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Abstract	Fruit flies of the genus *Drosophila* have become an important model for energy storage and metabolism at multiple levels of organization. *Drosophila* species differ substantially in their abilities to survive without food, and many species exhibit latitudinal clines in energy storage and starvation resistance. Variation in starvation resistance can also be generated using experimental evolution, by subjecting populations to starvation selection. Physiological analyses of starvation-selected flies reveal that the entire life history of the animal is affected, particularly larval traits associated with growth and energy storage. As adults, these animals contain large lipid stores, but at the cost of reduced fecundity. The genetic toolkit available for *Drosophila melanogaster* has also allowed researchers to identify the molecular basis for how energy is stored and distributed to tissues that need it. Insulin signaling and other pathways can be manipulated in tissue- and temporal-specific ways that are revealing fundamental energy regulatory mechanisms common to all animals.

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Chapter 4
Drosophila as a Model for Starvation: Evolution, Physiology, and Genetics

Allen G. Gibbs and Lauren A. Reynolds

4.1 Introduction

Drosophila melanogaster is one of the primary genetic models for understanding how nutritional limitation affects cellular physiology, because many of the molecular and cellular signaling pathways are shared among invertebrates and vertebrates. To a lesser extent, it is a model for organismal responses, although differences in endocrine systems sometimes make the link to vertebrates one of analogy rather than homology. *Drosophila* is also an excellent model for the evolution of starvation responses. The evolutionary history of the genus has been well studied, and *D. melanogaster's* short generation time and ease of maintenance have allowed experimental evolution studies on starvation resistance. We review here studies of starvation in *Drosophila* at multiple levels of organization, from species to molecules. A great advantage of *Drosophila* is the ability to traverse these levels relatively easily, and information across all levels is now being integrated in many labs around the world.

It is important to recognize at the outset that *D. melanogaster* is only a *model* for other species, including other *Drosophila* species. We were charged with reviewing the physiology of starvation specifically in *Drosophila*, and so we do not refer the large and interesting body of related work done with *Manduca, Locusta, Bombyx*, and a wide variety of other insects. The literature on *Drosophila* alone is extensive—our recent Web of Science search for "drosophila and feeding" returned nearly 2000 citations. This review will therefore necessarily skim the surface and omit a great deal of interesting information about starvation in *Drosophila*.

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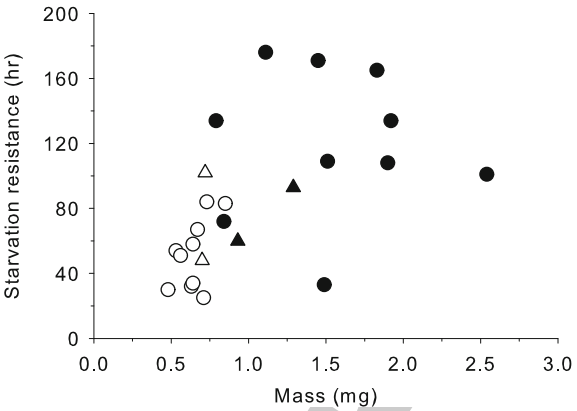

Fig. 4.1 Starvation resistance of 22 species of *Drosophila*. Male flies were assayed at 25°C. *Open circles*, tropical species; *filled circles*, temperate species. *Triangles* indicate tropical and temperate populations of *D. melanogaster* and *D. simulans*. Data modified from van Herrewege and David (1997)

4.2 Starvation Resistance in Natural Populations

The role of starvation stress in the ecology of *Drosophila* species is very poorly understood; in fact, the ecology of *Drosophila* in general is poorly understood. It is clear, however, that *Drosophila* species vary greatly in their ability to survive starvation stress. van Herrewege and David (1997) found that *Drosophila* species differed up to 5-fold in their survival in humid air. Starvation resistance was highly temperature dependent, with flies surviving approximately twice as long at 17°C as at 25°C. Species from temperate regions tended to survive longer than tropical species. The temperate species studied also tended to be larger, which may have contributed to longer survival times (Fig. 4.1). On the other hand, flies from temperate populations of two species were larger than tropical congeners, but size had little effect on starvation resistance.

