Lipid Metabolism in Parasitoids and their Parasitized Hosts

Mathilde Scheifler[1], Léonore Wilhelm[1] & Bertanne Visser[1]*

[1]Evolution and Ecophysiology group, Department of Functional and Evolutionary Entomology, University of Liège - Gembloux Agro-Bio Tech, Passage des Déportés 2, 5030 Gembloux, Belgium

Mathilde Scheifler: mathilde.scheifler@uliege.be
Léonore Wilhelm: leonore.wilhelm@orange.fr
Bertanne Visser: bertanne.visser@uliege.be
*corresponding author

Abstract

Parasitoids have an exceptional lifestyle where development is spent on or in a single host insect, but the adults are free-living. Unlike parasites, parasitoids always kill their host. How parasitoids use such a limiting resource, particularly lipids, is important for their chances to survive and reproduce. Lipid metabolism in parasitoids has been of interest to researchers already since the 60s and continues to fascinate ecologists, evolutionists, physiologists, and entomologists alike. One reason is that the majority of parasitoids do not accumulate triacylglycerols as adults. Early research revealed that some parasitoid larvae mimic the fatty acid composition of their host, which may result from a lack of *de novo* triacylglycerol synthesis. More recent work has focused on the evolution of lack of adult triacylglycerol accumulation and consequences for life history traits. In part 2 of this chapter we discuss research efforts on lipid metabolism in parasitoids from the 60s onwards. Parasitoids are master manipulators of their host's physiology, including lipid metabolism. Parasitoids have indeed evolved a range of mechanisms to affect the release, synthesis, transport, and take-up of lipids from their host. We detail the effects of parasitism on host physiology in part 3 of this chapter.

Keywords: Fat; Fitness; Host-parasitoid interaction; Parasitic wasp

1 Introduction

There are many intricacies when it comes to the fat metabolism of parasitoids. These insects have a unique lifestyle, where development takes place inside or on a single host (usually another insect or arthropod), but the adults are free-living (Godfray, 1994) (Fig. 1). During development, the parasitoid consumes the host and its nutritional resources, sometimes only partly or in its entirety in successive steps (Cuny and Poelman, 2022), ultimately leading to death of the host. Parasitoidism thus constitutes a parasitic relationship between a parasitoid and its host that differs from parasites in the extent to which the host is harmed (i.e., death), also in terms of fitness (Lafferty & Kuris, 2002). The parasitoid lifestyle evolved repeatedly in insects, including independent occurrences in beetles, flies, butterflies, and lacewings (Eggleton & Belshaw, 1992, 1993), but Hymenoptera (bees, ants, wasps, and sawflies) is the most speciose order when it comes to parasitoids (and potentially species in general, see Forbes et al., 2018). The large breath of parasitoid species worldwide, their unique lifestyle, and the plethora of strategies used by parasitoids to infect their hosts make them valuable and interesting biological model systems (Hoddle et al., 1998; Liu et al., 2015; Matthews et al., 2009; Quicray et al., 2023; Werren & Loehlin, 2009; Whitfield et al., 2017). This is true not only from a basic, fundamental scientific perspective, but also for the applied sciences, because parasitoids play a key role in regulating both natural and agricultural insect communities (i.e., as biocontrol agents against insect pests; Jervis, 2005).

This chapter starts with an overview of research done on lipid metabolism of insect parasitoids, from earlier works in the 60s to the most recent developments in part 2. Host fatty acid composition and fat content, as well as the ability of parasitoids to manipulate host lipids and their availability, is of critical importance for both immature and adult parasitoids. Part 3 focuses on the different mechanisms by which parasitoids can manipulate the fat metabolism of their hosts.

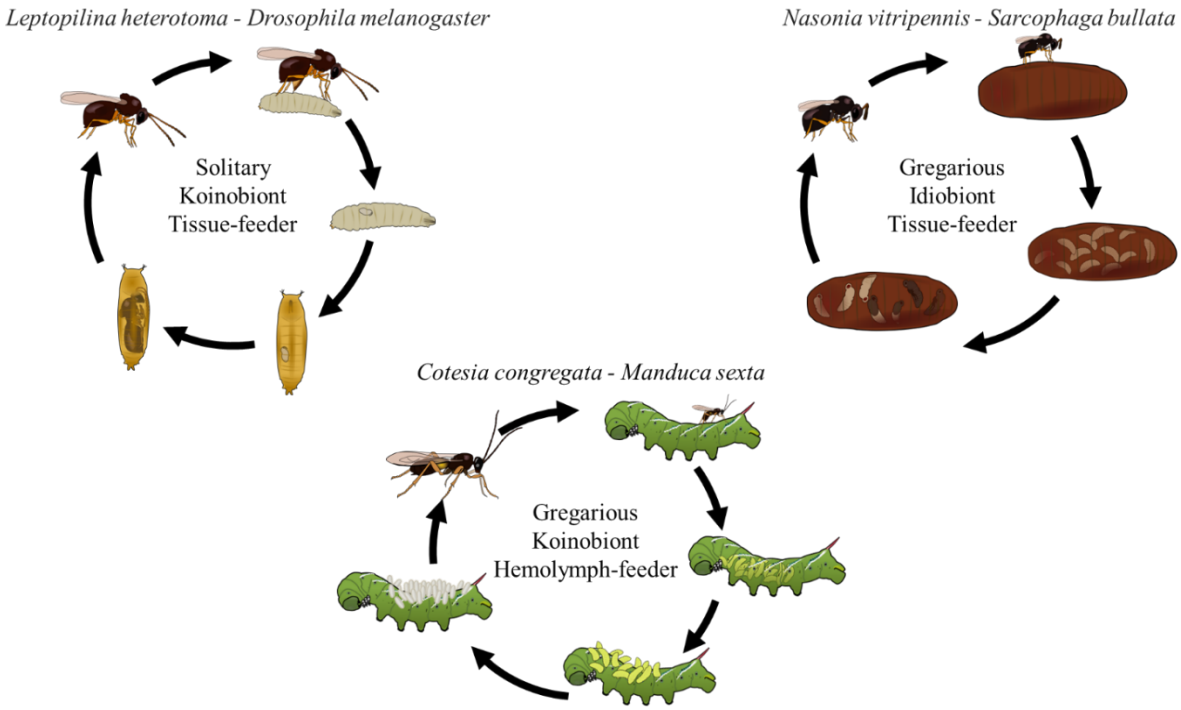

Fig. 1. The parasitoid life cycle showing the different effects on host development (idiobiont vs koinobiont), as well as parasitoid oviposition (solitary vs gregarious) and feeding strategies (hemolymph vs tissue-feeder). Represented species include the parasitoid *Leptopilina heterotoma* on the host *Drosophila melanogaster*, the parasitoid *Cotesia congregate* on the host *Manduca sexta*, and the parasitoid *Nasonia vitripennis* on the host *Sacrophaga bullata.*

2 Fatty acid synthesis and fat accumulation in parasitoids

Parasitoids have been of particular interest to biologists with respect to their lipid metabolism. There has, however, been some recent debate between researchers studying parasitoid lipid metabolism, mainly in terms of semantics (Visser et al., 2023). To avoid confusion about definitions and terminology related to lipid metabolism, we have proposed the use of stricter definitions that emphasize the difference between the processes of fatty acid synthesis and triacylglycerol/fat accumulation (Visser et al., 2023). This distinction is important, because these two processes are not synonymous with one another: fatty acids can be synthesized even if triacylglycerols are not accumulated. The main interest of evolutionary ecologists studying lipid metabolism in parasitoids has been on the accumulation of triacylglycerols in adults, because energy stored in the form of fat reserves can have a major impact on life histories and fitness (see Box 1 for a brief overview of the link between fat content and life histories in parasitoids). Parasitoids represent a curious case where triacylglycerols are generally not accumulated in response to superfluous feeding, unlike other animals that will readily accumulate triacylglycerols under the same nutritional conditions (Visser et al., 2010; Visser & Ellers, 2008; Box 1). While previously referred to as the “lack of lipogenesis” or “lack of lipid synthesis”, we now refer to this phenomenon as the “lack of adult triacylglycerol/fat accumulation” in parasitoids (Visser et al.,

2023), with fat being used synonymously with triacylglycerol. We continue part 2 of this chapter with a chronological account of the work done on lipid metabolism, with a main focus on fatty acid synthesis and fat accumulation in insect parasitoids.

Box 1. Survival of the fattest: Stored triacylglycerol levels impact longevity, reproduction, and other fitness-related traits in parasitoids

Triacylglycerols are the most comprehensive form in which energy can be stored, both in terms of the storage space needed within a cell and the higher caloric content per unit of weight (Arrese & Soulages, 2010). Oxidation of triacylglycerols further releases twice as much water compared to glycogen (i.e., the other metabolite for energy storage). Taking all this into consideration, it is not surprising that fat plays a critical role for insect life histories, including parasitoids (Jervis et al., 2008). We use earlier work on the *Drosophila*-parasitizing braconid wasp *Asobara tabida* as an exemplary case study to reveal the close link between fat reserves and life history traits (see Colinet et al., 2006; Giron & Casas, 2003a; Le Lann et al., 2014; Luo et al., 2010; Muller et al., 2017; Sheng et al., 2019 for similar findings in other parasitoid species).

Under laboratory conditions, *A. tabida* females can emerge with ~20% total body fat (Visser et al., 2010), but triacylglycerol levels never exceed those at emergence (Ellers, 1996; Le Lann et al., 2014; Visser et al., 2010), as depicted in Fig. B1A. Unlike most other insects (and animals) that rapidly start building up triacylglycerol stores when fed a surplus of sugars (see Fig. B1B for *Drosophila melanogaster* as an example; Visser et al., 2010), triacylglycerol content in *A. tabida* decreases quickly during the first week of life, and more steadily at a rate of ~0.004 mg per week thereafter (for comparison: when starved, *A. tabida* triacylglycerol-use is ~0.004 mg per day). Moreover, *A. tabida* strains with higher fat content also live longer (Ellers, 1996). Fat reserves are thus correlated with adult survival and used to fuel life.

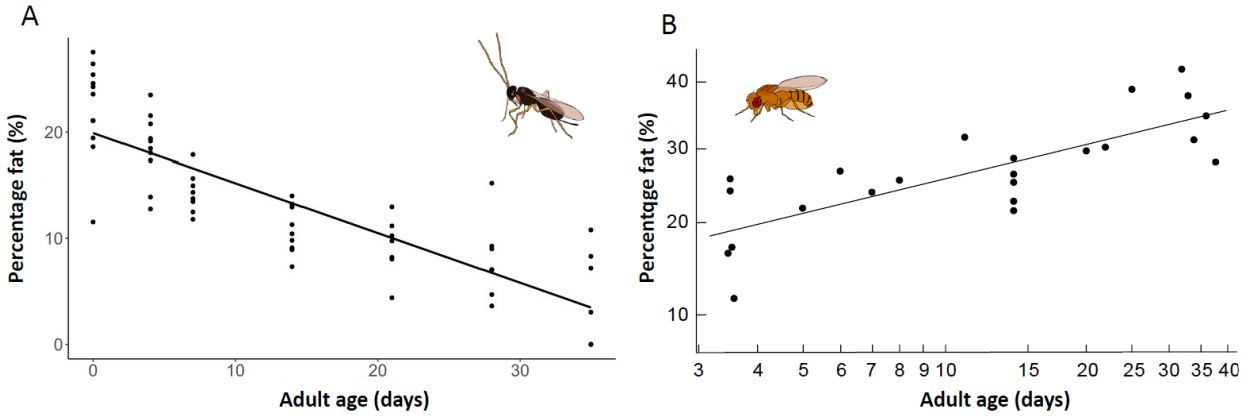

Fig. B1. The proportion of fat (in %) in adult *A. tabida* (A), and *D. melanogaster* (B) throughout life. Based on data from Ellers, 1996 and redrawn from Service, 1987.

A major part of the abdomen and fat body can be dedicated to storing fat. Triacylglycerol levels are indeed generally correlated with size in arthropods (Lease and Wolf, 2011), also in *A. tabida* (Ellers, 1996; Ellers et al., 1998). Larger, fatter females also have more eggs in the ovarioles (Ellers et al., 1998), and *A. tabida* females can emerge with ~160 yolk-poor (i.e., hydropic) eggs (Carton et al., 1986; Le Lann et al., 2014). Many more eggs can be produced during life (i.e., synovigeny; Fig. B2; Jervis et al., 2001), however, with realized fecundity ranging between 580 and 630 eggs when hosts are available in excess (Ellers and van Alphen, 1997). Similar to survival, allocation of fat reserves towards reproduction is highest during the first week of life. If the energetic cost of survival is similar between ovipositing and non-ovipositing females, then ~25ng fat is allocated into each egg during the first week of life. Once fat has been used for reproduction, these reserves cannot be used anymore for other functions (in contrast to some other parasitoid species that resorb eggs; Jervis et al., 2001). Limiting fat reserves can, therefore, lead to so-called trade-offs in life history traits, because energy can be invested either into reproduction or survival (Ellers, 1996) or early versus late reproduction (Seyahooei et al., 2020).

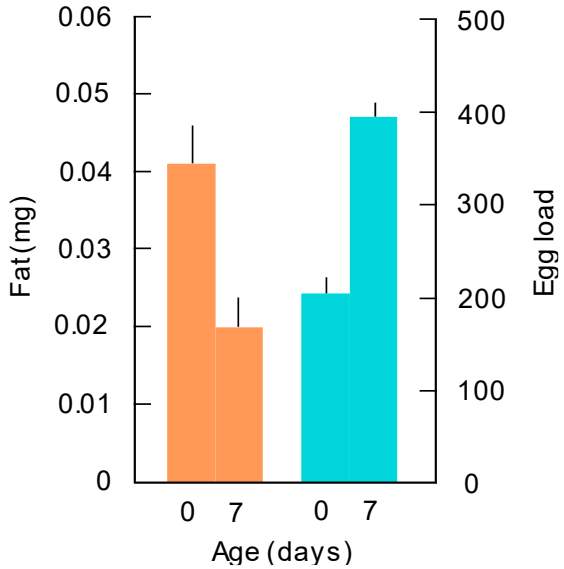

Fig. B2. Amount of fat (mean +1se) and egg load (mean +1se) of *A. tabida* females originating from a population in Kos, Greece, at emergence and 7 days after emergence (with access to food). Redrawn from Ellers & van Alphen, 1997.

Field experiments using a release-recapture approach revealed that dispersal of laboratory-reared *A. tabida* females is size-dependent (Ellers et al., 1998). Larger, fatter females can disperse over larger distances (>15 meters) compared to smaller females. Wild-caught *A. tabida* were generally smaller than laboratory-reared females, and size decreased as the season progressed (from July to September). Larger wild-caught females burned more fat than smaller females, and larger females carried more eggs at the time of capture. Another study examined the size of field-

caught females over several months (June to October) (Ellers et al., 2001). Female size varied, but larger, fatter females were generally captured at the start and the end of the field season. This pattern can be explained by the differences in temperature throughout the field season, with higher temperatures being reached during summer leading to an increase in metabolic activity and lipid oxidation. Alternatively, fat females have a selective advantage when entering and emerging from diapause early and late in the season, as large fat stores are required to survive months at low environmental temperatures (Ellers and van Alphen, 2002).

2.1 Early studies on parasitoid larval fatty acid and triacylglycerol synthesis

Parasitoid fatty acid metabolism started to receive some attention in the late 1960s. Several researchers took an interest in hymenopteran parasitoids, because unlike other insects (e.g., dipterans and lepidoptans; Barlow, 1964, 1965, 1966), some wasps did not seem to have their own characteristic fatty acid composition. Rather than a species-specific qualitative and quantitative fatty acid composition, several wasp species seemed to duplicate the fatty acid composition characteristic of their host. Bracken & Barlow (1967) were the first to investigate this intriguing phenomenon in the ichneumonid parasitoid *Exeristes comstockii*. Using unparasitized larvae of the dipteran host *Lucilia sericata*, the lepidopteran host *Galleria mellonella*, and the sawfly *Neodiprion sertifer* (basal Hymenoptera), hosts that show substantial interspecific quantitative differences in fatty acids, they showed that *E. comstockii* larvae readily duplicated the distinctive fatty acid composition of each of its hosts. The host-specific fatty acid composition of *E. comstockii* remained unchanged throughout pupation and into adulthood, meaning that the duplication phenomenon is not stage-specific. Similar findings were obtained for another ichneumonid parasitoid, *Itoplectis conquisitor*, when reared on the lepidopteran hosts *G. mellonella* and *Ostrinia nubilalis* (Thompson and Barlow, 1970), and in two parasitoid tachinid flies (Delobel and Pageaux, 1981).

To determine the generality of duplicated fatty acid compositions in parasitoids, Thompson & Barlow (1972) tested several other parasitoid species at the larval stage, including aphidids and braconids, as well as species from the superfamily Chalcidoidea, including pteromalids and eulophids. All icheumonids tested (n = 7) had similar fatty acid compositions as their hosts, but the same was true for the pteromalid *Spalangia cameroni* and the eulophid *Dahlbominus fuscipennis.* A subsequent study with 30 species revealed that while most ichneumonids had duplicated fatty acid compositions, this was not a general pattern for this group and duplication occurred also in species from other families (Thompson and Barlow, 1974). It was suggested that duplication of the fatty acid composition may be related to host range of the parasitoid, because most (although not all) species with duplicated compositions were generalists able to develop on a wide range of hosts. It should be noted, however, that in most experiments by Thompson and collaborators unparasitized hosts were compared to parasitoids. When fatty acid compositions differ between hosts and parasitoids, these changes could also result from manipulation of the host's fat metabolism by the parasitoid (see part 3 of this chapter). Furthermore, replication is rather low in the early works of Thompson and colleagues; hence some caution is needed when interpreting their results.

