adults consuming these foods. Adults fed eggs or pollen did not ingest different quantities of food, although females consumed marginally more pollen than males, and males consumed marginally more eggs than females. Digestion rates, as measured by decline of quantitative PCR marker in the predators, were not significantly different for males and females, and corresponded to a quantitative half-life of 56 min for pollen and 46 min for eggs. But when newly eclosed females were fed with only prey for 7 days, they subsequently did not measurably digest non-prey food over 8 h, compared with females fed previously on pollen, which digested it with an estimated half-life of 45 min. Thus, feeding experience with some prey may cause changes in the digestive system of the predator, which later impair digestion of non-prey foods such as pollen by omnivores. This may have implications for survival and reproduction of omnivorous natural enemies released into the field or diet-switching associated with movement among habitats.

Introduction

Many insect predators consume non-prey foods in addition to prey, and these non-prey foods may be essential to their survival, reproduction, and function as biological control agents (Lundgren, 2009). The degree to which a species can benefit from ingestion of prey and non-prey foods is highly dependent on its ability to digest them. The North American lady beetle *Coleomegilla maculata* DeGeer (Coleoptera: Coccinellidae) is an extremely polyphagous coccinellid; it can complete development and reproduce not only on a variety of prey, but also on various non-prey foods, including pollen of maize, *Zea mays* L. (Poaceae) (Lundgren & Wiedenmann, 2004; Michaud & Grant, 2005). The ability to digest different foods depends on the physiological capability of the ingesting insect, which in turn depends on intrinsic factors (such as age, sex, and genetics) and extrinsic factors (such as temperature and previous diet) (Chapman, 1998).

Foraging patterns often differ between sexes of arthropods, but the basis for this is poorly understood (Nakashima & Hirose, 2003; Stillwell et al., 2010). In coccinellids, adult males generally forage less and consume less food than females (Honěk, 1985; Johki et al., 1988; Dixon, 2000; Yasuda & Dixon, 2002). It is not clear, however, whether digestion of various foods (prey and non-prey) differs between the sexes.

*Correspondence: Donald C. Weber, USDA Agricultural Research Service, Invasive Insect Biocontrol & Behavior Laboratory, BARC-West Building 011A, Room 107, Beltsville, MD 20705, USA.
E-mail: Don.Weber@ars.usda.gov

No claim to original US government works

Entomologia Experimentalis et Applicata © 2011 The Netherlands Entomological Society

Polyphagous predators likely switch diets frequently under natural conditions, but the contributions of previous diet to their ability to consume new or different foods remain poorly understood, especially for omnivorous insects. The majority of studies on diet switching have focused on vertebrates (Starck, 2003; Battley & Piersma, 2005) and herbivorous insects (Stoyenoff et al., 1994; Chambers et al., 1998; Bernays et al., 2004; Behmer, 2009). For polyphagous herbivores, prior diet (so-called preconditioning) has long been recognized as strongly influencing bioassays for crop plant resistance (Smith, 2005). For insect herbivores, diet mixing and switching seem to be influenced both by nutrient regulation and by top-down risks such as predation (Chambers et al., 1998; Behmer, 2009), but trade-offs are generally not well documented, and detailed feeding behaviors of generalists and specialists differ (Bernays et al., 2004). Diet switching may be accompanied by rapid digestive adaptations, depending on the consumer species and the foods involved, especially if they are quite different in composition. For instance, passerine birds show radical changes in gut structure and function after a few days when switching between insect and fruit diets (Levey & Karasov, 1989), but these effects may be asymmetric, depending on the relative nutrient contents (Afik & Karasov, 1995; Lee & Houston, 1995; Starck, 1999; Hilton et al., 2000). Furthermore, the timescale required for digestive adjustments to changes in food intake and quality is poorly understood (McWilliams & Karasov, 2001, 2005). The duration and magnitude of fitness costs for switching between diets strongly influences advantages of omnivory which accrue from increased availability of food (Whelan et al., 2000).

Quantitative polymerase chain reaction (qPCR) provides a useful tool to determine ingestion and digestion of prey and non-prey diets containing known marker sequences of DNA. Laboratory study of predation using qPCR has shown that disappearance of prey DNA after ingestion by third instars of *C. maculata* is rapid, with a quantitative half-life ranging from 16 to 59 min, depending on subsequent diet (Weber & Lundgren, 2009a). Larval age affects the digestion rate of various foods in this species (Lundgren et al., 2005; Lundgren & Weber, 2010). For adult *C. maculata*, longevity and mobility promote radical changes in diet over time, with possible switches to and from prey and non-prey foods based on availability and preference. Diet switching within the adult stage, and whether previous diet can influence preference, and therefore ingestion and/or digestion, has not been examined for *C. maculata*.