Many *Drosophila* species have broad geographic ranges, allowing intraspecific studies of local adaptation in starvation resistance. The Indian subcontinent has been particularly well studied. Northern populations of several species have lower starvation resistance compared to southern, subtropical populations (Parkash et al. 1994; Parkash and Munjal 2000; Sisodia and Singh 2010). Starvation resistance also increases with latitude in Australian populations of *D. birchii* (Griffiths et al. 2005).

In eastern North America, an opposing latitudinal cline occurs. Populations of *D. melanogaster* in the north are more starvation resistant than southern populations (Schmidt et al. 2005; Schmidt and Paaby 2008). Robinson et al. (2000) also found no correlation between latitude and starvation resistance in *D. melanogaster* from South America. In Australia, differences in starvation resistance between populations of *D. melanogaster* were found, but these were not correlated with environmental conditions (Hoffmann et al. 2001, 2005; Hoffmann and Weeks 2007), whereas Philippine *Drosophila* species varied within, but not among, populations (van der Linde and Sevenster 2006).

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The explanation(s) for differing geographic patterns in starvation resistance are not clear. Parkash and Munjal (2000) argue that tropical populations are more susceptible to starvation because of higher metabolic rates related to high habitat temperatures. In North America, northern populations of *D. melanogaster* are more likely to undergo reproductive diapause under simulated winter conditions (Schmidt et al. 2005). Schmidt and Paaby (2008) concluded that females able to use reproductive diapause to overwinter are more resistant to stress in general, including starvation. Australian populations also differ in reproductive patterns in the winter (Mitrovski and Hoffmann 2001; Hoffmann et al. 2003), suggesting a potential link between reproduction and stress resistance.

It should also be noted that the latitudinal ranges for these studies differ. For example, the northernmost Indian populations studied were from similar latitudes to the southernmost North American populations. Differing types of selection at the extreme latitudes could result in higher starvation resistance in both regions. For example, global scale atmospheric circulation patterns (Hadley cells) create generally lower humidity approximately 30° north and south of the equator. Natural selection for surviving desiccation could tradeoff against starvation resistance (Parkash et al. 1994; Parkash and Munjal 2000; Parkash et al. 2012).

An alternative to comparative studies of starvation resistance is to study its evolution in the laboratory. *Drosophila melanogaster* is a widely used experimental model for the evolution of stress resistance (Garland and Rose 2009). The use of replicated populations (and unselected control populations) under controlled conditions allows correlations and tradeoffs between traits to be assessed and tested in a rigorous manner, although laboratory environments are not necessarily as simple as they appear (Gibbs and Gefen 2009). Starvation resistance evolves rapidly when populations are subjected to strong selection each generation (Rose et al. 1992). Selection on a poor diet (lemons) also results in increased starvation resistance (Harshman et al. 1999). Most studies have involved selection for adult starvation resistance, but at least one study on larval selection has been performed (Kolss et al. 2009).

4.3 Physiological Mechanisms of Starvation Resistance

At the organismal level, there are three mechanisms by which starvation resistance can be increased, as illustrated in Fig. 4.2. Animals can store more energy (lipids, carbohydrates, protein), they can consume it at a slower rate, or they can tolerate loss of a greater fraction of their initial energy supply. These mechanisms are not mutually exclusive. A fourth, behavioral strategy is cannibalism. When flies are starved in groups, in principle the longest survivors can consume those that have already died. This behavior is not seen in wildtype flies (Huey et al. 2004), but could evolve in starvation-selected populations.

Starvation resistance is positively correlated with lipid content among different *Drosophila* species (van Herrewege and David 1997; Bharathi et al. 2003). In fact,

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Fig. 4.2 Potential organismal mechanisms to increase starvation resistance. **a** Increased energy storage. **b** Reduced energy consumption. **c** Lower energetic threshold for mortality

the differences between tropical and temperate species seen in Fig. 4.1 are largely due to higher relative lipid content. Similar correlations between lipid content and starvation resistance occur within species (Parkash et al. 2005; Ballard et al. 2008; Sisodia and Singh 2010), although Jumbo-Lucioni et al. (2010) found that these traits were not genetically correlated in a set of 40 inbred lines. Greatly increased lipid storage is a consistent finding in starvation selection experiments (Chippindale et al. 1996; Djawdan et al. 1997; Harshman et al. 1999; Schwasinger-Schmidt et al. 2012). Lipid contents are generally much higher than in natural populations, suggesting that lipid storage has an evolutionary cost. Carbohydrates have received far less attention than lipids as energy stores, but also increase under starvation selection (Djawdan et al. 1997). Thus, energy storage, particularly in the form of lipids, is a consistent marker for starvation resistance.