The question arises why some parasitoids duplicate the fatty acid composition of their hosts and how they are doing that? One explanation is that the parasitoids readily take over the fat, and thereby the fatty acid composition, of the host without contributing *de novo* synthesized fatty acids themselves. Thompson & Barlow (1972b) tested this hypothesis using *E. comtockii* reared on the lepidopteran host *G. mellonella* and the dipteran host *L. sericata*. By injecting radiolabeled acetate (^{14}C-1-acetate) into the hosts, and rearing the parasitoid on both host species, it became apparent that *E. comstockii* larvae synthesized (as well as desaturated and elongated) fatty acids, with palmitate (C16:0) being the main synthetic product. While *de novo* fatty acid synthesis is clearly taking place, *E. comstockii* fatty acids also originate from direct incorporation of host fat. This was demonstrated by the presence of eicosenoic acid (C20:1) that was synthesized *de novo* by *G. mellonella* only and was also present in the fatty acid fraction of *E. comstockii* (but without radioactivity and thus synthesis). Thompson & Barlow (1972b) suggested that although parasitoids can synthesize fatty acids *de novo*, they may lack a mechanism to control fatty acid levels. Indeed, typically an increase in dietary fatty acids reduces *de novo* fatty acid synthesis; hence a lipid-rich dietary source, such as an insect host larva, should reduce rather than enhance fatty acid synthesis. Lacking the ability to regulate fatty acid levels could thus contribute to the duplication of fatty acid compositions observed in some parasitoid species.

If host fatty acid composition largely determines that of the parasitoid, what happens if the host is taken out of the equation altogether? Thompson & Barlow (1976) did the test: they reared larvae of another ichneumonid, *Exeristes roborator*, that also duplicates the fatty acid composition of its host, on a fatty-acid free artificial medium. Without any host, the parasitoid larvae readily synthesized fatty acids *de novo* with a composition that did not mimic that of any of its hosts. Moreover, adding host-extracted fatty acids to the artificial medium in relative abundances similar to the host's species-specific composition did not lead to duplication of the fatty acid composition. It is thus not the fatty acids, or their composition, *per se* that influence the parasitoid's fatty acid composition, yet the host itself has a major impact.

The developmental environment is strikingly different for parasitoids reared on an artificially defined fatty acid free-medium compared to a natural, developing host insect that is rich in lipids. Triacylglycerols indeed appeared to be less toxic for *E. roborator* to consume than fatty acids (Thompson, 1977), which makes sense considering that developing hosts typically contain substantial triacylglycerol quantities compared to free fatty acids. If host acylglycerols can be used by the parasitoid with relative ease, perhaps there is no need to invest in energetically costly triacylglycerol synthesis by the parasitoid itself. The study of Jones et al., (1982) compared triacylglycerol synthesis between ichneumonid species that duplicated the host's fatty acid composition, *E. roborator* and *I. conquisitor*, with species that have their own characteristic fatty acid composition irrespective of that of the host, i.e., the ichneumonids *Aphaereta pallipes*, *Hyposoter exigua*, and the chalcid *Brachymeria lasus.* What they found is that *E. roborator* and *I. conquisitor* did not incorporate glycerophosphate into acylglycerols, meaning that the *de novo* triacylglycerol pathway (also known as the Kennedy pathway) was not active. The species that did not duplicate the host's fatty acid composition readily incorporated glycerophosphate. Interestingly, all parasitoids were able to use the monoacylglycerol pathway, where

monoacylglycerols are sequentially acylated by acyl-CoA to form di- and subsequently triacylglycerols. For *E. roborator* 75% of triacylglycerols were formed from diacylglycerols, while this was 97% for *I. conquisitor*. The enzymes of the monoacylglycerol pathway further appeared to be substrate-specific in *E. roborator*, meaning that some fatty acid thioesters are more readily used to form triacylglycerols.

Overall, the work of Thompson and colleagues has shed an exceptional light on the mechanistic basis of the duplication of fatty acid compositions in some parasitoids. When high levels of triacylglycerols are available in a fat-rich host, partial catabolism of triacylglycerols into diacylglycerols (the main form in which lipid are transported through the hemolymph; Soulages & Wells, 1994; Turunen, 1979) can then facilitate a fast and relatively cheap means to synthesize new triacylglycerols for storage in the parasitoid larvae. The parasitoid larvae thus do not use the *de novo* trigylceride pathway, but rather the monoacylglycerol pathway to accomplish this. The similarity in composition is thought to result from acyltransferase specificity favoring fatty acids that are similar to that of the host.

2.2 The lack of adult triacylglycerol accumulation

The storage of fat reserves in periods of food abundance is one of the most conserved metabolic responses across all domains of life (Birsoy et al., 2013). Fat is a key energy substrate fueling insect life, including behavior and fitness (i.e., survival, reproduction; Box 1) (Arrese and Soulages, 2010). Although parasitoids use dietary carbohydrates to meet short-term energetic demands, parasitoids show an extraordinary physiological response to sugar feeding, unlike other insects. During the 90s and 00s, adults of several parasitoid species were found unable to convert excess carbohydrates into long-term storage in the form of fat. For example, the fat content of *A. tabida* (Ichneumonoidea) was highest at emergence and declined rapidly with age, despite continuous sugar feeding (see Box 1; Ellers, 1996). Similar findings were obtained for species in different superfamilies: Ichneumonoidea (*Ventura canescens* and *Diadegma insulare*; Casas et al., 2003; Lee et al., 2004); Cynipoidea (*Leptopilina heterotoma*; Eijs et al., 1998), Chalcidoidea (*Nasonia vitripennis*; *Eupelmus vuilletti*; (Giron and Casas, 2003b; Rivero and West, 2002), demonstrating that this extraordinary physiological phenotype was more common in parasitic Hymenoptera.

This lack of fat accumulation was proposed to be an evolutionary consequence of the parasitoid lifestyle (Visser and Ellers, 2008). Efficiently utilizing a single host insect and manipulating its nutrient content (see part 3 on host manipulation) generally leads to a lipid-rich environment for developing parasitoid larvae. Under such conditions, where host fat content is high, fat accumulation is no longer necessary (leading to relaxed selection; Lahti et al., 2009) or too costly to maintain (i.e., leading to selection against fat accumulation). Visser & Ellers (2008), therefore, hypothesized that parasitoids lost the ability for fat accumulation. A comparative approach with more than 90 insect species indeed revealed that *1)* loss of fat accumulation is ancestral in parasitic Hymenoptera; *2)* the loss of fat accumulation coincided with or followed the evolution of the parasitoid lifestyle; and *3)* there is parallel evolution, as the loss of fat accumulation evolved repeatedly and independently in parasitoid flies, beetles and wasps (Visser et al., 2010).

There were some exceptions, however, because several species did accumulate fat as adults. Analysis of several parasitoid-related traits revealed that fat accumulation re-evolved in generalist parasitoid species (i.e., parasitizing > 10 host species), including *L. heterotoma*, *Pteromalus puparum,* and *Gelis agilis* (Visser et al., 2010). An explanation is that manipulation of host fat content is difficult when many species can serve as potential hosts, making fat accumulation in adults critical for survival and reproduction when fat-poor hosts are used (Visser et al., 2010).

The question is which mechanism underlies the loss of fat accumulation in adult parasitoids? There can be several ways in which the fat accumulation phenotype was lost: *1)* the gene(s) underlying fatty acid or triacylglycerol synthesis and accumulation were lost (i.e., not present in the genome anymore; as in the yeast *Malassezia globosa*; Xu et al., 2007); *2)* the gene(s) have accumulated mutations in the coding regions, leading to non-functionality; *3)* the gene(s) remain present, but are silenced through regulatory processes. Consequently, either insufficient quantities of fatty acids and triacylglycerols are produced or accumulation itself is hampered. The loss or non-functionality of key genes in the fatty acid and triacylglycerol metabolism pathways is unlikely, however, because many genes (e.g., fatty acid synthase *fas*, Acetyl-CoA carboxylase *acc*, glycerol-3-phosphate-acyltransferase *gpat*) involved in the conversion of carbohydrates into triacylglycerols, are also essential for the synthesis of other lipid classes and are part of other key metabolic pathways (e.g., pyruvate metabolism, citrate cycle; phospholipids).

The first study on the molecular mechanisms and transcriptional profiles underlying the lack of fat accumulation in parasitoids was performed with the chalcid *N. vitripennis*. Genome analysis indeed revealed that coding sequences of *fas* and *acc* did not contain any stop codons, mutations, or signs of genetic damage (Visser et al., 2012). But contrary to findings in other animals, no effect of sugar feeding was found on the transcription levels of *fas* or *acc* (Visser et al., 2012), suggesting that fatty acid synthesis is not taking place. The same was found for *gpat*, involved in the early steps of acylglycerol synthesis (and part of the *de novo* triacylglycerol synthesis pathways). Genes involved in the monoacylglycerol pathway, e.g., *dgat*, did not respond to sugar-feeding neither. Functionality of *fas*, *acc*, as well as other genes and their enzymes were also confirmed in several other parasitoid wasp species (Kraaijeveld et al., 2019; Lammers et al., 2019; Prager et al., 2019; Visser et al., 2021). These results suggest that changes in gene expression, rather than structural genetic changes are involved in the lack of fat accumulation (Visser et al., 2012).

For transcriptomic studies, it is essential to know how the phenotype is affected. In the case of Visser et al., (2012), absence of fatty acid synthesis and fat accumulation were determined using stable isotope tracking methods (of deuterium into fatty acids of the neutral lipid fraction) and bulk fat extractions (comparing fat quantities between emerged and fed wasps), respectively. No incorporation of stable isotopes was found in fatty acids of the neutral lipid fraction, indicating that fatty acid synthesis did not take place, as suggested by the lack of gene transcription. In contrast the honeybee *Apis mellifera,* that readily synthesizes and stores fat, did incorporate isotopes into the neutral fat fraction. In *N. vitripennis,* no fat accumulation took place, because fat quantities decreased significantly during life. Even though fat does not accumulate, intermediary metabolites involved in fat metabolism can still be synthesized. For example, Ruther et al., (2021) found that

several parasitoid wasp species could synthesize fatty acids, and in the case of *N. vitripennis*, utilize these fatty acids in triacylglycerols and eggs (Multerer et al., 2022). However, no increase in bulk triacylglycerol stores was observed (Ruther et al., 2021). This means that even though fatty acids are synthesized and used to form triacylglycerols, this species still lacks adult triacylglycerol accumulation.

To further understand the (lack of) fat accumulation phenotype observed in parasitoids, Visser et al., (2017) compared fatty acid synthesis between *D. melanogaster*, showing typical and significant fat accumulation after feeding, a parasitoid that lacked fat accumulation, *E. vuilletti*, and two parasitoids that readily accumulate fat as adults, *G. agilis* and *Gelis aerator*. Fatty acid synthesis (of C16:0) was indeed very high for species that accumulate fat as adults, while for *E. vuilletti* that does not accumulate fat, no fatty acids were synthesized *de novo.* The same patterns were found when fatty acid synthesis was analyzed in the larvae of these species. There thus seems to be concurrence in fatty acid synthesis phenotypes between larvae and adults.

Work on fat metabolism in parasitoids has had two distinct spurts in terms of studies, one between the 60s and 80s focused on the similarity of fatty acid compositions between hosts and parasitoid larvae; the other still ongoing to understand why adult parasitoids do not accumulate fat. It may well be that despite the slightly different interests and focus, both phenomena result from the same underlying mechanism(s) and evolved in a similar way. Only one study has so far compared fatty acid compositions and fat accumulation strategies, using a rose gall wasp community, including the parasitoids *Orthopelma mediator* and *Pteromalus bedeguaris*(Visser et al., 2013). Both parasitoids did not accumulate fat as adults, and only the fatty acid composition of *O. mediator* was considerably different from its main host. *O. mediator* is an ichneumonid and *P. bedeguaris* a chalcid, both with a very limited host range. Fat accumulation strategy does thus not seem to be related to mimicking of the host's fatty acid composition, as *O. mediator* seems to have its own characteristic fatty acid composition compared to its two potential hosts. Host breath, with generalists being more likely to copy the fatty acid composition of the host, also does not seem to be related, because *P. bedeguaris* has a similar fatty acid composition as its hosts. This specific system may, however, not be ideal for evaluating the link between fat accumulation, host breadth and fatty acid compositions, because this community occupies a very particular ecological niche. More work is thus needed to determine whether the lack of fat accumulation coincides with mimicking of the fatty acid composition of the host.

2.3 More complex fat synthesis and accumulation phenotypes

While the majority of parasitoids do not accumulate fat as adults, for some parasitoid species repeated experiments hinted at more complicated patterns. For example, Moiroux et al., (2010) proposed that the ability of parasitoid wasps to accumulate fat was closely tied to geographic location and local environmental conditions. To test this, four geographically distinct *Leptopilina boulardi* populations were collected. Different fat accumulation phenotypes were found: two populations accumulated fat, while the two other populations did not (Moiroux et al., 2010). These observations could be related to genetic divergence between populations, as the two populations

that accumulated fat were genetically closer to each other than to populations that did not (Seyahooei et al., 2011; Visser et al., 2017).

Like Moiroux et al., (2010), a large-scale study on the ability of fat accumulation of field-caught *L. heterotoma* populations and other *Leptopilina* species also revealed contrasting fat accumulation phenotypes (Visser et al., 2018). These differences were found to be related to the fat content of the *Drosophila* host used. Indeed, parasitoids emerging from a lean host contained a lower quantity of fat, leading to fat accumulation in adults, while no fat accumulation was observed for parasitoids emerging with a high amount of fat (Visser et al., 2018). A more recent study with several *L. heterotoma* populations confirmed that this species can switch fat synthesis and accumulation on or completely off depending on the host's fat content. Wasps generally start synthesizing and accumulating fat when their developmental environment (i.e., a *Drosophila* larva) is fat-poor (Visser et al., 2021). Variation in fat accumulation strategies in *L. heterotoma* is plastic, meaning that a single genotype can generate different fat synthesis and accumulation phenotypes depending on environmental conditions (Fig. 2). Our future work will center around understanding how ecological conditions affect plastic fatty acid synthesis and fat accumulation.

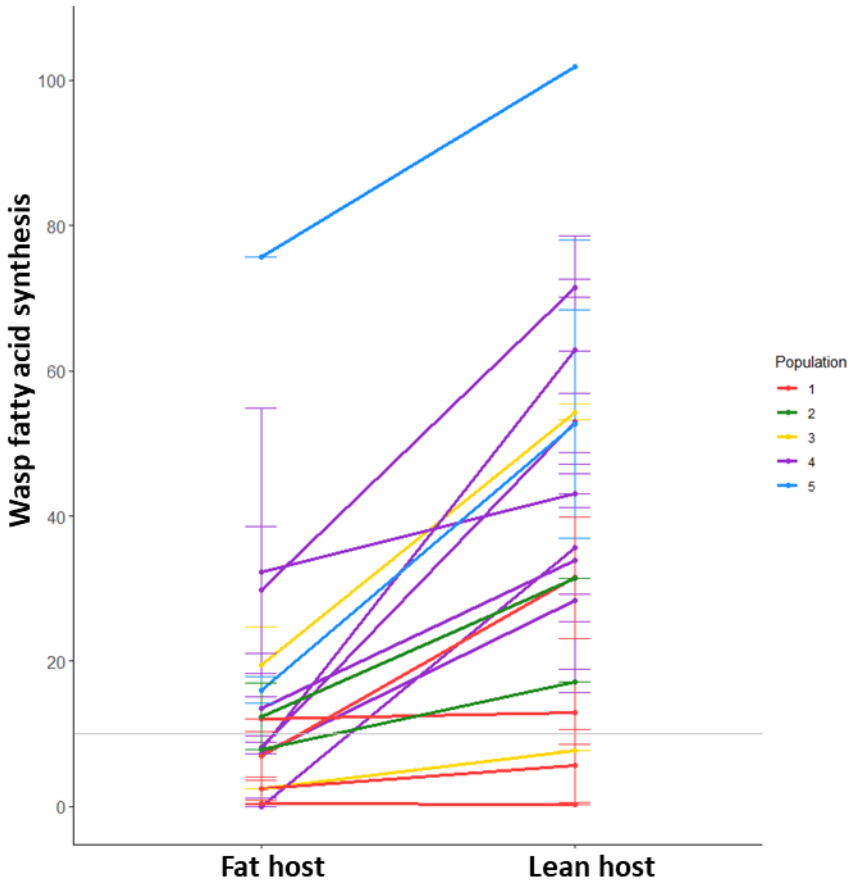

Fig 2. Fatty acid synthesis of *L. heterotoma* families (sharing 75% of their genome) originating from different populations (1 to 5). Reaction norms reveal that some families show plasticity in fatty acid synthesis (synthesis on lean hosts, no synthesis on fat hosts), while other families consitutively synthesize fatty acids or lack fatty acid synthesis (under the gray line). Variation in the slope of reaction norms suggests

that there is genetic variation for plasticity of fatty acid synthesis, meaning that plasticity itself can evolve in response to selection. Redrawn from Visser et al., 2021.