Here, we present experiments on feeding and digestion by *C. maculata*, using representative prey and non-prey foods, Colorado potato beetle [*Leptinotarsa decemlineata* (Say) (Coleoptera: Chrysomelidae) (CPB)] eggs and maize pollen, respectively. First, we test the hypothesis that adult female *C. maculata* consume more food and digest it faster than adult males do. Second, we test under no-choice conditions, whether ingestion and digestion rates of a food by *C. maculata* are faster when the beetles have experience digesting it, than when their prior diet is different.

Materials and methods

Test insects and diets

Coleomegilla maculata adults were collected from maize fields in Beltsville (MD, USA; 39°02′N, 76°56′W) and maintained in culture for 6 months prior to experiments. Larvae were reared on a 1:1 mixture (by weight) of Bee Pro pollen substitute (Mann Lake, Hackensack, MN, USA) and macerated dried freshwater amphipods (*Gammarus lacustris* Sars; Tetra Holding, Blacksburg, VA, USA). *Leptinotarsa decemlineata* were collected from potato fields in Beltsville and maintained on potato plants for at least ca. 8 months prior to experiments. Egg clutches between 24 and 48 h old (at 25 °C) were separated from the potato foliage using a fine paint brush for feeding to individual predators. Maize (NK4242; Northrup King Company, Golden Valley, MN, USA) plants were grown in 7.6-l pots (two plants per pot) in the greenhouse until anthesis, at ca. 27 °C, 40% r.h., and L14:D10 photoperiod. Plants were fertilized with 20-20-20 N-P-K (Plantex; Plant Product, Brampton, ON, Canada) and 1.26 ml of chelated Fe (Sequestrene 330 Fe; Becker Underwood, Ames, IA, USA) per liter water. Pollen was collected, sieved (mesh size: 55.6 openings cm^{-1}), and stored according to protocols outlined by Pilorget et al. (2010).

Experiment 1. Sex differences in ingestion and digestion of eggs or pollen

Newly eclosed *C. maculata* adults were held individually in 4-cm-diameter plastic Petri dishes, provided with a 20% (wt/wt) water solution of sucrose for 7 days, then starved (water was provided as a saturated cotton wick) for 24 h prior to experimentation. Each adult was observed (microscopically in the case of pollen) feeding for 5 min without interruption. Test animals (mean of 10 replicates per time per sex) were then sacrificed at 0, 15, 30, 60, 120, 240, and 480 min and placed immediately in 70% ethanol at −20 °C (Weber & Lundgren, 2009a). Sex was confirmed by dissection prior to DNA extraction.

Experiment 2. Effects of preceding diet on food ingestion and digestion

Newly eclosed females were held individually in 4-cm-diameter plastic Petri dishes, fed 7 days ad libitum

exclusively either on pollen or eggs, and also provided with a water-soaked cotton wick. Then, for each of these two initial (prior) diets, adults were starved for 24 h (water only on soaked cotton wick) and then fed for 5 min as described for Experiment 1, on eggs or pollen, resulting in two crossover (switched) diet treatments and two control (unswitched) diet treatments. Test animals (on average 8.25 ± 0.26 replicates per time per treatment) were again sacrificed at 0, 15, 30, 60, 120, 240, and 480 min and placed immediately in 70% ethanol at −20 °C. Sex was confirmed by dissection prior to DNA extraction.