The relationship between metabolic rates and starvation resistance is murkier. Surprisingly, no systematic comparative studies of metabolic rates in natural populations of *Drosophila* appear to have been done, at least not in the context of starvation stress. Metabolic rates differ substantially among species (Gibbs et al. 2003; Marron et al. 2003). Some of this variation may be related to water conservation, as desert (cactophilic) *Drosophila* have lower metabolic rates than other species after correction for body size and phylogenetic relationships (Gibbs et al. 2003). Tolerance of low energy content has not been studied (Rion and Kawecki 2007).

In starvation selection experiments, the evidence for evolution of reduced metabolism is mixed. Starvation-selected flies often have lower mass-specific metabolic rates than controls (Djawdan et al. 1997; Harshman et al. 1999). However, they are also larger because of their greater energy stores; when this is taken into consideration metabolic differences may disappear (Djawdan et al. 1997). Baldal et al. (2006) found that starvation-selected females actually tended to have higher metabolic rates than controls in the absence of food. No differences were seen when food was present, but metabolic rates are consistently lower when flies are starved than when they are fed (Djawdan et al. 1997; Baldal et al. 2006).

Harshman and Schmid (1998) also found no relationship between metabolic rates and starvation resistance. More recently, Schwasinger-Schmidt et al. (2012) found some support for the idea that starvation-selected flies are less active, and therefore should have lower metabolic rates (see also Hervant, Chap. 7). In summary, lower metabolic rates may contribute to increased starvation resistance in *Drosophila*, but their contribution is inconsistent and is certainly less significant than differences in energy storage.

4.4 Starvation and Life History Traits

A fundamental tradeoff in life history evolution exists between allocation of resources to survival and reproduction (see also Kirk, Chap. 3). This tradeoff can be alleviated by acquiring more resources (de Jong 1993), as exemplified by lipid accumulation in starvation-selected populations of *Drosophila*. Resource acquisition may have its own costs, however. Starvation-selected flies take longer to develop (Chippindale et al. 1996; Harshman et al. 1999) and have lower fecundity than controls (Wayne et al. 2006; Kolss et al. 2009). This is despite their larger body size and higher lipid content, factors that are generally correlated with higher fecundity in insects.

This conundrum may be explained by the complex life cycle of *Drosophila*. Holometabolous insects have striking differences in life history from vertebrates. In the case of *D. melanogaster*, eggs hatch into a larva that is essentially a feeding and growth machine. Over 3 days, the larva increases in mass by approximately 200-fold (Church and Robertson 1966). Soon thereafter it enters a 15–24 h wandering phase, during which it ceases feeding, leaves the media, and searches for a pupation site. The larva selects a spot, secretes a glue protein that adheres the animal to the substrate, and undergoes metamorphosis. Approximately 4 days later, an adult fly emerges from the pupal case. The adult feeds and allocates resources between somatic maintenance and reproduction. Thus, the life history of *Drosophila* can be broadly separated into 3 nutritional states: a feeding and growth stage, a non-feeding period lasting from late larval through early adult development, and a feeding but non-growing adult stage.

Drosophila pupae consume less than half of their stored lipids during metamorphosis, so flies eclose to adulthood with an energetic reserve (Merkey et al. 2011). Starvation-selected adults eclose with greater lipid stores than unselected controls, so that differences in energy storage occur before adulthood as well as in the young adult (Chippindale et al. 1996). This may be achieved by higher larval feeding rates to grow faster, extending the larval feeding period, reduced energy expenditure during metamorphosis, or some combination of these. Pre-adult stages of starvation-selected lines have not been well characterized, but selected lines do have longer egg-to-adult development times, suggesting a longer feeding period (Chippindale et al. 1996). Within these populations, individuals with longer development times also survived starvation longer.