3 Lipid metabolism in parasitized hosts

Part 2 of this chapter detailed the progress made with research on fatty acid synthesis and fat accumulation of the parasitoid itself, but the parasitoid's unique lifestyle has also led to the evolution of intricate mechanisms to manipulate host metabolism. A parasitoid relies on only a single host to complete its development and to obtain sufficient nutrients to fuel adult life. There is thus an incredible advantage for the parasitoid to "hijack" the host's metabolism for its own benefit. Considering that many parasitoids do not accumulate substantial fat quantities as adults (part 2) and fat is of key importance for life histories and fitness (Box 1), manipulating host lipid metabolism so that host lipids become more accessible or available for the parasitoid has a clear adaptive value.

Before exploring how host lipid metabolism can be manipulated, there are some parasitoid-specific traits that have a large impact on host manipulation. Parasitoids show tremendous diversity related to their mode of life and general biology (Fig. 1; Godfray, 1994; Quicke, 1997). An important distinction can be made, for example, between parasitoid species that arrest the host's development, idiobionts, and species that allow the host to continue feeding and growing, koinobionts. For idiobionts, resource availability for the parasitoid is largely fixed close to the time of oviposition, while for koinobionts resources generally keep on accumulating, at least for some time while the parasitoid is developing. Among koinobionts, a further distinction can be made between species that stop host development prematurely, reducing final host body size, and species that prolong host development, increasing final host body size (Cuny and Poelman, 2022). Several studies have indeed reported that host food consumption is reduced following parasitism by koinobiont parasitoids (Kaeslin et al., 2005; Morales et al., 2007; Pruijssers et al., 2009; Shi et al., 2015; Thompson, 1982a, 1983), while some parasitized hosts feed longer (Thompson and Redak, 2008). The extent to which a parasitoid can affect host development can also be dependent on the environment (i.e., phenotypic plasticity). The braconid *Meteorus pulchricornis*, for example, increases final size of the small lepidopteran host *Plutella xylostella* by 30%, while final size of the larger lepidopteran host *Mythimna separata* is increased by 95% (Harvey et al., 2010). The host species on which a koinobiont parasitoid develops can thus have a major impact on resource levels and resource availability, which in turn can have major consequences for fat synthesis and accumulation of the parasitoid itself (part 2.2; Visser et al., 2021).

Parasitoids are highly efficient in carrying over resources from their host, which for some species can mount to >90% of the host's body mass (Harvey et al., 2009). An increase in fat availability and content for the developing parasitoid can have a major impact on both larval survival and adult fitness (Rivers et al., 1998). When more fat can be carried over from the host, the parasitoid has more energetic reserves available for allocation into fitness-related traits (Box 1). On the other hand, too much host tissue can be detrimental when complete consumption of host tissues is required (Harvey, 1996; Harvey and Strand, 2002). Indeed, many parasitoids are so-called

"tissue-feeders", where most or all host tissues are consumed during the parasitoid's development (Fig. 1). Within the superfamily Ichneumonoidea, all gregarious (i.e., with multiple offspring emerging from a single host) koinobiont endoparasitoids (e.g., *Microplitis* sp. and *Cotesia* sp., as well as the family Chelonidae) have, however, evolved the ability to feed mostly on hemolymph (Harvey and Malcicka, 2016). These "hemolymph-feeders" inially only feed on hemolymph and part of the fat body of the host but exit the host during the last larval stage to pupate externally (Fig. 1). The adaptive significance of hemolymph feeding is that a wider range of host developmental stages and sizes can be parasitized, including hosts that are much larger than the parasitoid itself.

The question is whether hemolymph or tissue-feeding koinobionts have evolved different strategies to manipulate host metabolism. We could expect that for tissue-feeders, increasing fat body lipid content is more important, while for hemolymph-feeders an increase or steady flow of hemolymph lipids can increase the efficiency of scavenging from the host. This could be tested using host-parasitoid systems where the same host species is attacked by multiple parasitoids that differ in feeding strategy. Koinobiont microgastrine braconids and campoplegine ichneumonids may be ideal systems for testing this, e.g., *P. xylostella* parasitized by *Diadegma semiclausum* (tissue-feeder) and *Cotesia vestalis* (hemolymph-feeder), *Pieris brassicae* parasitized by *Hyposoter ebeninus* (tissue-feeder) and *Cotesia glomerata* (hemolymph-feeder), *Spodoptera littoralis* parasitized by *Hyposoter didymator* (tissue-feeder) and *Cotesia marginiventris* (hemolymph-feeder). Comparing host manipulation strategies of hemolymph versus tissue-feeders developing on the same host offers a unique opportunity to increase our understanding of the mechanisms underlying host manipulation.

3.1 The effects of parasitism, including the developing parasitoid itself

The easiest, and sometimes only, way to determine how host metabolism is affected by parasitism is to compare unparasized with parasitized hosts. Overall, host lipid levels can increase, remain stable, or decrease for a variety of reasons (Table 1). For example, parasitism of the locust *C. terminifera* by the parasitoid fly *Trichopsidea oestracea* led to a steep increase in overall lipid content, although the mechanism has remained unclear (Horwood and Hales, 1991). In another parasitoid fly, *Blepharipa sericariae*, the developing larvae were found to secrete a peptide that inhibits lipid transport in its host silkworm, *Philosamia cynthia prieri* (Hayakawa, 1987). *B. sericariae* eggs are consumed by the host during larval feeding, and after parasitoid hatching the parasitoid larvae remain in the second instar until the following spring when the larvae molt and start feeding on the host's pupal tissues. Lipid release from the fat body into the hemolymph was reduced by 50-70%, and lipid uptake by lipophorin (used for lipid transport) was inhibited by ~60% through the action of a parasitoid-secreted peptide. Similar results were obtained when the locust *Locusta migratoria* was used, where the decrease in lipid-uptake mainly affected the diacylglycerol component of the lipid fraction. This finding supports the idea that lipid uptake and transport in the hemolymph, which typically entails the transport of diacylglycerols in insects (Turunen, 1979; but see Ford & van Heusden (1994) who identified a lipophorin transporting triacylglycerols in *Aedes*

aegypti), is inhibited. Considering it takes about a year for *B. sericariae* to complete its development, inhibition of lipid transport by lipophorin conserves the triacylglycerol stores of the host's fat body. This is needed for the developing parasitoid to be able to complete its development in spring. *T. oestracea* takes a similar time to develop as *B. sericariae*; hence both parasitoid flies have optimized host use, either by increasing or conserving the lipid stores of their respective hosts.

So far, most work on host manipulation has been done on laboratory-reared hymenopteran parasitoids. Unlike the dipteran parasitoids described above, many hymenopteran parasitoids complete their development within several weeks. Major physiological changes can already be brought about within a short timespan, including a decrease in lipid levels. There are several, not mutually exclusive, reasons why lipid levels are lower in parasitized compared to unparasitized hosts: *1)* the host is not able to develop its own fat body (Dahlman, 1970); *2)* the host's physiology is redirected; 3) host and parasitoid compete for lipid resources, with both species consuming and utilizing lipids.

The koinobiont parasitoid *H. exiguae* feeds mainly on lepidopteran host hemolymph (*Trichoplusia ni*) during the first 8 days of development (when the host moults into its third and fourth instar), after which the larvae exit the host to pupate externally (Thompson, 1982b). Parasitized larvae had a lower concentration and total lipid content compared to unparasitized larvae, mainly near the end of parasitoid development. The reason is that parasitized, unlike unparasitized larvae, do not enter metamorphosis. Metamorphosis is an energetically costly process and disruption of the host *T. ni's* physiology inhibits the prepatory mechanisms to initate metamorphosis, including lipid accumulation, explaining the lower lipid levels during later developmental stages of parasitized hosts. Lower lipid levels in parasitized hosts may superficially seem a disadvantage for the parasitoid, but for *H. exiguae* redirection of the host's metabolism suffices for the parasitoid to successfully complete development.

Competition for host resources is also apparent in the *H. exiguae*-*T. ni* system. Lipid levels were found to be similar between parasitized and unparasitized starved hosts (Thompson, 1982b). In terms of the host's physiological state, parasitism thus seems to mimic starvation (at least to some extent, because starvation was found to be metabolically more demanding; Thompson, 1982a). In contrast to a starved host, however, a parasitized host still has access to food (at least in this system, where host development continues), which means that host and parasitoid are in direct competition for lipids (Dahlman and Greene, 1981; Nakamatsu and Tanaka, 2004). Lipids of parasitized *T. ni* were, however, not depleted completely, suggesting that the parasitoid utilizes resources in such a way that the host does not die prematurely (which would also lead to death of the parasitoid).

The above studies contribute to our general understanding of how lipid metabolism of the host is affected following parasitism, including the investigation of rare field-collected hosts that are typically more difficult to study. Experiments focusing solely on the effect of parasitism can, however, be confounded by other factors that can affect host metabolism, such as venom, teratocytes, and mutualistic viruses, which will be discussed in more detail in the following sections.

3.1 Venom-induced changes in host lipid metabolism

All female Hymenoptera produce venom in a specialized venom-gland that is a part of the reproductive system (Pennacchio and Strand, 2006; Poirié et al., 2014). The venom of parasitoids is injected in the host together with the egg(s) and consists of both proteinaceous and non-proteinacious compounds (Moreau and Asgari, 2015). The venom of ecto and endoparasitoids seems to serve different functions, for the former mainly inducing host paralysis and for the latter mainly interfering with the host's immune system. For all parasitoids, nutrient acquisition during development is critical for survival, investment in costly metamorphosis, and to fuel (at least part of) adult life. In this subsection, we will focus solely on the effects of venom on host lipid metabolism.

3.1.1 Lipid-related venom components

Venom components related to lipid metabolism have been identified in 23 different parasitoid species. The function of venom enzymes can be divided into four different categories: lipid catabolism, transport, synthesis, and storage (Table 2). When venom is injected, enzymes can immediately start freeing lipids for the developing parasitoid. Cathepsin for example, contributes to disruption of the host's fat body (Becchimanzi et al., 2020). On a cellular level, phospholipases play a key role for increasing nutrient transfer from the cytosol to the hemolymph and for disintegrating cells to release their content. Various phospholipases have been identified in parasitoid venom that differ in their specific site of action in the substrate. Phospholipase A1, for example, disrupts phospholipid packaging of cell membranes, leading to cell lysis, while phospholipase A2 hydrolyzes glycerophospholipids in membranes to release free fatty acids and lysophospholipids (Perez-Riverol et al., 2019). Phospholipases can indeed be part of a complex pathway affecting the host's lipid metabolism. The venom of *N. vitripennis*, for example, modifies cell membrane permeability leading to an influx of Na^+ in the cell (Danneels et al., 2010; Rivers et al., 2002). An increase in Na^+ can subsequently activate phospholipase C, leading to an increase in inositol-3-phosphate (a signaling molecule) and the release of Ca^{2+} from the mitochondrion. Calcium in turn activates phospholipase A2, stimulating fatty acid synthesis. Within parasitoid venom, phospholipases thus play an important role in making lipids available for parasitoid offspring.

The venom of 13 parasitoids was also found to contain several different lipases, mainly involved in the catabolism of different lipids, including mono, di-, and trigylcerides (Wang et al., 2020b). In *P. puparum*, investigation of lipases in the venom revealed that different lipase families are present, but also that many lipases are non-catalytic (Wang et al., 2020b). An explanation is that the non-catalytic lipases have acquired a new function, mainly for binding and transporting lipid.

Once lipids are released from the fat body, they need to be transported to the developing parasitoid through the hemolymph. Several hemolymph transport enzymes are contained within the venom, including apolipophorin and apolipoprotein (Table 2). In addition to these more typical enzymes involved in lipid transport, there have now been several reports of odorant binding proteins being part of the parasitoid venom (e.g., in *N. vitripennis, P. puparum*, and 8 other species;

Table 2). Odorant binding proteins typically serve for the transport of odorant molecules (e.g., pheromones) to olfactory receptors. In the parasitoid venom, odorant binding proteins are hypothesized to play a role as fatty acid and fatty acid ester carriers, as was found in several other insects (e.g., the ant *Camponotus japonicus*; Ishida et al., 2013; the blowfly *Phormia regina*; González et al., 2009). Like lipases, existing odorant binding proteins thus seem to have acquired new functions.

The venom of some species also contains enzymes that are involved in lipid synthesis, including fatty acids, glycosphingolipids, and diacylglycerols. So far, no clear explanation has been proposed as to why the venom would contain enzymes involved in lipogenesis. Focusing on fatty acid synthase for the synthess of fatty acids (mainly palmitate, C16:0), and phosphatidate phosphatase for the synthesis of diacylglycerols, some of the hosts exploited could be relatively lipid-poor. The aphid *Acyrtosiphon pisum* and the scale insect *Parasaissetia nigra,* for example, are plant sap-sucking insects, a nutritional resource that is expected to contain a lot of carbohydrates, but not many lipids. Lipid synthesis enzymes present in the venom can then utilize precursors, such as carbohydrates, from the host to increase lipid content and availability. The presence of fatty acid and diacylglycerol synthesizing enzymes in the venom may aid the developing parasitoid in obtaining sufficient lipids.

Three enzymes implicated in adipocyte maturation and/or lipid storage were found in several parasitoid species. An explanation for an increase of lipid storage in adipocytes could be that parasitism and envenomation alter metabolism in such a way that more precursors for acylglycerols become available. Indeed, the synthesis of fatty acids and diacylglycerols described above is an example. At the time of oviposition, the parasitoid is still in the egg or early larval stage, a time at which absorption of nutrients may be relatively little (compared to later developmental stages). For example, during the early stages of parasitism, the braconids *Aphidius ervi* and *Toxoneuron nigriceps* absorb nutrients through the epidermis (Caccia et al., 2005; Grimaldi et al., 2006). In parasitoid offspring in general, some time may be needed to develop a fully functioning gut and absorption through the epidermis or the anal vesicle in early larval stages may be more common (Edson and Vinson, 1977). Storage of large fat reserves is also expected to take some time, with fat droplets becoming clearly visible only during later larval instars (e.g., in *E. vuilletti* and *Gelis* sp., Pers. Obs.). When fat precursors accumulate in the hemolymph of the host, initial fat storage in adipocytes can provide a reserve to be consumed by the developing parasitoid at a later time. Increased fat storage was indeed found for the parasitoid *N. vitripennis* when parasitizing its preferred fly host *Sarcophaga bullata* (Rivers and Denlinger, 1995, 1994).

3.1.2 Venom-induced alterations in host lipid metabolism

Venom generally leads to an increase in lipid levels either in the whole body, the fat body or the hemolymph (Table 1). There are some exceptions, however, where lipid levels were lower, or no changes were observed. For example, in parasitized *S. littoralis*, Transmission Electron Microscopy revealed that the fat body rapidly released its content (glycogen and lipids) through cell vacuolization and reabsorption (Becchimanzi et al., 2017). This process was aided by haemocytes surrounding the fat body and increased cathepsin L activity. Hemolymph titers of

glycerolipids decreased during 48 hours, probably because the host's tissues require fat for ongoing, albeit reduced, metabolic activities. For the lepidopteran *Tenebrio molitor* parasitized by the bethylid *Scleroderma sichuanensis*, fat body and hemolymph lipid content also decreased following envenomation and parasitism (Zhuo et al., 2016). This decrease could be due both to consumption of the parasitoid and the host's requirement for lipid. Unlike parasitism, where the fat body was degraded, envenomation alone did not alter the appearance of the fat body, suggesting that factors other than venom are needed to rupture the fat body.

Host manipulation requires fine-tuned physiological interactions between parasitoid and host, which can be highly species-specific. For example, the parasitoid *N vitripennis* is highly polyphagous, being able to parasitize more than 60 different host species (Desjardins et al., 2010). Yet, despite its wide host range, *N. vitripennis* prefers to oviposit on the fly *S. bullata* (Desjardins et al., 2010). Rivers and Denlinger (1995) looked at the effect of parasitism by *N. vitripennis* on four distinct fly species, including *S. bullata, P. regina, Musca domestica,* and *Sarcodexia sternodontus.* Only in *S. bullata* marked increases in fat body and hemolymph lipids were observed (Rivers and Denlinger, 1995). For *P. regina* and *M. domestica* hemolymph lipids also increased following parasitism, but for the fly *S. sternodontus* fat body and hemolymph lipids decreased. For *S. bullata* both envenomation and parasitism actually led to increased fat content, which could result from active fatty acid and fat synthesis by the host. Parasitism by the wasp *Lysiphlebus japonica* of the aphid *Aphis glossypii* led to upregulation of almost all genes in the glycerolipid pathway, including diacylglyerol acyltransferase that produces triacylglycerols from diacylglycerols, revealing that venom can indeed induce lipogenesis in hosts (Zhang et al., 2015).