DNA extraction and amplification

DNA was extracted from individual *C. maculata* test adults using DNeasy Blood and Tissue extraction kits (#69506; Qiagen, Valencia, CA, USA). Samples (macerated whole individual adults with elytra and legs removed) were incubated in ATL buffer with Proteinase K for 3 h. Food-specific primer sets were as in Lundgren & Weber (2010): the 214 bp sequence of the COI gene of *L. decemlineata* (sequenced by Greenstone et al., 2007) and a 141 bp sequence from the COI gene of *Z. mays* (sequenced by Xiao et al., 2006). Marker size under 300 bp has been considered appropriate for predation studies (King et al., 2008). The amount of food DNA present in each *C. maculata* was quantified using qPCR on a Stratagene MX3000P thermocycler (Stratagene, La Jolla, CA, USA) in 25-μl reactions with the following ingredients: 9.5 μl PCR-grade water, 12.5 μl Quantitect SYBR Green PCR Master Mix (#204143; Qiagen), 1 μl template DNA (ca. 1–88 ng DNA; assessed using absorbance ratio produced at 260/280 nm), and 1 μl each of forward and reverse food-specific primer sets, both at concentrations of 225 nM. Amplification conditions were a single cycle of 95 °C, followed by 45–55 cycles of 94 °C for 15 s, 54 or 56 °C (for egg and pollen, respectively) for 30 s, and 74 °C for 30 s. To ensure that our PCR was amplifying the correct DNA, a food-specific dissociation (melting) temperature was determined for each PCR product by incubation at 95 °C for 60 s, then dropping the temperature to 55 °C and ramping up to 94 °C, monitoring fluorescence every 0.5 °C; the PCR product for *L. decemlineata* dissociates unimodally at 74.3 °C, and that for *Z. mays* at 77.7 °C. The final result of these PCRs was a Cycle threshold (Ct), the minimum number of PCR cycles necessary for the sample's fluorescence to be detected above background levels. For each value in which the Ct of a sample exceeded 45 cycles, a randomly selected value $45<Ct<55$ was generated. Cycle thresholds were determined for each test insect, along with positive food controls (five wells per plate), DNA from unfed *C. maculata* (three wells), and no-template (three wells) controls. These controls were extractions of the following material: three 3-day-old *L. decemlineata* eggs and 22 mg *Z. mays* pollen.

Data analysis

Analysis of covariance (ANCOVA; Milliken & Johnson, 2002) was performed on each of the two experiments using linear modeling of log-transformed estimates of quantity of food marker over time, by comparison with the positive food controls. The linear model of log-transformed data corresponds to the expected exponential decay rate for the DNA markers over time (Weber & Lundgren, 2009a). This model is consistent with the expectation that a constant proportion of target disappears per unit time, and that relatively small markers will persist longer regardless of absolute half-life (Deagle et al., 2006). For each experiment, the following hypotheses were tested: (1) Do beetles consume food at the same rate over the 5-min feeding period, i.e., does the detected amount at $t = 0$ differ by treatment? (2) Do the beetles digest each food they consume within 8 h, i.e., is the slope of the linear regression significantly different from zero? (3) Do sex or food type affect how fast food is digested, i.e., do the slopes differ among treatments in Experiment 1? and (4) Does initial (prior) diet affect the rate of digestion, i.e., do the slopes differ among treatments in Experiment 2?

These tests were carried out using Proc GLM (SAS, version 9.2; SAS Institute, 2008) according to the ANCOVA hypotheses of Milliken & Johnson (2002, chapter 2; time in this context is the covariate), using $\alpha = 0.05$. Data were compared within foods, but not between eggs and pollen, because of the different nature of the living tissue ingested and the variable rate at which different-sized amplicons are digested.

Results

Experiment 1. Sex differences in feeding

Male and female adult *C. maculata* did not differ significantly in the initial quantity of food ingested, nor in the digestion rate over 8 h (quantity ingested: $F_{1,136} = 0.51$, $P = 0.48$ for eggs; $F_{1,136} = 0.33$, $P = 0.57$ for pollen; digestion rate: $F_{1,136} < 0.01$, $P = 0.95$ for eggs; $F_{1,136} = 1.08$, $P = 0.30$ for pollen). The estimated mean quantitative half-lives for egg and pollen digestion were 46 and 56 min, respectively (Table 1).

Experiment 2. The effect of preceding diet on ingestion and digestion

Initial (prior) diet significantly influenced the rate of digestion of pollen as a test diet: females reared on *L. decemlineata* eggs digested pollen more slowly than females previously fed on pollen (digestion rate: $F_{1,110} = 4.09$, $P = 0.046$; Table 1, Figure 1). Females

Table 1 Estimated ingestion and digestion parameters for *Coleomegilla maculata* fed *Leptinotarsa decemlineata* eggs and *Zea mays* pollen. Predators were fed the initial food for 7 days, and then switched to the test diet, digestion rates of which were assessed using qPCR and primer sets specific for test foods over 8 h. Half-life is the time over which the linear regression decreases by one-half, according to the estimated slope of the linear regression of log-transformed marker quantities determined by qPCR