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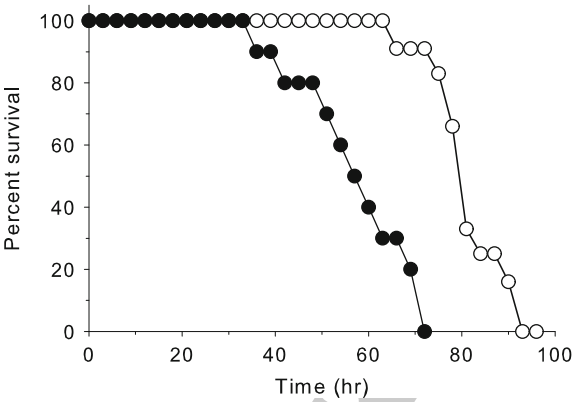

Fig. 4.3 Inhibition of programmed fat cell death increases starvation resistance in *D. melanogaster*. *Filled symbols*, control flies; *open symbols*, flies in which fat cell death was inhibited by expression of *diap* (*Drosophila* inhibitor of apoptosis) in the larval fat body. Data modified from Aguila et al. (2007)

Larvae store energy in the larval fat body. The fat body is unique to insects and serves many functions in addition to energy storage, including but not limited to immune responses, detoxification, and endocrine secretion (Hoshizaki 2005). In comparison to other larval tissues, larval fat body is unusual in that its cells remain intact during metamorphosis and are present in the young adult (Nelliot et al. 2006). Most larval tissues undergo programmed cell death in the pupa, with their contents being used to support proliferation of the imaginal disk cells that will form the adult tissues. Larval fat cells escape this fate, then undergo programmed cell death in the first 48 h of adult life (Aguila et al. 2007). Nutrients released at this time are used to support adult tissues and reproduction (Min et al. 2006; O'Brien et al. 2008).

Recent evidence suggests that the larval fat body has an important role in starvation resistance in young adult flies. Aguila et al. (2007) observed that newly eclosed female adults survived starvation stress over twice as long as 3–10-day-old females. The authors then used a genetic manipulation to delay death of the larval fat cells by approximately 2 days. These females survived starvation ~24 h longer than unmanipulated flies (Fig. 4.3). These flies also had lower fecundity, suggesting that larval resources are also important for reproduction (Aguila, Hoshizaki and Gibbs, unpublished observations).

Together, these findings suggest that starvation selection affects the physiology of the larval fat body. Increased lipid storage during the larval stage is certainly consistent with this idea. Because all cell division in this tissue occurs embryonically (Hoshizaki 2005), this probably reflects more lipid per cell rather than more fat cells. Starvation-selected females also have lower early adult fecundity than controls, despite having more ovarioles (Wayne et al. 2006). Preliminary evidence suggests that fat cell death is delayed in starvation-selected populations (Reynolds and Gibbs, unpublished data), which would cause lower fecundity. The onset of the wandering stage and developmental events in the fat body are regulated by the steroid hormone, 20-hydroxyecdysone (20E; Riddiford and Truman 1993; Rusten et al. 2004; Hoshizaki 2005; Bond et al. 2011). The hormonal basis for fat body changes in all stages of starvation-selected flies is unknown, but 20E signaling is likely to be involved.

4.5 Metabolic Responses to Starvation Stress

Drosophila melanogaster is a widely studied model for starvation responses, but the vast majority of studies have used the third and last larval instar. In adults, food deprivation causes increased activity (Connolly 1966; Knoppien et al. 2000; Farhadian et al. 2012). Increased energy consumption would appear counterintuitive, but in nature waiting for the next rotting banana to appear makes no sense (see also McCue et al., Chap. 8). Laboratory-selected flies do not have the option of finding a new food source, so they reduce their activity when food is absent (Williams et al. 2004). When food is returned, flies increase their feeding rate and allow more food to accumulate in their crop relative to unstarved controls (Farhadian et al. 2012).

