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# **Drosophila melanogaster: A Powerful Tiny Animal Model for the Study of Metabolic Hepatic Diseases**

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1 ***Drosophila melanogaster*: a powerful tiny animal model for the study of**  
2 **metabolic hepatic diseases**

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18  
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20  
21 **Running title: *Drosophila*, a model for NAFLD**

22

23

24 **Abstract**

25 Animal experimentation is limited by unethical procedures, time-consuming protocols, and high cost.  
26 Thus, the development of innovative approaches for disease treatment based on alternative models in  
27 a fast, safe, and economic manner is an important, yet challenging goal. In this paradigm, the fruit-  
28 fly *Drosophila melanogaster* has become a powerful model for biomedical research, considering its  
29 short life cycle and low-cost maintenance. In addition, biological processes are conserved and  
30 homologs of ~75% of human disease-related genes are found in the fruit-fly. Therefore, this model  
31 has been used in innovative approaches to evaluate and validate the functional activities of candidate  
32 molecules identified in *in vitro* large-scale analyses, as putative agents to treat or reverse pathological  
33 conditions. In this context, *Drosophila* offers a powerful alternative to investigate the molecular  
34 aspects of liver diseases, since no effective therapies are available for those pathologies.  
35 Nonalcoholic fatty liver disease is the most common form of chronic hepatic dysfunctions, which  
36 may progress to the development of chronic hepatitis and ultimately to cirrhosis, thereby increasing  
37 the risk for hepatocellular carcinoma (HCC). This deleterious situation reinforces the use of the  
38 *Drosophila* model to accelerate functional research aimed at deciphering the mechanisms that sustain  
39 the disease. In this short review, we illustrate the relevance of using the fruit-fly to address aspects of  
40 liver pathologies to contribute to the biomedical area.

41

42

43 **Introduction**

44 In modern societies, obesity has become a prevalent problem; according to the World Health  
45 Organization (WHO), more than 600 million adults were obese in 2016 (WGO, 2020). Consequently,  
46 metabolic dysfunctions have considerably increased, including hepatic diseases, which are one of the  
47 top 10 causes of death worldwide (Asrani et al., 2019; Powell et al., 2021; WHO, 2020). These  
48 diseases classified as acute or chronic may progress to irreversible damage and malfunction of the  
49 liver, while therapeutic strategies are still limited. To address this problem, innovative drugs and  
50 natural products have been explored; however, clinical trials have their own limitations, considering  
51 patient safety against the adverse side effects of pharmaceutical products, extensive time required for  
52 analyses, and ethical issues. Thus, to help investigating new compounds in preclinical stages, the  
53 *Drosophila melanogaster* model has become of utmost importance. *Drosophila* genetics have been  
54 used for studying many biological processes and is now a powerful alternative model to investigate  
55 metabolic changes associated with disease settings. The fruit-fly has a short life cycle, which enables  
56 analyses of the effects of drugs in various metabolic routes in a short time (Lee et al., 2020; Men et  
57 al., 2016; Tennessen et al., 2014). Furthermore, relevant biological processes and disease-related  
58 genes are conserved between human and *Drosophila* (Pandey and Nichols, 2011; Ugur et al., 2016),  
59 despite the flies lower genomic complexity compared to that of humans. In this minireview we  
60 discuss the advantages offered by the fruit-fly model to investigate hepatic dysfunctions in a  
61 translational perspective.

62

63 **The burden of hepatic diseases needs translational science**

64 The liver is a critical organ for the maintenance of body homeostasis; it controls several molecular  
65 and metabolic routes and also acts in detoxification processes (Ramos et al., 2021). In metabolic  
66 disorders, lipids may accumulate in the hepatocytes contributing to the establishment of fatty liver,  
67 also known as hepatic steatosis. This clinical condition, which happens when triacylglycerols (TAGs)  
68 represent at least 5% of the liver weight (Ikura, 2014; Powell et al., 2021; Ramos et al., 2021), can  
69 remain asymptomatic for several years or progress to more deleterious stages. Nonalcoholic fatty  
70 liver disease (NAFLD) is one of the most common forms of liver pathologies affecting ~25% of the  
71 world population (Younossi et al., 2018). In addition, drug abuse, environmental contaminants,  
72 imbalanced diet, and viral infections may also contribute to the development of NAFLDs (Byrne and  
73 Targher, 2015). More dramatically, NAFLD favors the development of chronic hepatitis that