Initial food	Test food	Sex	Intercept ± SE	Slope ± SE[1]	Half-life (min)
Experiment 1					
Sucrose	Egg	Male	−0.09 ± 0.33	−0.38 ± 0.09	47.0
Sucrose	Egg	Female	−0.41 ± 0.31	−0.39 ± 0.09	45.9
Sucrose	Pollen	Male	−1.17 ± 0.22	−0.37 ± 0.06	48.3
Sucrose	Pollen	Female	−1.00 ± 0.20	−0.29 ± 0.06	63.1
Experiment 2					
Pollen	Pollen	Female	−1.82 ± 0.32	−0.40 ± 0.09a	44.9
Egg	Pollen	Female	−2.12 ± 0.36	−0.13 ± 0.10b[2]	140.7
Egg	Egg	Female	−0.43 ± 0.34	−0.36 ± 0.10	49.6
Pollen	Egg	Female	−0.43 ± 0.29	−0.35 ± 0.08	52.2

[1]Different letters indicate significantly different digestion rates (slopes) of a given test food observed for paired treatments ($P<0.05$).

[2]The slope of the pollen following egg treatment did not differ significantly from zero ($t = -1.30$, $P = 0.20$; $n = 52$). All other digestion rates (slopes) differ significantly from zero ($P<0.001$ for all seven treatments).

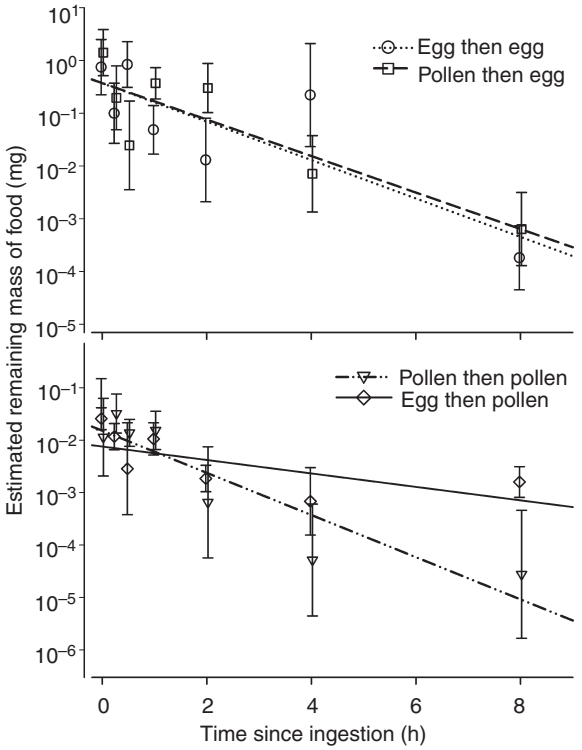

Figure 1 Digestion of food-specific DNA sequences by *Coleomegilla maculata* adult females, when fed *Leptinotarsa decemlineata* eggs or *Zea mays* pollen following the same or different food (logarithmic means ± SE). For the treatment of 'egg then pollen', the slope is not significantly different from zero, and it differs from the slope for the diet of 'pollen then pollen'. See Table 1 and text for statistical details.

reared on *L. decemlineata* eggs did not digest pollen measurably (i.e., the slope of the digestion regression was not significantly different from zero; $t = -1.30$, $P = 0.20$, $n = 52$). Pollen-reared females fed pollen digested half the pollen DNA within an estimated 45 min of cessation of feeding. *Leptinotarsa decemlineata* eggs were consumed and digested at similar rates regardless of the diet that they were reared on. The estimate of quantitative half-life for *L. decemlineata* DNA was 51 min for *C. maculata* previously fed eggs or pollen, similar to that for the sucrose-fed insects in Experiment 1. Estimated ingestion rates did not differ between the treatments.