The primary fuel consumed during starvation stress is lipid (Marron et al. 2003), by mechanisms closely resembling, and sometimes homologous to, mammalian regulation of lipolysis (Arrese and Soulages 2010). Neurosecretory cells in the ring gland secrete adipokinetic hormone (AKH), which activates lipolysis via G protein-mediated phosphorylation of one of the primary proteins associated with lipid droplets in the fat body, lipid storage droplet protein-1 (LSD1), a member of the perilipin protein family. As starvation progresses, transcription of *brummer* (*bmm*) is activated (Groenke et al. 2007). Brummer is the *Drosophila* homolog of adipose triglyceride lipase (Groenke et al. 2005). Lipids are transported in the hemolymph bound to lipophorins, probably in the form of diacylglycerides, rather than triacylglycerides (Canavoso et al. 2001). Oenocytes, specialized cells attached to the inner surface of the animal, take up some of these lipids and store them in a manner analogous to mammalian hepatocytes (Gutierrez et al. 2007). Most lipids, however, presumably are absorbed and metabolized by cells throughout the body.

In addition to AKH signaling, the insulin signaling pathway regulates nutrient uptake, storage, and metabolism. This pathway is well conserved between flies and mammals, making *Drosophila* an excellent model for mammals (Fig. 4.4). *Drosophila melanogaster* has 7 insulin-like peptides (dILPs) that are homologous to the insulin family in vertebrates, as well as a homologous insulin receptor. The dILPs are expressed at different times by different tissues, but there are some overlapping functions. The most important in terms of nutritional status are dILPs expressed by 7 neurosecretory cells (NSCs) in the brain. Ablation of these cells in larvae or adults results in elevated hemolymph trehalose and excess lipid accumulation, analogous to the condition in diabetic mammals (Belgacem and Martin 2006). However, release of dILPs is not dependent on lipid or carbohydrate levels; instead it depends on an amino acid sensing mechanism in the fat body (Geminard et al. 2009).

Drosophila have only one insulin receptor (InR), which can bind all 7 dILPs. Binding activates an intracellular signaling pathway strongly resembling, but less redundant than, mammalian insulin signaling (Teleman 2010). Events include activation of PI3 kinase (PI3 K), followed by the protein kinase Akt. Akt then

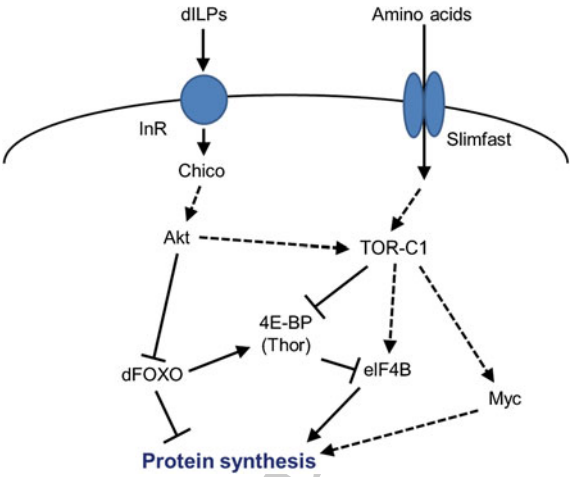

Fig. 4.4 Insulin/TOR signaling in *Drosophila*. Only members of these pathways mentioned in the text are shown. *Arrows* indicate activation of the downstream component; *blocked lines* indicate inhibition. *Dashed lines* indicate an indirect effect mediated by one or more intermediate steps. A more complete diagram can be found in Teleman (2010)

phosphorylates a variety of proteins, including dFOXO, the single *Drosophila* member of the FOXO family of transcription factors. dFOXO regulates transcription of numerous targets (Teleman et al. 2008), including 4E-binding protein (4E-BP, or Thor, a general inhibitor of translation). Phosphorylation of dFOXO decreases *Thor* expression, allowing greater protein synthesis.

Akt also indirectly regulates TOR (Target of Rapamycin), a central regulator of cellular metabolism. The TOR-C1 form of TOR increases ribosomal synthesis, inhibits translational repression by phosphorylating *Thor*, and stimulates amino acid uptake via the amino acid transporter, Slimfast. There is extensive crosstalk and feedback among various branches of the insulin signaling pathway. Accumulation of amino acids activates TOR, thereby activating amino acid transport. dFOXO regulates the expression of *myc*, a target of TOR that stimulates ribosome synthesis (Teleman et al. 2008). dFOXO and TOR pathways also intersect via their opposing effects on the expression and activity of 4E-BP.