The braconid parasitoid *Cotesia kariyai* (that parasitizes the lepidopteran *Pseudaletia separata*) can itself be parasitized by the pteromalid *Trichomalopsis apanteloctena* once *C. kariyai* leaves its own host to pupate externally (Nakamatsu and Tanaka, 2004). Lipid levels in developing pupae of *C. kariyai* gradually decrease, and lipid content of *C. kariyai* determines the size of *T. apanteloctena*. An increase in host fat content may indeed not be expected here, because the primary parasitoid host, *C. karyai,* may itself show little to no fat accumulation (as is the case for other *Cotesia* species; Visser et al., 2010).

Zooming in on the interaction between *N. vitripennis* an *S. bullata,* the elevation in host hemolymph lipids depended on the location where oviposition occurred on the host pupa. A posterior sting, where parasitism occurs under natural conditions, led to a steeper increase in lipids compared to an anterior sting (Rivers and Yoder, 1996). *N. vitripennis* larvae developing on posterior-oviposted hosts also contained more fat than anterior-oviposited hosts, suggesting indeed that more lipids are available. Elevation of host hemolymph lipids was also associated with the number of developing larvae. A higher number of eggs laid led to a higher increase in hemolymph lipid content. A similar finding was obtained for another gregarious parasitoid, *Trichomalopsis* near *americana*. This would suggest that venom increases nutrient content of the host in such a manner that competition can be avoided between multiple offspring developing from the same host and the host itself. Overall, the work on *N. vitripennis* suggests that idiobiont parasitoids may not be as restricted as previously thought when it comes to resource availability, including lipids (Rivers and Yoder, 1996).

Only few researchers investigated both the composition of the venom and their effects on host lipid metabolism. Wang et al., (2020b) looked at lipases and their diversification focusing on venom of the chalcid *P. puparum*, but also investigated how host lipid metabolism was affected following parasitism. Overall, parasitism led to a decrease of triacylglycerols and several phospholipids (e.g., sphingomyelin, phosphatidylcholine etc...) in the fat body, whereas these lipids increased in the hemolymph (Table 1). The increase of triacylglycerols in the hemolymph was concurrent with a decrease in diacylglycerols. In *P. pupurum* venom, diacylglycerol acyltransferase (DGAT2), catalyzing the last step of triacylglycerols synthesis from diacylglycerols, is not present. The venom does, however, contain multiple lipases (some with missing catalytic triads, potentially involved in lipid binding and transport), which suggests that the host's enzymatic machinery facilitates the conversion of di- to triacylglycerols. In the fat body, increasing triacylglycerol levels were mainly observed for highly unsaturated triacylglycerols, while triacylglycerols with fewer double bonds decreased. An increase in unsaturation generally increases triacylglycerol solubility. There was, however, no difference in unsaturation levels of triacylglycerols in the hemolymph; hence it is unclear what role the unsaturation plays in the fat body (i.e., higher solubility does not lead to increased transport and presence of unsaturated triacylglycerols in the hemolymph for use by the parasitoid larva). Desaturases were not found in the venom of *P. puparum,* but a desaturase was found to be upregulated in the venom glands. It thus remains unclear whether the wasp's venom or the host is responsible for the observed changes in triacylglycerol saturation levels.

The decrease of some phospholipids in the fat body and increase in the hemolymph further suggests that destruction of the fat body and fat body cell membranes ensues quickly after parasitism by *P. puparum* (Wang et al., 2020b). Parasitized hosts further had an increased cholesteryl ester content in the hemolymph, suggesting that cholesterol transport increased. Having been located in the Dufour's gland (i.e., which is part of the anatomy of the ovipositor) suggests that the cholesteryl esters may be derived from the venom. Lipases with potential cholesteryl esterase function have been identified from the salivary glands of developing *P. puparum* larvae, which could subsequently act on cholesteryl esters injected by the mother. This may allow the developing parasitoid to acquire essential sterols (that insects cannot synthesize) that may subsequently serve important functions as hormone-precursors, signaling molecules, and components of cell membranes. Sterols were further found to increase egg viability (when obtained through host-feeding in adults) (Mondy et al., 2006).

3.2 Polydnaviruses increase lipid availability for the developing parasitoid

Polydnavirus are viruses associated with endoparasitoid wasps (i.e., the primary hosts), persisting as proviruses (i.e., DNA sequences integrated in the host genome) in every germ and somatic cell of the wasp. Viral replication occurs only in calyx cells (that are part of the wasp's reproductive tract) during the pupal and adult stage. Polydnaviruses can be grouped into two distinct genera: Bracoviruses, associated with the braconid family, and Ichnoviruses, associated with the ichneumonid family (Strand and Burke, 2013). Braco- and Ichnoviruses each have a distinct morphology of the virion (that enters the secondary host, which is the host of the parasitoid) and

an independent evolutionary origin (Strand and Burke, 2012). Polydnaviruses do, however, have a similar life cycle, where virions are injected in the host during oviposition along with the egg and the venom (Strand and Burke, 2013). After virion injection, the virus integrates into the host genome to express virulence genes. The virus thus participates in the parasitization process, affecting the host's immune system, host growth, and metabolism (Strand and Burke, 2013). This subsection focuses on the effects polydnaviruses have on (secondary) host lipid metabolism.

The braconid *Chelanus inanitus* is an endoparasitoid that produces venom and a bracovirus. Kaeslin et al., (2005) disentangled the role of the venom, the bracovirus, and the developing parasitoid. This is possible when comparing parasitized hosts, with unparasitized hosts, but also using pseudoparasitized hosts, where the eggs within the mother are killed prior to oviposition using x-rays. Pseudoparasitized hosts thus receive the venom and the bracovirus, but the parasitoid larva does not hatch. Venom proteins disappear within 1-2 days after parasitization, while the polydnavirus remains throughout parasitoid development. The parasitoid larva, along with polydnavirus, cause an accumulation of whole-body lipids during development (Table 1). During the last host larval instar, most lipids had accumulated in parasitized hosts only, meaning that the parasitoid larva itself also plays a major role in increasing host fat accumulation.

In a recent study, Wang et al., (2021) revealed that the bracovirus associated with *C. vestalis* plays an important role in regulating lipid metabolism of the host moth *P. xylostella*. Following parasitization, whole-body triygylceride levels decreased, as did hemolymph fat levels, although there was a peak in the hemolymph at the onset of the last larval stage. The secondary host insect can acquire fatty acids from its diet that are subsequently absorbed by the gut, resynthesized into lipids, and transported to the fat body with lipoproteins. Parasitized hosts indeed showed reduced formation of neutral lipid droplets, meaning that an alteration of host lipid absorption and synthesis underlies the decrease in whole-body lipids. *C. vestalis* produces venom containing bracoviruses as well as teratocytes. To determine which parasitoid-associated factor led to depressed lipid levels, Wang et al., (2021) also used pseudoparasitized wasps, thereby removing the effect of teratocytes and the developing parasitoid. Injection of venom alone did not result in any changes; hence the bracovirus was found to be responsible for altered lipid levels in the host. Focusing on the mechanism at play, transcriptomics led to the identification of several bracovirus genes that could be involved in manipulating host lipids. Expression of one of these genes, *CvBV 9-2*, was indeed found to be responsible for reducing triacylglycerol levels in parasized larvae by increasing the expression of a tachykinin gene (*PxTk*) in the host gut suppressing lipogenesis.

3.3 Parasitoid-derived teratocytes increase fat availability for the parasitoid

Teratocytes are specialized cells derived from the dissociation of the cellular membrane surrounding the parasitoid embryo during its development that are released in the host's hemolymph during parasitoid hatching (Strand, 2014). Teratocytes are produced by some subfamilies within Braconidae (i.e., Microgastrinae, Meteorinae, Euphorinae, Aphidiinae) and Platygastridae (i.e., Scelioninae, Telenominae, Teleasinae), all of which are endoparasitoids (Dahlman, 1990; Strand, 2014). Teratocyte-like cells have also been reported in the Ichneumonidae

(Rouleux-Bonnin et al., 1999) and Chalcidoidea (Pedata et al., 2003; Strand, 1986). The number of teratocytes is species-specific, and can range from 10 (e.g., *Telenomus heliothidis*, Platygastridae; (Strand et al., 1988) to more than 1000 (e.g., *M. pulchricornis*, Braconidae; Suzuki & Tanaka, 2007) (Strand, 2014). Teratocytes help to disrupt host growth, inhibit host metamorphosis, but also seem to play a role in evading the host's immune system (Ali et al., 2013; Dahlman et al., 2003; Strand, 2014). Teratocytes further aid in nutrient acquisition for the developing parasitoids, particularly lipids (Falabella et al., 2005, 2000; Nakamatsu et al., 2002; Qin et al., 2000; Suzuki and Tanaka, 2007).

Ultrastructure studies revealed that once released in the host's hemolymph, teratocytes show both morphological and metabolic changes (Pennacchio et al., 1994; Strand et al., 1986; Volkoff and Colazza, 1992; Zhang et al., 1994), e.g., teratocyte size greatly increases (de Buron and Beckage, 1997; Strand and Wong, 1991; Volkoff and Colazza, 1992). To promote nutrient exchange between the teratocyte's intracellular and extracellular space, teratocytes exhibit long microvilli on their surface (to increase the surface for absorption/secretion), as well as large exosome-like spherical vesicles (containing lipids and other nutrients; (Hotta et al., 2001; Salvia et al., 2019; Shelby et al., 2014; Sluss, 1968). An abundant rough endoplasmic reticulum, numerous mitochondria and an extensive vacuolization are observed in the cytoplasm (de Buron and Beckage, 1997; Gerling and Orion, 1973; Sluss, 1968; Volkoff and Colazza, 1992). These morphological and metabolic characteristics confirm that teratocytes can absorb nutrients or secrete proteins into the host's hemolymph (Dahlman and Vinson, 1993; Salvia et al., 2019; Sluss, 1968). Teratocytes further do not divide after being released, but often become highly polyploid associated with an increase of the nuclear area, stimulating metabolic activity (Gerling and Orion, 1973; Hotta et al., 2001; Strand and Wong, 1991). In the insect fat body, DNA polyploidy caused by juvenile hormone stimulation was found to increase the transcription of vitellogenin (Dittmann et al., 1989; Nair et al., 1981), suggesting that teratocyte activity could be enhanced by polyploidy (Hotta et al., 2001).

Teratocytes supply nutrients to the developing parasitoid by digesting the host's fat body during early parasitoid larval stages when mouth parts are not yet formed. In the host-parasitoid system *P. separata*-*C. kariyai* system, triacylglycerol levels of the host decreased 6 days after parasitism but increased in the parasitoid's second instar larva from the 7th day (Nakamatsu et al., 2002). The increased lipase activity in the parasitoid larva, as well as the presence of lipid granules in the parasitoid midgut, confirmed the ingestion of host lipids by the parasitoid (Nakamatsu et al., 2002). Interestingly, teratocytes were attached to the host fat body and locally released collagenases (i.e., enzymes that break down the collagen sheath surrounding the host's fat body) to disrupt the host fat body matrix and release fat body cells (Nakamatsu et al., 2002). Teratocytes of other parasitoid species, such as the braconids *Microplitis mediator* or *M. pulchricornis*, seem to play a similar role (Qin et al., 2000; Suzuki and Tanaka, 2007).

Teratocytes release several other enzymes that can enhance host fat body digestion until complete consumption: a teratocyte-specific carboxylesterase, assumed to be involved in the hydrolysis of host lipids (*Dinocampus coccinellae*; (Gopalapillai et al., 2005), enolases and lipases (*A. ervi*, *Microplitis demolitor*, *D. coccinellae*; (Burke and Strand, 2014; Falabella et al., 2009; Kadono-Okuda et al., 1998), and cathepsin (Burke and Strand, 2014). These lipid-catabolic

enzymes were also found in the venom of some parasitoid species (Table 2). Finally, in the parasitoid *T. nigriceps*, teratocytes produced a chitinase during the last larval stage of the parasitoid that seemed to be involved in the digestion of host cuticle (i.e., lipids), aiding the egression of the parasitoid larvae from the host to pupate externally (Cônsoli et al., 2005).

In addition to fat body disruption, teratocytes also facilitate the transport of lipids from the host hemolymph to the developing larvae. For example, teratocytes of *A. ervi* produce an extracellular fatty acid binding protein that transports fatty acids in the host's hemolymph (Falabella et al., 2005, 2000; Pennacchio et al., 1999). This protein showed a high affinity for C14-C18 saturated fatty acids, oleic acid (C18:1), as well as a longer chain polyunsaturated fatty acid (arachidonic acid; C20:4) (Falabella et al., 2005). Immunolocalization revealed that the fatty acid binding protein was distributed around lipid particles abundantly present in the hemolymph of the parasitized host, but also in the external epidermal layer and the midgut lumen of parasitoid larvae (Caccia et al., 2012; Falabella et al., 2005). Altogether these findings suggest that *1)* fatty acids can be absorbed by the epidermal epithelium of the developing parasitoid, as was already found for amino acids and sugars (Caccia et al., 2012) and *2)* fatty acid binding protein transports key fatty acids in the host hemolymph to the growing parasitoid larva, which can subsequently be absorbed and stored as triacylglycerols (Caccia et al., 2012). Similar lipid transport enzymes were found in parasitoid venom, such as annexin, apolipophorins and calreticulin (Table 2) (Burke and Strand, 2014).

A decrease in teratocyte number during later stages of parasitoid development has been observed in several parasitoid species (de Buron and Beckage, 1997; Gopalapillai et al., 2005; Kadono-Okuda et al., 1995; Suzuki and Tanaka, 2007; Volkoff and Colazza, 1992). Such a decrease can be explained by the appearance of multiple bleb structures (i.e., teratocyte anatomical deformations resulting from the enlargement or coalescence of microvilli; Buron & Beckage, 1997; Zhang et al., 1994) on the teratocycte membrane, which is symptomatic of apoptotic cells (de Buron and Beckage, 1997; Hotta et al., 2001). Another explanation is that teratocytes are progressively consumed by the parasitoid larva(e) (Kadono-Okuda et al., 1995; Strand and Wong, 1991). Teratocytes absorb nutrients and produce proteins that can be released in host's hemolymph, but also stored inside the cells (Okuda and Kadono-Okuda, 1995). Indeed, teratocytes usually display a high abundance of proteins (e.g., glycoproteins, vitellogenin, amino-acids) as well as lipids (i.e., lipid droplets) (de Buron and Beckage, 1997; Gopalapillai et al., 2005; Kadono-Okuda et al., 1998; Okuda and Kadono-Okuda, 1995) that can constitute an additionnal source of nutrients for successful parasitoid development. On the contrary, no decrease in teratocyte number was observed in other parasitoids, such as *C. kariyai*, suggesting that in this species, the teratocytes are not consumed by the parasitoid and may have another potential role in host regulation or parasitoid development at a later stage (Hotta et al., 2001; Suzuki and Tanaka, 2007).

4 Conclusions and future perspectives

Parasitoids are fascinating creatures, particularly with regard to their lipid metabolism. Parasitoid larvae can mimic the fatty acid composition of their host, because there is no *de novo* trigylceride synthesis. The adults of many parasitoid species do not accumulate fat at all, with the exception of

some polyphagous species that typically develop on fat-poor hosts. More recently, fat synthesis and accumulation was found to vary in response to environmental conditions, i.e., is plastic, in the wasp *L. heterotoma*. This may be more common also in other parasitoid species (e.g., *L. boulardi, N. vitripennis*), although this remains to be explicitly tested. Having a system where fat synthesis and accumulation phenotypes vary opens up a lot of new research opportunities. For example, we need to know under which ecological conditions certain phenotypes are favored or not, preferably using natural populations. We can further continue to dig into the mechanisms underlying fat synthesis and accumulation (and the lack thereof), by experimentally manipulating parasitoid phenotypes (e.g., by changing host fat content; Enriquez et al., 2022). Several other research directions focusing on parasitoid lipids, including crossroads with biomedicine and parasitoid-microbe interactions, can also be envisioned (as highlighted by Visser et al., 2023) . This shows that, despite the considerable research effort into parasitoid lipid metabolism since the 60s, there is still a great diversity of research opportunities that can (and hopefully will be) pursued.

Parasitoids have become masters in host manipulation with the sheer number of mechanisms by which host lipid metabolism can be affected as proof. The diversity of parasitoids and thus host manipulation strategies may seem daunting to try and elucidate, because most responses are host and parasitoid-species specific. Using hosts and parasitoids that share an evolutionary history is, therefore, essential to further our understanding of host manipulation in a biologically meaningful way. *P. puparum* is one of the few species with which complementary studies have been performed to understand the entire process of host manipulation, from physiology to genes and their diversification (Wang et al., 2020a; Wang et al., 2021). Extending such thorough investigation also to other systems (i.e., hemolymph-feeders, koinobionts), also in a comparative context, will certainly enrich our understanding of host manipulation. There is further much to learn from parasitoid host manipulation strategies, even for our own benefit. For example, some venom components can be used in biological control of insect pests (Danneels et al., 2010; Moreau and Asgari, 2015). Virulence factors associated with teratocytes have also been proposed for use in transgenic plants able to resist pest attack (Merlin et al., 2021). Parasitoids can thus inspire the development of new technologies, perhaps even beyond insect pest control. For example, parasitoid venom components were suggested as potential pharmaceuticals against allergies, blood clotting, and as an antibiotic against microbial infections (Moreau and Asgari, 2015).