Discussion

Preceding diet affected pollen digestion, to the extent that prey-fed females did not effectively digest pollen when it suddenly became available, whereas females fed pollen for 7 days had the ability to digest this food efficiently. This was a significant and unidirectional dietary conditioning phenomenon in the diet crossover experiment with *C. maculata*: insects failed to digest a subsequent maize pollen meal at a rate comparable with females previously fed pollen. Yet females fed only sucrose in the first experiment digested pollen at a similar rate to those which were previously pollen fed. This appears to be an example of negative dietary conditioning, in which prey consumption in this case diminishes the quality or quantity of enzymes that digest resistant pollen components which protect the DNA within the maize pollen. The dynamics of digestion of pollen are not well known for insects that ingest pollen grains whole, as is true for all beetles (Roulston & Cane, 2000). Digestion can include osmotic shock, germination, pseudo-germination, exudation, and enzyme penetration and may involve contributions from microbial symbionts (Roulston & Cane, 2000; Johnson & Nicolson, 2001). The pollen coat (exine) is typically a resistant barrier to

digestion, which requires particular adaptations to release the contents of the pollen grain. Thus, the presence of effective enzymes, even if in combination with other pollen digestion mechanisms, is likely a critical aspect of pollen digestion in pollen-feeding beetles studied (Johnson & Nicolson, 2001; Human & Nicolson, 2003; Lundgren, 2009). The fact that pollen-fed females were able to digest it well suggests that they have a more effective complement of pollen-digesting enzymes, either their own or possibly those which originate in the diet itself (Lundgren, 2009).

Male and female *C. maculata* ingested and digested foods at similar rates. Although the differences were not significant, females consumed approximately twice as much pollen as males and males consumed slightly more eggs (logarithmic means). The feeding period of 5 min was likely too short to show differences in ingestion which have been found over longer periods. For periods of several days, Hazzard & Ferro (1991) found that female *C. maculata* consumed 50% more eggs over 24 h, and Lundgren et al. (2005) found ca. 10× more maize pollen in female than in male *C. maculata* collected from a maize field during anthesis. The similar digestive rates of these foods in males and females reflect the inherent omnivory of both sexes of this species, and further suggests that observations on sex-specific differences in pollen consumption rates are likely driven by foraging behavior rather than physiological differences between the sexes.

Among arthropod predators, omnivory is now recognized to be very frequent, including consumption of non-prey foods such as pollen, fungi, plant parts, and nectar (Coll & Guershon, 2002; Lundgren, 2009; Weber & Lundgren, 2009b). By definition, this entails diet mixing and diet switching or shifting over time, based on availability and selection of food items by the individual predator (Chambers et al., 1998; Singer & Bernays, 2003; Behmer, 2009). In the field, plants provide food and other resources which frequently mediate interactions between omnivores and their prey and non-prey foods (Eubanks & Styrsky, 2005). For example, pollen from a single species can be extremely abundant, but usually only for a short time during the year (Lundgren & Wiedenmann, 2004); similarly, specific prey populations (e.g., aphids and insect eggs) are often plentiful but ephemeral resources (Voss & Ferro, 1989; Michaud & Jyoti, 2008). The result is that individual predators likely have to shift their diets over their adult lives, and may encounter a large number of potential food items, particularly if the predator moves from habitat to habitat, or with resource pulses (Yang et al., 2008). *Coleomegilla maculata* balances its diet between insect prey and non-prey foods, and populations alter their reliance on selected prey items when alternative foods (e.g., pollen or other prey types) become available (Hazzard & Ferro, 1991; Cottrell & Yeargan, 1998; Lundgren et al., 2004). These spatiotemporal patterns of food availability demand and select for mobility and/or physiological adaptability to ingestion and digestion of a wide range of foods. Quality or value of the different foods available to omnivores, however, depends on their ability to effectively digest them, which in turn may depend on the pattern of diet switching and mixing: the order and duration in which different foods are encountered and ingested. For biological control by omnivorous predators, knowledge of the implications of prior diet and subsequent diet switching is essential to the optimal functional and numeric response of biocontrol agents in conservation, augmentative, and inundative approaches.

Acknowledgements

We thank Michael Athanas and Meiling Webb for predator rearing and experimental assistance, and Janet Fergen for assisting with DNA extraction and amplification. We also thank the two anonymous reviewers for their helpful suggestions. Mention of trade names or commercial products is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the USDA.

References

Afik D & Karasov WH (1995) The trade-offs between digestion rate and efficiency in warblers and their ecological implications. Ecology 76: 2247–2257.

Battley PF & Piersma T (2005) Adaptive interplay between feeding ecology and features of the digestive tract in birds. Physiological and Ecological Adaptations to Feeding in Vertebrates (ed. by JM Starck & T Wang), pp. 201–228. Science Publishers, Enfield, NH, USA.

Behmer ST (2009) Insect herbivore nutrient regulation. Annual Review of Entomology 54: 165–187.