The alphabet-soup description above includes only a few components of the insulin/TOR signaling pathway, but it provides a framework for understanding how starvation affects signaling. During starvation in *Drosophila*, secretion of dILPs by the neurosecretory cells decreases. Food-seeking behavior increases, mediated by neural S6 kinase, a downstream target of insulin signaling. AKH secretion also stimulates activity (Lee and Park 2004; Isabel et al. 2005). Phosphatidylinositol-(3,4,5)-triphosphate levels decline, Akt becomes dephosphorylated, and dFOXO is recruited to the nucleus. *Thor* expression increases, and existing Thor protein becomes dephosphorylated and can inhibit elongation initiation factor eIF4B, thereby inhibiting protein synthesis. dFOXO and TOR inputs inhibit *myc* transcription, thereby inhibiting ribosome biogenesis. The overall result is a general reduction in energy-intensive biosynthetic activities. In addition TOR-mediated autophagy of fat cell contents commences, generating nutrients that

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can be used to support metabolism in the rest of the body (Scott et al. 2004; McPhee and Baehrecke 2009).

This general pattern is likely to differ in a tissue-specific manner. It can also vary depending upon developmental stage. The pupa does not feed, yet needs to devote a significant fraction of metabolism to building adult tissues. Beginning in the wandering stage of the third instar, 20E signaling induces the larval fat body to express d*ILP6* (Slaidina et al. 2009) and activates lipid catabolism (Wang et al. 2010). Inhibition of d*ILP6* transcription in the fat body results in smaller adults, but these have high triglyceride levels and are more starvation resistant than control flies. Additional experiments revealed that d*ILP6* expression is regulated by dFOXO, providing a further example of the intersection between these pathways. In another example of signaling crosstalk, recent work suggests that dFOXO regulates expression of *dDOR*, a coactivator of the ecdysone receptor (Francis et al. 2010).

Mammalian researchers will note that we have barely mentioned sugar homeostasis in our discussion of insulin signaling (see Champagne et al., Chap. 19). To some extent this is due to the focus on the *Drosophila* larva, a very rapidly growing stage that requires high levels of amino acids to support biosynthesis. In fact, a common control treatment for 'starvation' (lack of amino acids) is a diet containing sucrose to allow animals to continue to manufacture ATP.

In *Drosophila*, the primary signal for insulin secretion is the presence of amino acids, not carbohydrates. The primary site for sensing overall nutritional status is the fat body (Colombani et al. 2003). One or more factors secreted by the fat body stimulates dILP secretion by the NSCs when amino acids are abundant (Geminard et al. 2009). When amino acid levels are low or the Slimfast amino acid transporter is inactivated, dDILP secretion is reduced. Thus, the NSCs and fat body are in reciprocal communication with each other. The identity of the signal released by the fat body is unknown, but the fat body is known to produce numerous growth factors (Britton and Edgar 1998; Kawamura et al. 1999).

Under prolonged starvation, an additional energy source available to female flies is reabsorbed eggs (Wilson 1985; McCall 2004). Oogenesis is initiated from germline stem cells situated at the anterior tip of each ovariole, the germarium. An egg chamber or follicle forms, comprising the oocyte and nurse cells enclosed in a layer of follicle cells (Wu et al. 2008). In well-fed laboratory strains of *D. melanogaster*, new egg chambers are formed continuously over most of an adult female's life span. Reabsorption during starvation is initiated by apoptosis of the nurse cells (Terashima and Bownes 2005, 2006), and there is increased cell death in the germarium (Drummond-Barbosa and Spradling 2001; Pritchett et al. 2009). One might predict that starvation-selected flies would contain fewer ovarioles than control flies, but this is not the case (Wayne et al. 2006). Reduced fecundity in these populations may instead be caused by lower activity of the germline stem cells or increased egg reabsorption, but this has not been investigated.

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4.6 Genomics of Starvation Resistance

As the first multicellular eukaryote with a sequenced genome, *D. melanogaster* has been the subject of numerous genomic analyses, including several related to starvation stress. Harbison et al. (2004) identified nearly 400 genes associated with starvation resistance, many of them associated with cell fate determination. The authors suggest that these genes may affect resource allocation during development, setting the conditions for survival later. This pattern is consistent with selection experiments in which larval resource acquisition is a major determinant of adult starvation resistance (Chippindale et al. 1996). Analyses of quantitative trait loci (QTLs) have identified several genomic regions associated with differences in starvation resistance and energy storage (Vieira et al. 2000; Harbison et al. 2005; Wang et al. 2005).