Acknowledgements

We are grateful to Jeff Harvey for providing insights into the biology of several parasitoid species and Jacintha Ellers for providing the original data used to make Box 1 Figure B1A. We would further like to thank Thomas Enriquez for making the cartoons used in Figure 1. MS and BV were supported by the Fonds National de Recherche Scientifique.

Tables

805 **Table 1. Overview of studies looking at the effect of parasitism on host lipid levels (mainly triglycerides) in the whole body, fat body and/or**
806 **hemolymph.** A distinction is made between the effects of parasitism in general, the venom (with or without the egg, i.e., envenomation), polydnaviruses
807 (with or without the egg, i.e., pseudoparasitization or injection), and teratocytes (with or without the egg, i.e., injection).

Parasitoid species	Parasitoid family	G/S	I/K	Gen/ Sp	Ecto/ Endo	Host stage attacked	H/T	Host species	Host order	Host Treat-ment	Host whole body	Host fat body	Host hemo-lymph	References
PARASITISM IN GENERAL														
Apanteles galleriae	Braconidae	S	K	Gen	Endo	L	T	*Achoria grisella*	Lepidoptera	Par	Sim	-	-	Nurullahoğlu et al., 2004
Cotesia congregata	Braconidae	G	K	Sp	Endo	L	H	*Manduca sexta*	Lepidoptera	Par	Low	-	-	Thompson and Redak, 2008
Cotesia flavipes	Braconidae	G	K	Sp	Endo	L	H	*Diatraea saccharalis*	Lepidoptera	Par	-	Sim	Low	Salvador and Cônsoli, 2008
								Diatraea flavipennella	Lepidoptera	Par	-	Sim	Low	dos Passos et al., 2019
Glyptapanteles liparidis	Braconidae	G	K	Sp	Endo	L	H	*Lymantria dispar*	Lepidoptera	Par	Low	-	Sim	Bischof and Ortel, 1996
Cardiochiles nigriceps	Braconidae	S	K	Sp	Endo	L	H	*Heliothis virescens*	Lepidoptera	Par	Sim	-	Sim	Barras et al., 1970
Trichomalopsis apanteloctena	Pteromalidae	S	I	Gen	Ecto	P	T	*Cotesia kariyai*	Hymenoptera	Par	Low	-	-	Nakamatsu and Tanaka, 2004
Trichopsidea oestracea	Nemestridae	S	K	Sp	Endo	N	T	*Chortoicetes terminifera*	Orthoptera	Par	Hig	-	-	Horwood and Hales, 1991
VENOM														
Bracon nigricans	Braconidae	G	I	Gen	Ecto	L	T	*Spodoptera littoralis*	Lepidoptera	Env	-	Low	Low	Becchimanzi et al., 2020, 2017
Habrobracon brevicornis	Braconidae	G	I	Gen	Ecto	L	T	*Galleria mellonella*	Lepidoptera	Env	-	Low	Hig	Kryukova et al., 2021
Lysiphlebia japonica	Braconidae	S	K	Gen	Endo	A	T	*Aphis gossypii*	Hemiptera	Par	Hig	-	-	Xueke et al., 2017
Euplectrus separatae	Eulophidae	G	K	Sp	Ecto	L	T	*Mythimna separata*	Lepidoptera	Par	-	Low	Hig	Nakamatsu and Tanaka, 2003
								Mythimna separata	Lepidoptera	Env	-	Low	Hig	Nakamatsu and Tanaka, 2004, 2003
Nasonia vitripennis	Pteromalidae	G	I	Gen	Ecto	P	T	*Sarcophaga bullata*	Diptera	Env	Hig	Hig	Hig	Rivers and Denlinger, 1995, 1994
								Sarcophaga bullata	Diptera	Env	-	-	Hig	Rivers et al., 1998
								Sarcophaga bullata	Diptera	Par	-	Hig	-	Rivers and Denlinger, 1995

								Sarcophaga bullata	Diptera	Env	-	Hig	-	Rivers and Denlinger, 1995
								Sarcodexia sternodontus	Diptera	Env	-	Low	Low	Rivers and Denlinger, 1995
								Phormia regina	Diptera	Env	-	Low	Hig	Rivers and Denlinger, 1995
								Musca domestica	Diptera	Env	-	Sim	Hig	Rivers and Denlinger, 1995
								Sarcophaga bullata	Diptera	Par	-	Hig	Hig	Rivers and Yoder, 1996
Trichomalopsis apanteloctena	Pteromalidae	S	I	Gen	Ecto	P	T	*Cotesia kariyai*	Hymenoptera	Env	Sim	-	-	Nakamatsu and Tanaka, 2004
Muscidifurax zaraptor	Pteromalidae	S	I	Gen	Ecto	P	T	*Sarcophaga bullata*	Diptera	Env	-	-	Hig	Rivers et al., 1998
Trichomalopsis near americana	Pteromalidae	G	I		Ecto	P	T	*Sarcophaga bullata*	Diptera	Env	-	Low	Hig	Rivers et al., 1998
Pteromalus puparum	Pteromalidae	G	I	Gen	Endo	P	T	*Pieris rapae*	Lepidoptera	Par	-	Hig	Hig	Wang et al., 2020a
Scleroderma sichuanensis	Bethylidae	G	I	Gen	Ecto	P	T	*Tenebrio molitor*	Coleoptera	Par	-	Low	Low	Zhuo et al., 2016
										Env	-	Sim	Low	Zhuo et al., 2016
TERATOCYTES														
Meteorus pulchricornis	Braconidae	S	K	Gen	Endo	L	T	*Mythimna separata*	Lepidoptera	Par	-	Low	-	Suzuki and Tanaka, 2007
Cotesia kariyai	Braconidae	G	K	Sp	Endo	L	H	*Mythimna separata*	Lepidoptera	Par	-	Low	-	Nakamatsu et al., 2002
Microplitis croceipes	Braconidae	S	K	Sp	Endo	L	H	*Heliothis virescens*	Lepidoptera	Par	-	Low	-	Zhang et al., 1997
										Inj	-	Low	-	Zhang et al., 1997
Dinocampus coccinellae	Braconidae	S	K	Gen	Endo	L/A	T	*Hippodamia convergens*	Coleoptera	Par	-	Low	-	Sluss, 1968
Dinocampus coccinellae	Braconidae	S	K	Gen	Endo	L/A	T	*Coccinella septempunctata*	Coleoptera	Par	-	Low	-	Gopalapillai et al., 2005
POLYDNAVIRUS														
Microplitis demolitor	Braconidae	S	K	Gen	Endo	L	H	*Chrysodeixis includens*	Lepidoptera	Inj	Low	-	-	Pruijssers et al., 2009
Cotesia vestalis	Braconidae	S	K	Sp	Endo	L	H	*Plutella xylostella*	Lepidoptera	Par	Low	Low	-	Wang et al., 2021
Chelonus inanitus	Braconidae	S	K	Gen	Endo	E/L	H	*Spodoptera littoralis*	Lepidoptera	Par	Hig	-	-	Kaeslin et al., 2005
										Ps	Hig	-	-	Kaeslin et al., 2005

G, Gregarious; S, solitary; I, Idiobiont; K, Koinobiont; Gen, Generalist; Sp, Specialist; Ecto, Ectoparasitoid; Endo, Endoparasitoid; A, Adult; E, Egg; L, Larva; N, Nymph; P, Pupa; H, Hemolymph-feeder; T, Tissue-feeder; Env, Envenomation; Inj, Injection; Par, Parasitization; Ps, Pseudoparasitization. Hig, Higher; Low, Lower, Sim, Similar. NB: *Trichomalopsis apanteloctena* is a hyperparasitoid.

809 **Table 2. Overview of genes and/or enzymes involved in lipid metabolism found in the venom of different parasitoid species.** We did not
810 distinguish between different homologs or orthologs. For each enzyme, a potential function is indicated based on functional studies in animals, including
811 humans.
812

Enzyme	Function	Species		
Lipid catabolism				
Carboxylesterase	Degradation of triglycerides, mainly long-chain triglyceride (Deng et al., 2021)	*Anisopteromalus calandrae*[1]	*Microplitis mediator*[12]	*Torymus sinensis*[23]
		Bracon nigricans[3]	*Ooencyrtus telenomicida*[15]	
		Hyposoter didymator[8]	*Psyttalia lounsburyi*[18]	
Cathepsin (D, L, J)	Digestive enzymes (e.g. degradation of the fat body) (Becchimanzi et al., 2020; Cristofoletti et al., 2003; Yang et al., 2020)	*B. nigricans*[3]	*Microctonus hyperodae*[11]	*Toxoneuron nigriceps*[24]
		Leptopilina heterotoma[10]	*M. mediator*[12]	*T. sinensis*[23]
		Microctonus aethiopoides[11]	*O. telenomicida*[15]	
Enolase	Mediates host tisue degradation (Falabella et al., 2009; Grossi et al., 2016)	*M. mediator*[12]	*Psyttalia concolor*[18]	*Tetrastichus brontispae*[22]
		O. telenomicida[15]	*P. lounsburyi*[18]	*T. nigriceps*[24]
Enoyl-coA hydratase	Metabolizing fatty acids in beta oxidation to produce both acetyl CoA and ATP	*B. nigricans*[3]	*O. telenomicida*[15]	
Fatty Acid Binding Protein	Fatty acid import, storage and export (also cholesterol and phospholipids) (Furuhashi and Hotamisligil, 2008)	*Diversinervus elegans*[6]	*M. mediator*[12]	
Lipase (3, A, H)	Digestion, transport, processing of dietary lipids (Wang et al., 2020b)	*B. nigricans*[3]	*O. telenomicida*[15]	*Pteromalus puparum*[21]
		Chelonus inanitus[4]	*M. aethiopoides*[11]	*P. lounsburyi*[18]
		Leptopilina boulardi[9]	*M. hyperodae*[11]	*T. sinensis*[23]
		L. heterotoma[10]	*Nasonia vitripennis*[13-14]	
		M. mediator[12]	*Pimpla hypochondriaca*[16]	
Low-density lipoprotein receptor	Low-density lipoprotein, mediating endocytosis of vitellogenin and lipophorin	*Aphidius ervi*[2]	*N. vitripennis*[13-14]	*P. puparum*[21]
		M. mediator[12]	*O. telenomicida*[15]	*T. sinensis*[23]
Low-density lipoprotein receptor-like venom protein	Central role in cholesterol and other lipoprotein metabolism (Scieuzo et al., 2021)	*A. calandrae*[1]	*N. vitripennis*[13-14]	*O. telenomicida*[15]
Phospholipase (A1, A2, B, C)	Hydrolyse phospholipid substrates at specific ester bonds (Richmond and Smith, 2011)	*B. nigricans*[3]	*L. heterotoma*[10]	*P. concolor*[18]
		Cotesia chilonis[5]	*M. mediator*[12]	*P. lounsburyi*[18]
		Eupelmus orientalis[7]	*O. telenomicida*[15]	*T. nigriceps*[24]

		D. elegans[6]	*Pimpla turionellae*[17]	
Vitellogenin receptor	Low density lipoprotein receptor that transports lipids into a recipient cell	*M. aethiopoides*[11]	*O. telenomicida*[15]	
Lipid transport (in the hemolymph)				
Annexin	Ca^{2+}-dependent lipid binding protein that could be involved in membrane transport processes	*L. heterotoma*[10] *M. mediator*[12]	*O. telenomicida*[15] *P. concolor*[18]	
Apolipophorin	Hemolymph lipid transport (Weers and Ryan, 2006)	*B. nigricans*[3] *D. elegans*[6] *L. heterotoma*[9]	*O. telenomicida*[15] *M. mediator*[12] *T. brontispae*[22]	*T. sinensis*[23]
Apolipoprotein D-like	Lipid transport processes in the insect hemolymph (Scieuzo et al., 2021)	*M. mediator*[12]	*O. telenomicida*[15]	*T. sinensis*[23]
Calreticulin	Chaperoning and regulation of Ca^{2+} homeostasis in the endoplasmic reticulum lumen	*A. calandrae*[1] *C. chilonis*[5] *H. didymator*[8] *M. hyperodae*[11] *M. aethiopoides*[11]	*M. mediator*[12] *N. vitripennis*[13-14] *O. telenomicida*[15] *P. puparum*[19,21] *P. concolor*[18]	 *P. lounsburyi*[18] *T. brontispae*[22] *T. sinensis*[23] *T. nigriceps*[24]
Odorant binding protein	Solubilizing and carrying free fatty acids released by lipases (Ishida et al., 2013; Pelosi et al., 2018)	*A. calandrae*[1] *B. nigricans*[3] *C. inanitus*[4] *L. heterotoma*[10]	*M. mediator*[12] *N. vitripennis*[13-14] *O. telenomicida*[15] *P. puparum*[20-21]	*T. brontispae*[22] *T. sinensis*[23]
Lipid synthesis				
3-oxoacyl-ACP reductase	Fatty acid synthesis and polyunsaturated fatty acid synthesis	*B. nigricans*[3]		
Fatty acid synthase	Catalyzing the *de novo* synthesis of fatty acids	*A. ervi*[2] *D. elegans*[6]	*M. mediator*[12] *O. telenomicida*[15]	*T. brontispae*[22]
n-acetyllactosaminide beta-n-acetylglucosaminyltransferase	Glycosphingolipid synthesis	*L. heterotoma*[10]	*O. telenomicida*[15]	*T. nigriceps*[24]
Phosphatidate phosphatase	Conversion of phosphatidate to diglyceride	*D. elegans*[6]	*O. telenomicida*[15]	*T. brontispae*[22]
Lipid storage				

Adipocyte plasma membrane-associated protein-like	Maturation of adipocytes and their capacity to store lipids (Sarjeant and Stephens, 2012)	*O. telenomicida*[15]	*T. sinensis*[23]	
Insulin-like growth factor-binding protein	Regulation of lipid metabolism, lipid accumulation, adipocyte differentiation (Kim and Lee, 2014; Pan et al., 2021)	*L. heterotoma*[10] *M. mediator*[12]	*O. telenomicida*[15] *T. sinensis*[23]	
Regucalcin	Ca^{2+} signaling, lipid accumulation in adipocytes (Doğan et al., 2021)	*B. nigricans*[3]	*M. mediator*[12]	*O. telenomicida*[15]

[1]Perkin et al., 2015; [2]Colinet et al., 2014; [3]Becchimanzi et al., 2020; [4]Vincent et al., 2010; [5]Teng et al., 2017; [6]Liu et al., 2017; [7]Doury et al., 1997; [8]Dorémus et al., 2013; [9]Colinet et al., 2013; [10]Heavner et al., 2013; [11]Crawford et al., 2008 [12]Lin et al., 2019; [13]de Graaf et al., 2010; [14]Sim and Wheeler, 2016; [15]Cusumano et al., 2018; [16]Dani et al., 2005; [17]Uçkan et al., 2006; [18]Mathé-Hubert et al., 2016; [19]Zhu et al., 2010; [20]Wang et al., 2015; [21]Yan et al., 2016 [22]Liu et al., 2018; [23]Scieuzo et al., 2021; [24]Laurino et al., 2016

813

References

Ali MR, Seo J, Lee D, Kim Y. Teratocyte-secreting proteins of an endoparasitoid wasp, *Cotesia plutellae*, prevent host metamorphosis by altering endocrine signals. Comp Biochem Physiol A Mol Integr Physiol 2013;166:251–62. https://doi.org/10.1016/j.cbpa.2013.06.028.

Arrese EL, Soulages JL. Insect Fat Body: Energy, Metabolism, and Regulation. Annu Rev Entomol 2010;55:207–25. https://doi.org/10.1146/annurev-ento-112408-085356.

Barlow JS. Effects of the diet on the composition of body fat in *Musca domestica* L. Can J Zool 1966;44:775–9. https://doi.org/10.1139/z66-077.

Barlow JS. Effects of diet on the composition of body fat in *Agria affinis* (Fallén). Can J Zool 1965;43:337–40. https://doi.org/10.1139/z65-032.

Barlow JS. Fatty acids in some insect and spider fats. Can J Biochem 1964;42:1365–74. https://doi.org/10.1139/o64-148.

Barras DJ, Joiner RL, Vinson SB. Neutral lipid composition of the tobacco budworm, *Heliothis virescens* (Fab.), as affected by its habitual parasite, *Cardiochiles nigriceps* viereck. Comp Biochem Physiol 1970;36:775–83. https://doi.org/10.1016/0010-406X(70)90532-3.

Becchimanzi A, Avolio M, Bostan H, Colantuono C, Cozzolino F, Mancini D, et al. Venomics of the ectoparasitoid wasp *Bracon nigricans*. BMC Genomics 2020;21:34. https://doi.org/10.1186/s12864-019-6396-4.

Becchimanzi A, Avolio M, Di Lelio I, Marinelli A, Varricchio P, Grimaldi A, et al. Host regulation by the ectophagous parasitoid wasp *Bracon nigricans*. J Insect Physiol 2017;101:73–81. https://doi.org/10.1016/j.jinsphys.2017.07.002.

Birsoy K, Festuccia WT, Laplante M. A comparative perspective on lipid storage in animals. J Cell Sci 2013;126:1541–52. https://doi.org/10.1242/jcs.104992.