## ***Drosophila melanogaster* in translational studies of liver diseases**

74 potentially progresses to cirrhosis, increasing the risk for HCC (Friedman et al., 2018; Powell et al.,  
75 2021). The worldwide panel of liver pathologies also comprises chronic viral hepatitis, alcoholic  
76 liver disease, metabolic and cholestatic liver dysfunctions (Asrani et al., 2019; WGO, 2020). Although  
77 NAFLD was identified more than 50 years ago (Parise, 2019; Sanyal, 2019), the therapeutic options  
78 to treat or at least to control these diseases rely on long-term procedures and are still limited to  
79 weight loss and diet modification, stressing the critical needs of innovative strategies based on fast  
80 and reliable preclinical tests.

81 Translational science aims at using the scientific discoveries from preclinical models to set up the  
82 bases of clinical trials (Gilliland et al., 2016; van Erk et al., 2021). Knowledge on liver pathologies  
83 has now increased, leading to the characterization of the biochemical mechanisms underlying disease  
84 progression and to the identification of specific biomarkers (Eslam and George, 2020; Friedman et  
85 al., 2018; Samuel and Shulman, 2018). Advances in our understanding of lipid and sugar metabolism  
86 along with omics studies and mechanistic investigations, have revealed important regulatory  
87 functions of the transcriptions factors sterol-regulatory-element-binding-proteins (SREBPs),  
88 carbohydrate-responsive-element-binding-protein (ChREBP), liver-X-receptors (LXRs) and  
89 peroxisome-proliferator-activated-receptors (PPARs). In addition, the patatin-like-phospholipase-  
90 domain-containing-3 (PNPLA3), a triacylglycerol-lipase, has been shown to contribute to the control  
91 of energy expenditure or storage; PNPLA3 expression is regulated by nutritional sources, especially  
92 carbohydrates (Bruschi et al., 2017). Furthermore, the mechanistic-target-of-rapamycin (mTOR),  
93 which coordinates cell growth at the organismal level, is implicated in metabolic-related disease (Ji et  
94 al., 2021; Saxton and Sabatini, 2017). Other studies have demonstrated that epigenetic processes  
95 have been linked to liver diseases, but also that the genetic profile of an individual is directly  
96 correlated to the severity of the disease, since different alleles encode metabolic enzymes with  
97 variable functional activities (Anstee et al., 2019; Ramos et al., 2021). This has opened an unexplored  
98 field in the search for innovative therapeutics, including oligonucleotide usage to reduce through  
99 RNA-interference (RNAi) the levels of molecules relevant for liver homeostasis. In this context,  
100 translational research provides a safe way to test innovative drugs at a large scale using animal  
101 models. Rodents, have been long used in preclinical trials, despite the time-consuming analyses,  
102 while unpredictable side-effects of the drugs in humans cannot be excluded (Bryda, 2013; Van  
103 Norman, 2019). Thus, alternative models provide a golden solution, considering that many societies  
104 limit animal experimentation to protect them against cruelty (Doke and Dhawale, 2015; Freires et al.,  
105 2017).