Microarray experiments have shown that up to 25% of the transcriptome can be affected by starvation (Harbison et al. 2005). The first such transcriptome analysis was performed by Zinke et al. (2002). The focus of this study was sugar-related gene expression, so larvae fed sugar were compared with starved larvae and those fed with sugar and protein. Several genes associated with lipid catabolism were upregulated specifically in starved larvae, whereas lipid synthetic genes were upregulated in larvae fed only sugar. These results are consistent with the idea that starved larvae rely on endogenous lipid to survive, while sugar-fed larvae use this resource to make ATP, with any excess going to lipid synthesis. Surprisingly, Harbison et al. (2005) found that genes for biosynthetic proteins tended to increase in expression in starved flies. Transcriptional networks affecting energy storage and metabolism have also been identified (Jumbo-Lucioni et al. 2010). Transcripts correlated with lipid content included several that have human homologs and have been associated with obesity in mice.

The studies above assayed whole-body gene transcription, but different tissues will respond differently to starvation (e.g. fat body and oenocytes). Immune function genes are downregulated in several tissues (Farhadian et al. 2012). In ovaries, changes in expression of multiple members of the insulin/TOR signaling are consistent with an inhibition of protein synthesis and cell growth (Terashima and Bownes 2005). Decreased expression of ovary-specific genes, such as yolk proteins, can also be detected in whole-animal experiments (Bauer et al. 2006). Starvation selection also affects gene expression. Sorensen et al. (2007) found that over 200 genes were constitutively downregulated in starvation-selected lines, including many involved in transcription and glycolysis, suggesting that overall metabolism may be lower. Interestingly, the specific genes identified differed from those differentially expressed during starvation stress (Harbison et al. 2005). Thus, acute and evolutionary responses to starvation appear to rely on different mechanisms.

Genomic studies of starvation in natural populations of *Drosophila* have also been performed. In both North America and Australia, latitudinal clines in allele

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frequency of the insulin receptor have been observed in *D. melanogaster* (Paaby et al. 2010). In North America, this cline parallels a cline in starvation resistance (Schmidt et al. 2005; Schmidt and Paaby 2008). No latitudinal clines were detected, however, for the InR substrate, Chico. This finding is consistent with genomic comparisons among *Drosophila* species, which show that evolution of downstream members of the insulin signaling pathway tends to be more constrained than that of upstream proteins (Alvarez-Ponce et al. 2009, 2012).

4.7 Summary

More is known about starvation responses in *Drosophila* than in any other insect, perhaps any other animal. The genetic resources available for *D. melanogaster* have made it a widely used model to study regulation of energy storage and mobilization. For example, many aspects of TOR signaling were initially identified in *Drosophila*, then studied in mammalian systems (Martin and Hall 2005). Genetic advantages notwithstanding, fruitflies are too small for convenient study of some aspects of starvation. For this reason, hemolymph transport of lipids is far better understood in larger insects such as *Manduca* (Arrese et al. 2001). Presumably *Drosophila* also convert triacylglycerides to diacylglycerides before releasing them into the hemolymph, but this has not been well studied. Life history differences among species will also affect how insects respond to starvation. Adult *Bombyx* moths do not feed, so starvation-induced reabsorption of eggs does not make sense and presumably does not occur. *Drosophila* is an excellent *model*, but comparative studies of insect starvation are still needed.

Comparative studies within the genus *Drosophila* should be very informative. *Drosophila* use a wide variety of host plants in nature, differing greatly in their spatial and temporal availability, as well as nutritional content (Markow and O'Grady 2008). Starvation resistance varies widely across the genus. Within species, local populations exhibit variation that in many cases suggests local adaptation to environmental conditions. At the time of this writing, genome sequences are available for 19 species of *Drosophila*, from many different nutritional habitats. A century of genetic research on *D. melanogaster*, intensive study of evolution in the genus *Drosophila*, and rapidly expanding genomic resources for *D. melanogaster* and its relatives provide many opportunities to deepen our understanding of starvation biology in insects and other animals.

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