Bernays EA, Singer MS & Rodrigues D (2004) Foraging in nature: foraging efficiency and attentiveness in caterpillars with different diet breadths. Ecological Entomology 29: 389–397.

Chambers PG, Raubenheimer D & Simpson SJ (1998) The functional significance of switching interval in food mixing by *Locusta migratoria*. Journal of Insect Physiology 44: 77–85.

Chapman RF (1998) The Insects: Structure and Function. Cambridge University Press, Cambridge, UK.

Coll M & Guershon M (2002) Omnivory in terrestrial arthropods: mixing plant and prey diets. Annual Review of Entomology 47: 267–297.

Cottrell TE & Yeargan KV (1998) Effect of pollen on *Coleomegilla maculata* (Coleoptera: Coccinellidae) population density, predation, and cannibalism in sweet corn. Environmental Entomology 27: 1402–1410.

Deagle BE, Eveson JP & Jarman SN (2006) Quantification of damage in DNA recovered from highly degraded samples – a case study on DNA in faeces. Frontiers in Zoology 3: 11.

Dixon AFG (2000) Insect Predator-Prey Dynamics: Ladybird Beetles and Biological Control. Cambridge University Press, Cambridge, UK.

Eubanks MD & Styrsky JD (2005) Effects of plant feeding on the performance of omnivorous 'predators'. Plant-Provided Foods for Carnivorous Insects (ed. by FL Wäckers, PCJ van Rijn & J Bruin), pp. 148–177. Cambridge University Press, Cambridge, UK.

Greenstone MH, Rowley DL, Weber DC, Payton ME & Hawthorne DJ (2007) Feeding mode and prey detectability half-lives in molecular gut analysis: an example with two predators of the Colorado potato beetle. Bulletin of Entomological Research 97: 201–209.

Hazzard RV & Ferro DN (1991) Feeding responses of adult *Coleomegilla maculata* (Coleoptera: Coccinellidae) to eggs of Colorado potato beetle (Coleoptera: Chrysomelidae) and green peach aphids (Homoptera: Aphididae). Environmental Entomology 20: 644–651.

Hilton GM, Furness RW & Houston DC (2000) The effects of diet switching and mixing on digestion in seabirds. Functional Ecology 14: 145–154.

Honěk A (1985) Activity and predation of *Coccinella septempunctata* in the field (Col., Coccinellidae). Zeitschrift für Angewandte Entomologie 100: 399–409.

Human H & Nicolson SW (2003) Digestion of maize and sunflower pollen by the spotted maize beetle *Astylus atromaculatus* (Melyridae): is there a role for osmotic shock? Journal of Insect Physiology 49: 633–643.

Johki Y, Obata S & Matsui M (1988) Distribution and behaviour of five species of aphidophagous ladybirds (Coleoptera) around aphid colonies. Ecology and Effectiveness of Aphidophaga (ed. by E Niemczyk & AFG Dixon), pp. 35–38. SPB Academic Publishing, The Hague, The Netherlands.

Johnson SA & Nicolson SW (2001) Pollen digestion by flower-feeding Scarabaeidae: protea beetles (Cetoniini) and monkey beetles (Hopliini). Journal of Insect Physiology 47: 725–733.

King RA, Read DS, Traugott M & Symondson WOC (2008) Molecular analysis of predation: a review of best practice for DNA-based approaches. Molecular Ecology 17: 947–963.

Lee WB & Houston DC (1995) The rate of change of gut anatomy in voles in relation to diet quality. Journal of Zoology 236: 341–345.

Levey DJ & Karasov WH (1989) Digestive responses of temperate birds switched to fruit or insect diets. The Auk 106: 675–686.

Lundgren JG (2009) Relationships of Natural Enemies and Non-Prey Foods. Springer, Dordrecht, The Netherlands.

Lundgren JG & Weber DC (2010) Changes in digestive rate of a predatory beetle over its larval stage: implications for dietary breadth. Journal of Insect Physiology 56: 431–437.

Lundgren JG & Wiedenmann RN (2004) Nutritional suitability of corn pollen for the predator *Coleomegilla maculata* (Coleoptera: Coccinellidae). Journal of Insect Physiology 50: 567–575.

Lundgren JG, Razzak AA & Wiedenmann RN (2004) Population responses and food consumption by predators *Coleomegilla maculata* and *Harmonia axyridis* (Coleoptera: Coccinellidae) during anthesis in an Illinois cornfield. Environmental Entomology 33: 958–963.