Bischof C, Ortel J. The effects of parasitism by *Glyptapanteles liparidis* (Braconidae: Hymenoptera) on the hemolymph and total body composition of gypsy moth larvae (*Lymantria dispar*, Lymantriidae: Lepidoptera). Parasitol Res 1996;82:687–92. https://doi.org/10.1007/s004360050186.

Bracken GK, Barlow JS. Fatty acid composition of *Exeristes comstockii* (Cress) reared on different hosts. Can J Zool 1967;45:57–61. https://doi.org/10.1139/z67-007.

Burke GR, Strand MR. Systematic analysis of a wasp parasitism arsenal. Mol Ecol 2014;23:890–901. https://doi.org/10.1111/mec.12648.

de Buron I, Beckage NE. Developmental changes in teratocytes of the braconid wasp *Cotesia congregata* in larvae of the tobacco hornworm, *Manduca sexta*. J Insect Physiol 1997;43:915–30. https://doi.org/10.1016/S0022-1910(97)00056-5.

Caccia S, Grimaldi A, Casartelli M, Falabella P, Eguileor M de, Pennacchio F, et al. Functional analysis of a fatty acid binding protein produced by *Aphidius ervi* teratocytes. J Insect Physiol 2012;58:621–7. https://doi.org/10.1016/j.jinsphys.2011.12.019.

Caccia S, Leonardi MG, Casartelli M, Grimaldi A, de Eguileor M, Pennacchio F, et al. Nutrient absorption by *Aphidius ervi* larvae. J Insect Physiol 2005;51:1183–92. https://doi.org/10.1016/j.jinsphys.2005.06.010.

Carton Y, Bouletreau M, van Alphen JJM, van Lenteren JC. The Drosophila parasitic wasps. In: Ashburner M, Carson HL, Thompson JN, editors. The genetics and biology of Drosophila (volume 3), London: Academic Press; 1986, p. 348–94.

Casas J, Driessen G, Mandon N, Wielaard S, Desouhant E, van Alphen J, et al. Energy dynamics in a parasitoid foraging in the wild. J Anim Ecol 2003;72:691–7. https://doi.org/10.1046/j.1365-2656.2003.00740.x.

Colinet D, Anselme C, Deleury E, Mancini D, Poulain J, Azéma-Dossat C, et al. Identification of the main venom protein components of *Aphidius ervi*, a parasitoid wasp of the aphid model *Acyrthosiphon pisum*. BMC Genomics 2014;15:342. https://doi.org/10.1186/1471-2164-15-342.

Colinet D, Deleury E, Anselme C, Cazes D, Poulain J, Azema-Dossat C, et al. Extensive inter- and intraspecific venom variation in closely related parasites targeting the same host: The case of *Leptopilina* parasitoids of *Drosophila*. Insect Biochem Mol Biol 2013;43:601–11. https://doi.org/10.1016/j.ibmb.2013.03.010.

Colinet H, Hance T, Vernon P. Water Relations, Fat Reserves, Survival, and Longevity of a Cold-exposed Parasitic Wasp *Aphidius colemani* (Hymenoptera: Aphidiinae). Environ Entomol 2006;35:228–36. https://doi.org/10.1603/0046-225X-35.2.228.

Cônsoli FL, Brandt SL, Coudron TA, Vinson SB. Host regulation and release of parasitism-specific proteins in the system *Toxoneuron nigriceps–Heliothis virescens*. Comp Biochem Physiol B Biochem Mol Biol 2005;142:181–91. https://doi.org/10.1016/j.cbpc.2005.07.002.

Crawford AM, Brauning R, Smolenski G, Ferguson C, Barton D, Wheeler TT, et al. The constituents of *Microctonus sp.* parasitoid venoms. Insect Mol Biol 2008;17:313–24. https://doi.org/10.1111/j.1365-2583.2008.00802.x.

Cristofoletti PT, Ribeiro AF, Deraison C, Rahbé Y, Terra WR. Midgut adaptation and digestive enzyme distribution in a phloem feeding insect, the pea aphid *Acyrthosiphon pisum*. J Insect Physiol 2003;49:11–24. https://doi.org/10.1016/S0022-1910(02)00222-6.

Cuny MAC, Poelman EH. Evolution of koinobiont parasitoid host regulation and consequences for indirect plant defence. Evol Ecol 2022;36:299–319. https://doi.org/10.1007/s10682-022-10180-x.

Cusumano A, Duvic B, Jouan V, Ravallec M, Legeai F, Peri E, et al. First extensive characterization of the venom gland from an egg parasitoid: structure, transcriptome and functional role. J Insect Physiol 2018;107:68–80. https://doi.org/10.1016/j.jinsphys.2018.02.009.

Dahlman DL. Evaluation of teratocyte functions: An overview. Arch Insect Biochem Physiol 1990;13:159–66. https://doi.org/10.1002/arch.940130303.

Dahlman DL. Trehalose levels in parasitized and nonparasitized Tobacco Hornworm, *Manduca sexta*, Larvae. Ann Entomol Soc Am 1970;63:615–7.

Dahlman DL, Greene JR. Larval Hemolymph Protein Patterns in Tobacco Hornworms Parasitized by *Apanteles congregatus*. Ann Entomol Soc Am 1981;74:130–3. https://doi.org/10.1093/aesa/74.1.130.

Dahlman DL, Rana RL, Schepers EJ, Schepers T, DiLuna FA, Webb BA. A teratocyte gene from a parasitic wasp that is associated with inhibition of insect growth and development inhibits host protein synthesis. Insect Mol Biol 2003;12:527–34. https://doi.org/10.1046/j.1365-2583.2003.00439.x.

Dahlman DL, Vinson SB. Teratocytes: Developmental and Biochemical Characteristics. In: Beckage NE, Thompson S N, Federici BA, editors. Parasites and Pathogens of Insects. 1st ed., London: Academic Press; 1993, p. 145–65. https://doi.org/10.1016/B978-0-08-091649-1.50012-8.

Dani MP, Edwards JP, Richards EH. Hydrolase activity in the venom of the pupal endoparasitic wasp, *Pimpla hypochondriaca*. Comp Biochem Physiol B Biochem Mol Biol 2005;141:373–81. https://doi.org/10.1016/j.cbpc.2005.04.010.

Danneels EL, Rivers DB, de Graaf DC. Venom proteins of the parasitoid wasp *Nasonia vitripennis*: Recent discovery of an untapped pharmacopee. Toxins (Basel) 2010;2:494–516. https://doi.org/10.3390/toxins2040494.

Delobel B, Pageaux JF. Influence de l'alimentation sur la composition en acides gras totaux de diptères tachinaires. Entomol Exp Appl 1981;29:281–8. https://doi.org/10.1111/j.1570-7458.1981.tb03070.x.

Deng Y, Kim BY, Lee KY, Yoon HJ, Wan H, Li J, et al. Lipolytic Activity of a Carboxylesterase from Bumblebee (*Bombus ignitus*) Venom. Toxins (Basel) 2021;13:239. https://doi.org/10.3390/toxins13040239.

Desjardins CA, Perfectti F, Bartos JD, Enders LS, Werren JH. The genetic basis of interspecies host preference differences in the model parasitoid *Nasonia*. Heredity (Edinb) 2010;104:270–7. https://doi.org/10.1038/hdy.2009.145.

Dittmann F, Kogan PH, Hagedorn HH. Ploidy levels and DNA synthesis in fat body cells of the adult mosquito, *Aedes aegypti*: The role of juvenile hormone. Arch Insect Biochem Physiol 1989;12:133–43. https://doi.org/10.1002/arch.940120302.

Doğan C, Hänniger S, Heckel DG, Coutu C, Hegedus DD, Crubaugh L, et al. Characterization of calcium signaling proteins from the fat body of the Colorado Potato Beetle, *Leptinotarsa decemlineata* (Coleoptera: Chrysomelidae): Implications for diapause and lipid metabolism. Insect Biochem Mol Biol 2021;133:103549. https://doi.org/10.1016/j.ibmb.2021.103549.

Dorémus T, Urbach S, Jouan V, Cousserans F, Ravallec M, Demettre E, et al. Venom gland extract is not required for successful parasitism in the polydnavirus-associated endoparasitoid *Hyposoter didymator* (Hym. Ichneumonidae) despite the presence of numerous novel and conserved venom proteins. Insect Biochem Mol Biol 2013;43:292–307. https://doi.org/10.1016/j.ibmb.2012.12.010.

Doury G, Bigot Y, Periquet G. Physiological and Biochemical Analysis of Factors in the Female Venom Gland and Larval Salivary Secretions of the Ectoparasitoid Wasp *Eupelmus orientalis*. J Insect Physiol 1997;43:69–81. https://doi.org/10.1016/S0022-1910(96)00053-4.

Edson KM, Vinson SB. Nutrient absorption by the anal vesicle of the braconid wasp, *Microplitis croceipes*. J Insect Physiol 1977;23:5–8. https://doi.org/10.1016/0022-1910(77)90101-9.

Eggleton P, Belshaw R. Comparisons of dipteran, hymenopteran and coleopteran parasitoids: provisional phylogenetic explanations. Biol J Linn Soc 1993;48:213–26. https://doi.org/10.1006/bijl.1993.1015.

Eggleton P, Belshaw R. Insect parasitoids: an evolutionary overview. Philos Trans R Soc B 1992;337:1–20. https://doi.org/10.1098/rstb.1992.0079.

Eijs IEM, Ellers J, van Duinen G-J. Feeding strategies in drosophilid parasitoids: the impact of natural food resources on energy reserves in females. Ecol Entomol 1998;23:133–8. https://doi.org/10.1046/j.1365-2311.1998.00117.x.

Ellers J. Fat and Eggs: an Alternative Method To Measure the Trade-Off Between Survival and Reproduction in Insect Parasitoids. Neth J Zool 1996;46:227–35. https://doi.org/10.1163/156854295X00186.

Ellers J, van Alphen JJM. A trade-off between diapause duration and fitness in female parasitoids. Ecol Entomol 2002;27:279–84. https://doi.org/10.1046/j.1365-2311.2002.00421.x.

Ellers J, van Alphen JJM. Life history evolution in *Asobara tabida*: plasticity in allocation of fat reserves to survival and reproduction. J Evol Biol 1997;10:771–85. https://doi.org/10.1046/j.1420-9101.1997.10050771.x.

Ellers J, van Alphen JJM, Sevenster JG. A field study of size-fitness relationships in the parasitoid *Asobara tabida*. J Anim Ecol 1998;67:318–24. https://doi.org/10.1046/j.1365-2656.1998.00195.x.

Ellers J, Bax M, van Alphen JJM. Seasonal changes in female size and its relation to reproduction in the parasitoid *Asobara tabida*. Oikos 2001;92:209–314. https://doi.org/10.1034/j.1600-0706.2001.920213.x.

Enriquez T, Lievens V, Nieberding CM, Visser B. Pupal size as a proxy for fat content in laboratory-reared and field-collected *Drosophila* species. Sci Rep 2022;12:12855. https://doi.org/10.1038/s41598-022-15325-0.

Falabella P, Perugino G, Caccialupi P, Riviello L, Varricchio P, Tranfaglia A, et al. A novel fatty acid binding protein produced by teratocytes of the aphid parasitoid *Aphidius ervi*. Insect Mol Biol 2005;14:195–205. https://doi.org/10.1111/j.1365-2583.2004.00548.x.

Falabella P, Riviello L, De Stradis ML, Stigliano C, Varricchio P, Grimaldi A, et al. *Aphidius ervi* teratocytes release an extracellular enolase. Insect Biochem Mol Biol 2009;39:801–13. https://doi.org/10.1016/j.ibmb.2009.09.005.

Falabella P, Tremblay E, Pennacchio F. Host regulation by the aphid parasitoid *Aphidius ervi*: the role of teratocytes. Entomol Exp Appl 2000;97:1–9. https://doi.org/10.1046/j.1570-7458.2000.00710.x.

Forbes AA, Bagley RK, Beer MA, Hippee AC, Widmayer HA. Quantifying the unquantifiable: why Hymenoptera, not Coleoptera, is the most speciose animal order. BMC Ecol 2018;18:21. https://doi.org/10.1186/s12898-018-0176-x.

Ford PS, van Heusden MC. Triglyceride-Rich Lipophorin in *Aedes aegypti* (Diptera: Culicidae). J Med Entomol 1994;31:435–41. https://doi.org/10.1093/jmedent/31.3.435.

Furuhashi M, Hotamisligil GS. Fatty acid-binding proteins: role in metabolic diseases and potential as drug targets. Nat Rev Drug Discov 2008;7:489–503. https://doi.org/10.1038/nrd2589.

Gerling D, Orion T. The giant cells produced by *Telenomus remus* (Hymenoptera: Scelionidae). J Invertebr Pathol 1973;21:164–71. https://doi.org/10.1016/0022-2011(73)90197-3.

Giron David, Casas J. Mothers reduce egg provisioning with age. Ecol Lett 2003a;6:273–7. https://doi.org/10.1046/j.1461-0248.2003.00429.x.

Giron D, Casas J. Lipogenesis in an adult parasitic wasp. J Insect Physiol 2003b;49:141–7. https://doi.org/10.1016/S0022-1910(02)00258-5.

Godfray HCJ. Parasitoids: Behavioral and Evolutionary Ecology. vol. 67. Princeton University Press; 1994. https://doi.org/10.2307/j.ctvs32rmp.

González D, Zhao Q, McMahan C, Velasquez D, Haskins WE, Sponsel V, et al. The major antennal chemosensory protein of red imported fire ant workers. Insect Mol Biol 2009;18:395–404. https://doi.org/10.1111/j.1365-2583.2009.00883.x.

Gopalapillai R, Kadono-Okuda K, Okuda T. Molecular cloning and analysis of a novel teratocyte-specific carboxylesterase from the parasitic wasp, *Dinocampus coccinellae*. Insect Biochem Mol Biol 2005;35:1171–80. https://doi.org/10.1016/j.ibmb.2005.05.010.

de Graaf DC, Aerts M, Brunain M, Desjardins CA, Jacobs FJ, Werren JH, et al. Insights into the venom composition of the ectoparasitoid wasp *Nasonia vitripennis* from bioinformatic and

proteomic studies. Insect Mol Biol 2010;19:11–26. https://doi.org/10.1111/j.1365-2583.2009.00914.x.

Grimaldi A, Caccia S, Congiu T, Ferrarese R, Tettamanti G, Rivas-Pena M, et al. Structure and function of the extraembryonic membrane persisting around the larvae of the parasitoid *Toxoneuron nigriceps*. J Insect Physiol 2006;52:870–80. https://doi.org/10.1016/j.jinsphys.2006.05.011.

Grossi G, Grimaldi A, Cardone RA, Monné M, Reshkin SJ, Girardello R, et al. Extracellular matrix degradation via enolase/plasminogen interaction: Evidence for a mechanism conserved in Metazoa. Biol Cell 2016;108:161–78. https://doi.org/10.1111/boc.201500095.

Harvey JA. *Venturia canescens* parasitizing *Galleria mellonella* and *Anagasta kuehniella*: is the parasitoid a conformer or regulator? J Insect Physiol 1996;42:1017–25. https://doi.org/10.1016/S0022-1910(96)00069-8.

Harvey JA, Malcicka M. Nutritional integration between insect hosts and koinobiont parasitoids in an evolutionary framework. Entomol Exp Appl 2016;159:181–8. https://doi.org/10.1111/eea.12426.

Harvey JA, Sano T, Tanaka T. Differential host growth regulation by the solitary endoparasitoid, *Meteorus pulchricornis* in two hosts of greatly differing mass. J Insect Physiol 2010;56:1178–83. https://doi.org/10.1016/j.jinsphys.2010.03.018.

Harvey JA, Strand MR. The Developmental Strategies of Endoparasitoid Wasps Vary with Host Feeding Ecology. Ecology 2002;83:2439–51. https://doi.org/10.2307/3071805.

Harvey JA, Wagenaar R, Bezemer TM. Interactions to the fifth trophic level: secondary and tertiary parasitoid wasps show extraordinary efficiency in utilizing host resources. J Anim Ecol 2009;78:686–92. https://doi.org/10.1111/j.1365-2656.2008.01516.x.

Hayakawa Y. Inhibition of lipid transport in insects by a parasitic factor. Comp Biochem Physiol B: Comp Biochem 1987;87:279–83. https://doi.org/10.1016/0305-0491(87)90140-4.

Heavner ME, Gueguen G, Rajwani R, Pagan PE, Small C, Govind S. Partial venom gland transcriptome of a *Drosophila* parasitoid wasp, *Leptopilina heterotoma*, reveals novel and shared bioactive profiles with stinging Hymenoptera. Gene 2013;526:195–204. https://doi.org/10.1016/j.gene.2013.04.080.

Hoddle MS, van Driesche RG, Sanderson JP. Biology and use of the whitefly parasitoid *Encarsia formosa*. Annu Rev Entomol 1998;43:645–69. https://doi.org/10.1146/annurev.ento.43.1.645.

Horwood MA, Hales DF. Fat body changes in a locust, *Chortoicetes terminifera* (Walker) (Orthoptera: Acrididae), parasitized by a nemestrinid fly. Arch Insect Biochem Physiol 1991;17:53–63. https://doi.org/10.1002/arch.940170107.

Hotta M, Okuda T, Tanaka T. *Cotesia kariyai* teratocytes: growth and development. J Insect Physiol 2001;47:31–41. https://doi.org/10.1016/S0022-1910(00)00089-5.

Ishida Y, Ishibashi J, Leal WS. Fatty Acid Solubilizer from the Oral Disk of the Blowfly. PLoS One 2013;8:e51779. https://doi.org/10.1371/journal.pone.0051779.

Jervis MA. Insects As Natural Enemies. Dordrecht: Springer Netherlands; 2005. https://doi.org/10.1007/978-1-4020-2625-6.

Jervis MA, Ellers J, Harvey JA. Resource Acquisition, Allocation, and Utilization in Parasitoid Reproductive Strategies. Annu Rev Entomol 2008;53:361–85. https://doi.org/10.1146/annurev.ento.53.103106.093433.

Jervis MA, Heimpel GE, Ferns PN, Harvey JA, Kidd NAC. Life-history strategies in parasitoid wasps: a comparative analysis of 'ovigeny.' J Anim Ecol 2001;70:442–58. https://doi.org/10.1046/j.1365-2656.2001.00507.x.

Jones D, Barlow JS, Thompson SN. *Exeristes*, *Itoplectis*, *Aphaereta*, *Brachymeria*, and *Hyposoter* species: In vitro glyceride synthesis and regulation of fatty acid composition. Exp Parasitol 1982;54:340–51. https://doi.org/10.1016/0014-4894(82)90043-1.

Kadono-Okuda K, Sakurai H, Takeda S, Okuda T. Synchronous growth of a parasitoid, *Perilitus coccinellae*, and teratocytes with the development of the host, *Coccinella septempunctata*. Entomol Exp Appl 1995;75:145–9. https://doi.org/10.1111/j.1570-7458.1995.tb01920.x.

Kadono-Okuda K, Weyda F, Okuda T. *Dinocampus* (=*Perilitus*) coccinellae teratocyte-specific polypeptide: its accumulative property, localization and characterization. J Insect Physiol 1998;44:1073–80. https://doi.org/10.1016/S0022-1910(98)00063-8.

Kaeslin M, Pfister-Wilhelm R, Lanzrein B. Influence of the parasitoid *Chelonus inanitus* and its polydnavirus on host nutritional physiology and implications for parasitoid development. J Insect Physiol 2005;51:1330–9. https://doi.org/10.1016/j.jinsphys.2005.08.003.

Kim MS, Lee D-Y. Serum insulin-like growth factor-binding protein-3 level correlated with glycemic control and lipid profiles in children and adolescents with type 1 diabetes. Journal of Pediatric Endocrinology and Metabolism 2014;27. https://doi.org/10.1515/jpem-2013-0358.

Kraaijeveld K, Neleman P, Mariën J, de Meijer E, Ellers J. Genomic Resources for *Goniozus legneri* , *Aleochara bilineata* and *Paykullia maculata* , Representing Three Independent Origins of the Parasitoid Lifestyle in Insects. G3 Genes Genom Genet 2019;9:987–91. https://doi.org/10.1534/g3.119.300584.

Kryukova NA, Mozhaytseva KA, Rotskaya UN, Glupov V V. *Galleria mellonella* larvae fat body disruption (Lepidoptera: Pyralidae) caused by the venom of *Habrobracon brevicornis* (Hymenoptera: Braconidae). Arch Insect Biochem Physiol 2021;106:e21746. https://doi.org/10.1002/arch.21746.

Lafferty KD, Kuris AM. Trophic strategies, animal diversity and body size. Trends Ecol Evol 2002;17:507–13. https://doi.org/10.1016/S0169-5347(02)02615-0.

1058 Lahti DC, Johnson NA, Ajie BC, Otto SP, Hendry AP, Blumstein DT, et al. Relaxed selection in
1059 the wild. Trends Ecol Evol 2009;24:487–96. https://doi.org/10.1016/j.tree.2009.03.010.

1060 Lammers M, Kraaijeveld K, Mariën J, Ellers J. Gene expression changes associated with the
1061 evolutionary loss of a metabolic trait: lack of lipogenesis in parasitoids. BMC Genomics
1062 2019;20:309. https://doi.org/10.1186/s12864-019-5673-6.

1063 Le Lann C, Visser B, Mériaux M, Moiroux J, van Baaren J, van Alphen JJM, et al. Rising
1064 temperature reduces divergence in resource use strategies in coexisting parasitoid species.
1065 Oecologia 2014;174:967–77. https://doi.org/10.1007/s00442-013-2810-9.

1066 Laurino S, Grossi G, Pucci P, Flagiello A, Bufo SA, Bianco G, et al. Identification of major
1067 *Toxoneuron nigriceps* venom proteins using an integrated transcriptomic/proteomic approach.
1068 Insect Biochem Mol Biol 2016;76:49–61. https://doi.org/10.1016/j.ibmb.2016.07.001.

1069 Lease HM, Wolf BO. Lipid content of terrestrial arthropods in relation to body size, phylogeny,
1070 ontogeny and sex. Physiol Entomol 2011;36:29–38. https://doi.org/10.1111/j.1365-
1071 3032.2010.00767.x.

1072 Lee JC, Heimpel GE, Leibee GL. Comparing floral nectar and aphid honeydew diets on the
1073 longevity and nutrient levels of a parasitoid wasp. Entomol Exp Appl 2004;111:189–99.
1074 https://doi.org/10.1111/j.0013-8703.2004.00165.x.

1075 Lin Z, Wang R-J, Cheng Y, Du J, Volovych O, Han L-B, et al. Insights into the venom protein
1076 components of *Microplitis mediator*, an endoparasitoid wasp. Insect Biochem Mol Biol
1077 2019;105:33–42. https://doi.org/10.1016/j.ibmb.2018.12.013.

1078 Liu N-Y, Wang J-Q, Zhang Z-B, Huang J-M, Zhu J-Y. Unraveling the venom components of an
1079 encyrtid endoparasitoid wasp *Diversinervus elegans*. Toxicon 2017;136:15–26.
1080 https://doi.org/10.1016/j.toxicon.2017.06.011.

1081 Liu N-Y, Xu Z-W, Yan W, Ren X-M, Zhang Z-Q, Zhu J-Y. Venomics reveals novel ion transport
1082 peptide-likes (ITPLs) from the parasitoid wasp *Tetrastichus brontispae*. Toxicon 2018;141:88–93.
1083 https://doi.org/10.1016/j.toxicon.2017.11.008.

1084 Liu T-X, Stansly PA, Gerling D. Whitefly parasitoids: Distribution, life history, bionomics, and
1085 utilization. Annu Rev Entomol 2015;60:273–92. https://doi.org/10.1146/annurev-ento-010814-
1086 021101.

1087 Luo S, Li J, Liu X, Lu Z, Pan W, Zhang Q, et al. Effects of six sugars on the longevity, fecundity
1088 and nutrient reserves of *Microplitis mediator*. Biol Control 2010;52:51–7.
1089 https://doi.org/10.1016/j.biocontrol.2009.09.002.

1090 Mathé-Hubert H, Colinet D, Deleury E, Belghazi M, Ravallec M, Poulain J, et al. Comparative
1091 venomics of *Psyttalia lounsburyi* and *P. concolor*, two olive fruit fly parasitoids: a hypothetical
1092 role for a GH1 β-glucosidase. Sci Rep 2016;6:35873. https://doi.org/10.1038/srep35873.

Matthews RW, González JM, Matthews JR, Deyrup LD. Biology of the parasitoid *Melittobia* (Hymenoptera: Eulophidae). Annu Rev Entomol 2009;54:251–66. https://doi.org/10.1146/annurev.ento.54.110807.090440.

Merlin BL, Pino LE, Peres LEP, Prataviera F, Ortega EMM, Cônsoli FL. Beyond host specificity: the biotechnological exploitation of chitolectin from teratocytes of *Toxoneuron nigriceps* to control non-permissive hosts. J Pest Sci (2004) 2021;94:713–27. https://doi.org/10.1007/s10340-020-01290-y.

Moiroux J, le Lann C, Seyahooei MA, Vernon P, Pierre J-S, van Baaren J, et al. Local adaptations of life-history traits of a *Drosophila* parasitoid, *Leptopilina boulardi*: does climate drive evolution? Ecol Entomol 2010;35:727–36. https://doi.org/10.1111/j.1365-2311.2010.01233.x.

Mondy N, Corio-Costet M-F, Bodin A, Mandon N, Vannier F, Monge J-P. Importance of sterols acquired through host feeding in synovigenic parasitoid oogenesis. J Insect Physiol 2006;52:897–904. https://doi.org/10.1016/j.jinsphys.2006.03.007.

Morales J, Medina P, Viñuela E. The influence of two endoparasitic wasps, *Hyposoter didymator* and *Chelonus inanitus*, on the growth and food consumption of their host larva *Spodoptera littoralis*. BioControl 2007;52:145–60. https://doi.org/10.1007/s10526-006-9026-4.

Moreau SJM, Asgari S. Venom proteins from parasitoid wasps and their biological functions. Toxins (Basel) 2015;7:2385–412. https://doi.org/10.3390/toxins7072385.

Muller D, Giron D, Desouhant E, Rey B, Casas J, Lefrique N, et al. Maternal age affects offspring nutrient dynamics. J Insect Physiol 2017;101:123–31. https://doi.org/10.1016/j.jinsphys.2017.07.011.

Multerer M-T, Wendler M, Ruther J. The biological significance of lipogenesis in *Nasonia vitripennis*. Proc Biol Sci 2022;289:20220208. https://doi.org/10.1098/rspb.2022.0208.

Nair KK, Chen TT, Wyatt GR. Juvenile hormone-stimulated polyploidy in adult locust fat body. Dev Biol 1981;81:356–60. https://doi.org/10.1016/0012-1606(81)90300-6.

Nakamatsu Y, Fujii S, Tanaka T. Larvae of an endoparasitoid, *Cotesia kariyai* (Hymenoptera: Braconidae), feed on the host fat body directly in the second stadium with the help of teratocytes. J Insect Physiol 2002;48:1041–52. https://doi.org/10.1016/S0022-1910(02)00192-0.

Nakamatsu Y, Tanaka T. Food Resource Use of Hyperparasitoid *Trichomalopsis apanteloctena* (Hymenoptera: Pteromalidae), an Idiobiotic Ectoparasitoid. Ann Entomol Soc Am 2004;97:994–9. https://doi.org/10.1603/0013-8746(2004)097[0994:FRUOHT]2.0.CO;2.

Nakamatsu Y, Tanaka T. Venom of ectoparasitoid, *Euplectrus sp.* near plathypenae (Hymenoptera: Eulophidae) regulates the physiological state of *Pseudaletia separata* (Lepidoptera: Noctuidae) host as a food resource. J Insect Physiol 2003;49:149–59. https://doi.org/10.1016/S0022-1910(02)00261-5.

Nurullahoğlu Z, Kan U, Sak O, Ergi E. Total Lipid and Fatty Acid Composition of *Apanteles galleriae* and Its Parasitized Host. Ann Entomol Soc Am 2004;97:1000–6. https://doi.org/10.1603/0013-8746(2004)097[1000:TLAFAC]2.0.CO;2.

Okuda T, Kadono-Okuda K. *Perilitus coccinellae* teratocyte polypeptide: Evidence for production of a teratocyte-specific 540 kDa protein. J Insect Physiol 1995;41:819–25. https://doi.org/10.1016/0022-1910(95)00009-J.

Pan J, Cen L, Zhou T, Yu M, Chen X, Jiang W, et al. Insulin-like growth factor binding protein 1 ameliorates lipid accumulation and inflammation in nonalcoholic fatty liver disease. J Gastroenterol Hepatol 2021;36:3438–47. https://doi.org/10.1111/jgh.15627.

dos Passos EM, Wanderley-Teixeira V, Porto ALF, Marques EJ, Teixeira ÁAC, Silva FSO. *Cotesia flavipes* Cameron (Hymenoptera: Braconidae) alters the nutrients in the hemolymph, fat body, and cytochemistry of *Diatraea flavipennella* Box (Lepidoptera: Crambidae) hemocytes. Semin Cienc Agrar 2019;40:539–54. https://doi.org/10.5433/1679-0359.2019v40n2p539.

Pedata PA, Garonna AP, Zabatta A, Zeppa P, Romani R, Isidoro N. Development and morphology of teratocytes in *Encarsia berlesei* and *Encarsia citrina*: first record for Chalcidoidea. J Insect Physiol 2003;49:1063–71. https://doi.org/10.1016/j.jinsphys.2003.08.003.

Pelosi P, Zhu J, Knoll W. Odorant-Binding Proteins as Sensing Elements for Odour Monitoring. Sensors 2018;18:3248. https://doi.org/10.3390/s18103248.

Pennacchio F, Fanti P, Falabella P, Digilio MC, Bisaccia F, Tremblay E. Development and nutrition of the braconid wasp, *Aphidius ervi* in aposymbiotic host aphids. Arch Insect Biochem Physiol 1999;40:53–63. https://doi.org/10.1002/(SICI)1520-6327(1999)40:1<53::AID-ARCH6>3.0.CO;2-J.

Pennacchio F, Strand MR. Evolution of developmental strategies in parasitic Hymenoptera. Annu Rev Entomol 2006;51:233–58. https://doi.org/10.1146/annurev.ento.51.110104.151029.

Pennacchio F, Vinson SB, Tremblay E. Morphology and ultrastructure of the serosal cells (teratocytes) in *Cardiochiles nigriceps* Viereck (Hymenoptera: Braconidae) embryos. Int J Insect Morphol Embryol 1994;23:93–104. https://doi.org/10.1016/0020-7322(94)90003-5.

Perez-Riverol A, Lasa AM, dos Santos-Pinto JRA, Palma MS. Insect venom phospholipases A1 and A2: Roles in the envenoming process and allergy. Insect Biochem Mol Biol 2019;105:10–24. https://doi.org/10.1016/j.ibmb.2018.12.011.

Perkin LC, Friesen KS, Flinn PW, Oppert B. Venom gland components of the ectoparasitoid wasp, *Anisopteromalus calandrae*. J Venom Res 2015;6:19–37.

Poirié M, Colinet D, Gatti JL. Insights into function and evolution of parasitoid wasp venoms. Curr Opin Insect Sci 2014;6:52–60. https://doi.org/10.1016/j.cois.2014.10.004.

Prager L, Bruckmann A, Ruther J. De novo biosynthesis of fatty acids from α-D-glucose in parasitoid wasps of the *Nasonia* group. Insect Biochem Mol Biol 2019;115:103256. https://doi.org/10.1016/j.ibmb.2019.103256.

Pruijssers AJ, Falabella P, Eum JH, Pennacchio F, Brown MR, Strand MR. Infection by a symbiotic polydnavirus induces wasting and inhibits metamorphosis of the moth *Pseudoplusia includens*. Journal of Experimental Biology 2009;212:2998–3006. https://doi.org/10.1242/jeb.030635.

Qin Q, Gong H, Ding T. Two Collagenases Are Secreted by Teratocytes from *Microplitis mediator* (Hymenoptera: Braconidae) Cultured in Vitro. J Invertebr Pathol 2000;76:79–80. https://doi.org/10.1006/jipa.2000.4950.

Quicke DL. Parasitic Wasps. London: Springer; 1997.

Quicray M, Wilhelm L, Enriquez T, He S, Scheifler M, Visser B. The *Drosophila* -parasitizing wasp *Leptopilina heterotoma* : A comprehensive model system in ecology and evolution. Ecol Evol 2023;13:e9625. https://doi.org/10.1002/ece3.9625.

Richmond GS, Smith TK. Phospholipases A1. Int J Mol Sci 2011;12:588–612. https://doi.org/10.3390/ijms12010588.

Rivero A, West S. The physiological costs of being small in a parasitic wasp. Evol Ecol Res 2002;4:407–20.

Rivers DB, Denlinger DL. Venom-Induced Alterations in Fly Lipid Metabolism and Its Impact on Larval Development of the Ectoparasitoid *Nasonia vitripennis* (Walker) (Hymenoptera: Pteromalidae). J Invertebr Pathol 1995;66:104–10. https://doi.org/10.1006/jipa.1995.1071.

Rivers DB, Denlinger DL. Redirection of metabolism in the flesh fly, *Sarcophaga bullata*, following envenomation by the ectoparasitoid *Nasonia vitripennis* and correlation of metabolic effects with the diapause status of the host. J Insect Physiol 1994;40:207–15. https://doi.org/10.1016/0022-1910(94)90044-2.

Rivers DB, Pagnotta MA, Huntington ER. Reproductive Strategies of 3 Species of Ectoparasitic Wasps Are Modulated by the Response of the Fly Host *Sarcophaga bullata* (Diptera: Sarcophagidae) to Parasitism. Ann Entomol Soc Am 1998;91:458–65. https://doi.org/10.1093/aesa/91.4.458.

Rivers DB, Rocco MM, Frayha AR. Venom from the ectoparasitic wasp *Nasonia vitripennis* increases Na+ influx and activates phospholipase C and phospholipase A2 dependent signal transduction pathways in cultured insect cells. Toxicon 2002;40:9–21. https://doi.org/10.1016/S0041-0101(01)00132-5.