Lundgren JG, Huber A & Wiedenmann RN (2005) Quantification of consumption of corn pollen by the predator *Coleomegilla maculata* (Coleoptera: Coccinellidae) during anthesis in an Illinois cornfield. Agricultural and Forest Entomology 7: 53–60.

McWilliams SR & Karasov WH (2001) Phenotypic flexibility in digestive system structure and function in migratory birds and its ecological significance. Comparative Biochemistry and Physiology Part A 128: 579–593.

McWilliams SR & Karasov WH (2005) Migration takes guts – digestive physiology of migratory birds and its ecological significance. Birds of Two Worlds (ed. by R Greenberg & PP Marra), pp. 67–78. Smithsonian Institution Press, Washington, DC, USA.

Michaud JP & Grant AK (2005) Suitability of pollen sources for the development and reproduction of *Coleomegilla maculata* (Coleoptera: Coccinellidae) under simulated drought conditions. Biological Control 32: 363–370.

Michaud JP & Jyoti JL (2008) Dietary complementation across life stages in the polyphagous lady beetle *Coleomegilla maculata*. Entomologia Experimentalis et Applicata 126: 40–45.

Milliken GA & Johnson DE (2002) Analysis of Messy Data, Volume III: Analysis of Covariance. Chapman & Hall/CRC, New York, NY, USA.

Nakashima Y & Hirose Y (2003) Sex differences in foraging behaviour and oviposition site preference in an insect predator, *Orius sauteri*. Entomologia Experimentalis et Applicata 106: 79–86.

Pilorget L, Buckner J & Lundgren JG (2010) Sterol limitation in a pollen-fed omnivorous lady beetle (Coleoptera: Coccinellidae). Journal of Insect Physiology 56: 81–87.

Roulston TH & Cane JH (2000) Pollen nutritional content and digestibility for animals. Plant Systematics and Evolution 222: 187–209.

SAS Institute (2008) SAS for Windows (version 9.2). SAS Institute, Cary, NC, USA.

Singer MS & Bernays EA (2003) Understanding omnivory needs a behavioral perspective. Ecology 84: 2532–2537.

Smith CM (2005) Plant Resistance to Arthropods: Molecular and Conventional Approaches. Springer, Dordrecht, The Netherlands.

Starck JM (1999) Phenotypic flexibility of the avian gizzard: rapid, reversible and repeated changes of organ size in response to changes in dietary fibre content. Journal of Experimental Biology 202: 171–179.

Starck JM (2003) Shaping up: how vertebrates adjust their digestive system to changing environmental conditions. Animal Biology 53: 245–257.

Stillwell RC, Blanckenhorn WU, Teder T, Davidowitz G & Fox CW (2010) Sex differences in phenotypic plasticity affect varia-

tion in sexual size dimorphism in insects: from physiology to evolution. Annual Review of Entomology 55: 227–245.

Stoyenoff JL, Witter JA, Montgomery ME & Chilcote CA (1994) Effects of host switching on gypsy moth (*Lymantria dispar* (L.)) under field conditions. Oecologia 97: 143–157.

Voss RV & Ferro DN (1989) Role of reproductive diapause in the population dynamics of the Colorado potato beetle. Environmental Entomology 17: 863–871.

Weber DC & Lundgren JG (2009a) Detection of predation using qPCR: effect of prey quantity, elapsed time, chaser diet, and sample preservation on detectable quantity of prey DNA. Journal of Insect Science 9: 41.

Weber DC & Lundgren JG (2009b) Assessing the trophic ecology of the Coccinellidae: their roles as predators and as prey. Biological Control 51: 199–214.

Whelan CJ, Brown JS, Schmidt KA, Steele BB & Willson MF (2000) Linking consumer–resource theory and digestive physiology: application to diet shifts. Evolutionary Ecology Research 2: 911–934.

Xiao H, Zhang F & Zheng Y (2006) The 5′ stem-loop and its role in mRNA stability in maize S cytoplasmic male sterility. Plant Journal 47: 864–872.

Yang LH, Bastow JL, Spence KO & Wright AN (2008) What can we learn from resource pulses? Ecology 89: 621–634.

Yasuda H & Dixon AFG (2002) Sexual size dimorphism in the two spot ladybird beetle *Adalia bipunctata*: developmental mechanism and its consequences for mating. Ecological Entomology 27: 493–498.