Rivers DB, Yoder JA. Site-specific effects of parasitism on water balance and lipid content of the parasitic wasp *Nasonia vitripennis* (Hymenoptera: Pteromalidae). Eur J Entomol 1996;93:75–82.

Rouleux-Bonnin F, Renault S, Rabouille A, Periquet G, Bigot Y. Free serosal cells originating from the embryo of the wasp *Diadromus pulchellus* in the pupal body of parasitized leek-moth, *Acrolepiosis assectella*. Are these cells teratocyte-like? J Insect Physiol 1999;45:479–84. https://doi.org/10.1016/S0022-1910(98)00149-8.

Ruther J, Prager L, Pokorny T. Parasitic wasps do not lack lipogenesis. Proc R Soc B: Biol Sci 2021;288:20210548. https://doi.org/10.1098/rspb.2021.0548.

Salvador G, Cônsoli FL. Changes in the hemolymph and fat body metabolites of *Diatraea saccharalis* (Fabricius) (Lepidoptera: Crambidae) parasitized by *Cotesia flavipes* (Cameron) (Hymenoptera: Braconidae). Biol Control 2008;45:103–10. https://doi.org/10.1016/j.biocontrol.2007.12.007.

Salvia R, Grimaldi A, Girardello R, Scieuzo C, Scala A, Bufo SA, et al. *Aphidius ervi* Teratocytes Release Enolase and Fatty Acid Binding Protein Through Exosomal Vesicles. Front Physiol 2019;10:715. https://doi.org/10.3389/fphys.2019.00715.

Sarjeant K, Stephens JM. Adipogenesis. Cold Spring Harb Perspect Biol 2012;4:a008417–a008417. https://doi.org/10.1101/cshperspect.a008417.

Scieuzo C, Salvia R, Franco A, Pezzi M, Cozzolino F, Chicca M, et al. An integrated transcriptomic and proteomic approach to identify the main *Torymus sinensis* venom components. Sci Rep 2021;11:5032. https://doi.org/10.1038/s41598-021-84385-5.

Service PM. Physiological Mechanisms of Increased Stress Resistance in *Drosophila melanogaster* Selected for Postponed Senescence. Physiol Zool 1987;60:321–6.

Seyahooei M, van Alphen JJM, Kraaijeveld K. Genetic structure of *Leptopilina boulardi* populations from different climatic zones of Iran. BMC Ecol 2011;11:4. https://doi.org/10.1186/1472-6785-11-4.

Seyahooei MA, Kraaijeveld K, Bagheri A, van Alphen JJM. Adult size and timing of reproduction in five species of *Asobara* parasitoid wasps. Insect Sci 2020;27:1334–45. https://doi.org/10.1111/1744-7917.12728.

Shelby KS, Habibi J, Puttler B. An Ultrastructural and Fluorescent Study of the Teratocytes of *Microctonus aethiopoides* Loan (Hymenoptera: Braconidae) from the Hemocoel of Host Alfalfa Weevil, *Hypera postica* (Gyllenhal) (Coleoptera: Curculionidae). Psyche (Camb Mass);2014:1–9. https://doi.org/10.1155/2014/652518.

Sheng S, Zhang X, Zheng Y, Wang Jiao, Zhou Y, Liao C, et al. Effect of six sugars on the longevity, oviposition performance and nutrition accumulation in an endoparasitoid, *Meteorus pulchricornis* (Hymenoptera: Braconidae). J Asia Pac Entomol 2019;22:263–8. https://doi.org/10.1016/j.aspen.2019.01.010.

1231 Shi M, Dong S, Li M, Yang Y, Stanley D, Chen X. The Endoparasitoid, *Cotesia vestalis*, Regulates
1232 Host Physiology by Reprogramming the Neuropeptide Transcriptional Network. Sci Rep
1233 2015;5:8173. https://doi.org/10.1038/srep08173.

1234 Sim AD, Wheeler D. The venom gland transcriptome of the parasitoid wasp *Nasonia vitripennis*
1235 highlights the importance of novel genes in venom function. BMC Genomics 2016;17:571.
1236 https://doi.org/10.1186/s12864-016-2924-7.

1237 Sluss R. Behavioral and anatomical responses of the convergent lady beetle to parasitism by
1238 *Perilitus coccinellae* (Schrank) (Hymenoptera: Braconidae). J Invertebr Pathol 1968;10:9–27.
1239 https://doi.org/10.1016/0022-2011(68)90259-0.

1240 Soulages JL, Wells MA. Lipophorin: The Structure of an Insect Lipoprotein and Its Role in Lipid
1241 Transport in Insects. Adv Protein Chem 1994;45:371–415. https://doi.org/10.1016/S0065-
1242 3233(08)60644-0.

1243 Strand MR. Teratocytes and their functions in parasitoids. Curr Opin Insect Sci 2014;6:68–73.
1244 https://doi.org/10.1016/j.cois.2014.09.005.

1245 Strand MR. The physiological interactions of parasitoids with their hosts and their influence on
1246 reproductive strategies. In: Waage J, Greahead D, editors. Insect Parasitoids, London: Academic
1247 Press; 1986, p. 97–136.

1248 Strand MR, Burke GR. Polydnavirus-wasp associations: Evolution, genome organization, and
1249 function. Curr Opin Virol 2013;3:587–94. https://doi.org/10.1016/j.coviro.2013.06.004.

1250 Strand MR, Burke GR. Polydnaviruses as Symbionts and Gene Delivery Systems. PLoS Pathog
1251 2012;8:e1002757. https://doi.org/10.1371/journal.ppat.1002757.

1252 Strand MR, Meola SM, Vinson SB. Correlating pathological symptoms in *Heliothis virescens* eggs
1253 with development of the parasitoid *Telenomus heliothidis*. J Insect Physiol 1986;32:389–402.
1254 https://doi.org/10.1016/0022-1910(86)90052-1.

1255 Strand MR, Vinson SB, Nettles WC, Xie ZN. In vitro culture of the egg parasitoid *Telenomus*
1256 *heliothidis*: the role of teratocytes and medium consumption in development. Entomol Exp Appl
1257 1988;46:71–8. https://doi.org/10.1111/j.1570-7458.1988.tb02269.x.

1258 Strand MR, Wong EA. The growth and role of *Microplitis demolitor* teratocytes in parasitism of
1259 *Pseudoplusia includens*. J Insect Physiol 1991;37:503–15. https://doi.org/10.1016/0022-
1260 1910(91)90027-W.

1261 Suzuki M, Tanaka T. Development of *Meteorus pulchricornis* and regulation of its noctuid host,
1262 *Pseudaletia separata*. J Insect Physiol 2007;53:1072–8.
1263 https://doi.org/10.1016/j.jinsphys.2007.06.006.

1264 Teng Z-W, Xiong S-J, Xu G, Gan S-Y, Chen X, Stanley D, et al. Protein Discovery: Combined
1265 Transcriptomic and Proteomic Analyses of Venom from the Endoparasitoid *Cotesia chilonis*
1266 (Hymenoptera: Braconidae). Toxins (Basel) 2017;9:135. https://doi.org/10.3390/toxins9040135.

Thompson SN. The nutritional physiology of *Trichoplusia ni* parasitized by the insect parasite, *Hyposoter exiguae*, and the effects of parallel-feeding. Parasitology 1983;87:15–28. https://doi.org/10.1017/S0031182000052380.

Thompson SN. Immediate Effects of Parasitization by the Insect Parasite, *Hyposoter exiguae* on the Nutritional Physiology of Its Host, *Trichoplusia ni*. J Parasitol 1982a;68:936–41. https://doi.org/10.2307/3281009.

Thompson SN. Effects of the insect parasite, *Hyposoter exiguae* on the total body glycogen and lipid levels of its host, *Trichoplusia ni*. Comp Biochem Physiol B: Comp Biochem 1982b;72:233–7. https://doi.org/10.1016/0305-0491(82)90040-2.

Thompson SN. Lipid nutrition during larval development of the parasitic wasp, *Exeristes*. J Insect Physiol 1977;23:579–83. https://doi.org/10.1016/0022-1910(77)90051-8.

Thompson SN, Barlow JS. Regulation of Lipid Metabolism in the Insect Parasite, *Exeristes roborator* (Fabricius). J Parasitol 1976;62:303–6. https://doi.org/10.2307/3279292.

Thompson SN, Barlow JS. The Fatty Acid Composition of Parasitic Hymenoptera and its Possible Biological Significance. Ann Entomol Soc Am 1974;67:627–32. https://doi.org/10.1093/aesa/67.4.627.

Thompson SN, Barlow JS. Influence of Host Fatty Acid Composition on That of Ichneumonoid and Chalcidoid Wasps. J Parasitol 1972a;58:836–9. https://doi.org/10.2307/3278331.

Thompson SN, Barlow JS. Synthesis of fatty acids by the parasite *Exeristes comstockii* (Hymenop.) and two hosts, *Galleria mellonella* (Lep.) and *Lucilia sericata* (Dip.). Can J Zool 1972b;50:1105–10. https://doi.org/10.1139/z72-147.

Thompson SN, Barlow JS. The change in fatty acid composition pattern of *Itoplectis conquisitor* (Say) reared on different hosts. J Parasitol 1970;56:845–6.

Thompson SN, Redak RA. Parasitism of an insect *Manduca sexta* L. alters feeding behaviour and nutrient utilization to influence developmental success of a parasitoid. J Comp Physiol B 2008;178:515–27. https://doi.org/10.1007/s00360-007-0244-6.

Turunen S. Digestion and absorption of lipids in insects. Comp Biochem Physiol A Physiol 1979;63:455–60. https://doi.org/10.1016/0300-9629(79)90171-3.

Uçkan F, Ergin E, Rivers DB, Gençer N. Age and diet influence the composition of venom from the endoparasitic wasp *Pimpla turionellae* L. (Hymenoptera: Ichneumonidae). Arch Insect Biochem Physiol 2006;63:177–87. https://doi.org/10.1002/arch.20154.

Vincent B, Kaeslin M, Roth T, Heller M, Poulain J, Cousserans F, et al. The venom composition of the parasitic wasp *Chelonus inanitus* resolved by combined expressed sequence tags analysis and proteomic approach. BMC Genomics 2010;11:693. https://doi.org/10.1186/1471-2164-11-693.

Visser B, Alborn HT, Rondeaux S, Haillot M, Hance T, Rebar D, et al. Phenotypic plasticity explains apparent reverse evolution of fat synthesis in parasitic wasps. Sci Rep 2021;11:7751. https://doi.org/10.1038/s41598-021-86736-8.

Visser B, van Dooremalen C, Vázquez Ruiz A, Ellers J. Fatty acid composition remains stable across trophic levels in a gall wasp community. Physiol Entomol 2013;38:306–12. https://doi.org/10.1111/phen.12035.

Visser B, Ellers J. Lack of lipogenesis in parasitoids: A review of physiological mechanisms and evolutionary implications. J Insect Physiol 2008;54:1315–22. https://doi.org/10.1016/j.jinsphys.2008.07.014.

Visser B, Hance T, Noël C, Pels C, Kimura MT, Stökl J, et al. Variation in lipid synthesis, but genetic homogeneity, among *Leptopilina* parasitic wasp populations. Ecol Evol 2018;8:7355–64. https://doi.org/10.1002/ece3.4265.

Visser B, le Lann C, den Blanken FJ, Harvey JA, van Alphen JJM, Ellers J. Loss of lipid synthesis as an evolutionary consequence of a parasitic lifestyle. Proc Natl Acad Sci USA 2010;107:8677–82. https://doi.org/10.1073/pnas.1001744107.

Visser B, Le Lann C, Hahn DA, Lammers M, Nieberding CM, Alborn HT, et al. Many parasitoids lack adult fat accumulation, despite fatty acid synthesis: A discussion of concepts and considerations for future research. Curr Opin Insect Sci 2023;3:100055. https://doi.org/10.1016/j.cris.2023.100055.

Visser B, Roelofs D, Hahn DA, Teal PEA, Mariën J, Ellers J. Transcriptional changes associated with lack of lipid synthesis in parasitoids. Genome Biol Evol 2012;4:752–62. https://doi.org/10.1093/gbe/evs065.

Visser B, Willett DS, Harvey JA, Alborn HT. Concurrence in the ability for lipid synthesis between life stages in insects. R Soc Open Sci 2017;4:160815. https://doi.org/10.1098/rsos.160815.

Volkoff N, Colazza S. Growth patterns of teratocytes in the immature stages of *Trissolcus basalis* (Woll.) (Hymenoptera: Scelionidae), an egg parasitoid of *Nezara viridula* (L.) (Heteroptera: Pentatomidae). Int J Insect Morphol Embryol 1992;21:323–36. https://doi.org/10.1016/0020-7322(92)90027-K.

Wang J, Jin H, Schlenke T, Yang Y, Wang F, Yao H, et al. Lipidomics reveals how the endoparasitoid wasp *Pteromalus puparum* manipulates host energy stores for its young. Biochim Biophys Acta Mol Cell Biol Lipids 2020a;1865:158736. https://doi.org/10.1016/j.bbalip.2020.158736.

Wang J, Song J, Fang Q, Yao H, Wang F, Song Q, et al. Insight into the Functional Diversification of Lipases in the Endoparasitoid *Pteromalus puparum* (Hymenoptera: Pteromalidae) by Genome-scale Annotation and Expression Analysis. Insects 2020b;11:227. https://doi.org/10.3390/insects11040227.

Wang L, Zhu JY, Qian C, Fang Q, Ye GY. Venom of the parasitoid wasp *Pteromalus puparum* contains an odorant binding protein. Arch Insect Biochem Physiol 2015;88:101–10. https://doi.org/10.1002/arch.21206.

Wang Y, Wu X, Wang Z, Chen T, Zhou S, Chen J, et al. Symbiotic bracovirus of a parasite manipulates host lipid metabolism via tachykinin signaling. PLoS Pathog 2021;17:e1009365. https://doi.org/10.1371/journal.ppat.1009365.

Weers PMM, Ryan RO. Apolipophorin III: Role model apolipoprotein. Insect Biochem Mol Biol 2006;36:231–40. https://doi.org/10.1016/j.ibmb.2006.01.001.

Werren JH, Loehlin DW. The parasitoid wasp *Nasonia*: An emerging model system with haploid male genetics. Cold Spring Harb Protoc 2009;4:pdb.emo134. https://doi.org/10.1101/pdb.emo134.

Whitfield JB, Austin AD, Fernandez-Triana JL. Systematics, Biology, and Evolution of Microgastrine Parasitoid Wasps. Annu Rev Entomol 2017;63:389–406. https://doi.org/10.1146/annurev-ento-020117-043405.

Xu J, Saunders CW, Hu P, Grant RA, Boekhout T, Kuramae EE, et al. Dandruff-associated *Malassezia* genomes reveal convergent and divergent virulence traits shared with plant and human fungal pathogens. Proc Natl Acad Sci USA 2007;104:18730–5. https://doi.org/10.1073/pnas.0706756104.

Xueke G, Shuai Z, Junyu L, Limin L, Lijuan Z, Jinjie C. Lipidomics and RNA-Seq Study of Lipid Regulation in Aphis gossypii parasitized by *Lysiphlebia japonica*. Sci Rep 2017;7:1364. https://doi.org/10.1038/s41598-017-01546-1.

Yan Z, Fang Q, Wang L, Liu J, Zhu Y, Wang F, et al. Insights into the venom composition and evolution of an endoparasitoid wasp by combining proteomic and transcriptomic analyses. Sci Rep 2016;6:19604. https://doi.org/10.1038/srep19604.

Yang H, Zhang R, Zhang Y, Liu Q, Li Y, Gong J, et al. Cathepsin-L is involved in degradation of fat body and programmed cell death in *Bombyx mori*. Gene 2020;760:144998. https://doi.org/10.1016/j.gene.2020.144998.

Zhang D, Dahlman DL, Järlfors UE. Effects of *Microplitis croceipes* Teratocytes on Host Haemolymph Protein Content and Fat Body Proliferation. J Insect Physiol 1997;43:577–85. https://doi.org/10.1016/S0022-1910(96)00118-7.

Zhang D, Dahlman DL, Järlfors UE, Southgate HH, Wiley SP. Ultrastructure of Microplitis croceipes (Cresson) (Braconidae : Hymenoptera) teratocytes. Int J Insect Morphol Embryol 1994;23:173–87. https://doi.org/10.1016/0020-7322(94)90016-7.

Zhang S, Luo J-Y, Lv L-M, Wang C-Y, Li C-H, Zhu X-Z, et al. Effects of *Lysiphlebia japonica* (Ashmead) on cotton-melon aphid *Aphis gossypii* Glover lipid synthesis. Insect Mol Biol 2015;24:348–57. https://doi.org/10.1111/imb.12162.

Zhu J-Y, Yin Ye G, Fang Q, Hu C. Alkaline Phosphatase from Venom of the Endoparasitoid Wasp, *Pteromalus puparum*. J Insect Sci 2010;10:14. https://doi.org/10.1673/031.010.1401.

Zhuo Z-H, Yang W, Xu D-P, Yang C-P, Yang H. Effects of *Scleroderma sichuanensis* Xiao (Hymenoptera: Bethylidae) venom and parasitism on nutritional content regulation in host *Tenebrio molitor* L. (Coleoptera: Tenebrionidae). Springerplus 2016;5:1017. https://doi.org/10.1186/s40064-016-2732-1.