106

107 **The fruit-fly model**

108 In this paradigm, the fruit-fly has emerged as a powerful system to study pathological conditions.  
109 The *Drosophila* life-cycle comprises four developmental stages: embryonic, larval, pupal and adult.  
110 Development from embryo to adult takes about ten days and adult lifespan one to two months  
111 (Johnson and Stolzing, 2019), allowing metabolic investigations on hundreds of offspring either at  
112 juvenile (larva and pupa) or adult stages. Given its powerful genetics, the *Drosophila* model has  
113 proved an efficient strategy for the study of several types of human pathologies, including metabolic  
114 (Baenas and Wagner, 2019; Musselman and Kuhnlein, 2018; Perrimon et al., 2016) neurological  
115 (Coll-Tane et al., 2019; Link and Bellen, 2020; Mariano et al., 2020; Salazar et al., 2020), cardiac  
116 (Birse et al., 2010; Diop et al., 2015; Guida et al., 2019; Piazza and Wessells, 2011), digestive  
117 (Musselman and Kuhnlein, 2018; Nayak and Mishra, 2019) and nephrocytic (Millet-Boureima et al.,  
118 2018; Rani and Gautam, 2018) comorbidities. In addition, tumor models may be induced in larvae or  
119 adult flies by genome manipulation (Dong et al., 2007; Gonzalez, 2013; Hirabayashi et al., 2013;  
120 Igaki et al., 2006; Pagliarini and Xu, 2003; Samji et al., 2021). The fruit-fly enables functional large-  
121 scale analysis to validate relevant molecules and biomarkers in a faster way than with rodent-based  
122 studies ( Fernandez-Hernandez et al., 2016; Papanikolopoulou et al., 2019; Bangi, 2020; Bossen et  
123 al., 2019; Richardson and Portela, 2018).

124 Most of the genes and metabolic routes involved in human hepatic diseases are conserved in  
125 *Drosophila* (Table 1). The conservation between fly and mammalian genes is of utmost interest for  
126 translational studies aiming for a deeper understanding of cellular dysfunctions for which,  
127 investigation is technically restricted in mammalian models and impossible in humans.

128 In contrast to mammals, most invertebrates do not contain an organ equivalent to the liver, although  
129 hepatic functions are conserved. In insects, the fat body (FB) has long been considered as the liver  
130 counterpart (Li et al., 2019), although recent studies suggest that the oenocytes also accomplish  
131 hepatic-related functions (Gutierrez et al., 2007; Storelli et al., 2019). In *Drosophila*, the FB is an  
132 organ that spreads throughout the entire organism and oenocytes are groups of cells located  
133 underneath the abdominal external cuticle. In humans, dietary nutrients are transferred to the liver  
134 through a portal system and lipids are transported through the lymph stream as chylomicrons.  
135 *Drosophila* has an open circulatory system so that nutrients crossing the intestinal epithelium enter  
136 the haemolymph, and thus, the body-wide distribution of the FB favors nutrient uptake (Figure 1A).

137 Further, given its eating sources (mostly rotting fruits), the fruit-fly needs a powerful detoxification  
138 system that involves the gut, the FB and the oenocytes (Huang et al., 2019; Iredale, 2010; Yang et al.,  
139 2007). Over the past two decades, several studies on metabolic hepatic dysfunctions have taken  
140 advantage of the fly model (Ghosh et al., 2020; Hader et al., 2003; Hofbauer et al., 2020; Liao et al.,  
141 2020; Villanueva et al., 2019) to describe the functional activities of molecules relevant for human  
142 pathologies (Hmeljak and Justice, 2019; Heier and Kuhnlein, 2018; Perrimon et al., 2016; Ugur et al.,  
143 2016).

144

### 145 **Homeostatic dysfunctions in NAFLD**

146 Type 2 diabetes mellitus (T2DM) is another metabolic-related disease progressing exponentially. The  
147 T2DM hallmark is the development of insulin resistance, due to impaired response to the hormone or  
148 its reduced production by pancreatic  $\beta$ -cells (Boland et al., 2017; Li et al., 2021), thereby resulting in  
149 high glucose levels, increased general oxidative stress, vascular problems, and serious secondary  
150 perturbations in the physiology and the metabolism of the body (Chatterjee et al., 2017; Galicia-  
151 Garcia et al., 2020). In diabetic patients, lipid metabolism is modified, and the liver may accumulate  
152 fatty acids (FAs), which facilitates the establishment of NAFLD (Kuchay et al., 2020; Sanyal, 2019).

153 In NAFLD, the metabolism of both lipid and glucose in hepatic cells is disrupted (Sanyal, 2019; Shen  
154 et al., 2017). The accumulation of lipids in the hepatocytes results from changes in lipid uptake, *de*  
155 *novo* synthesis of FAs,  $\beta$ -oxidation and export of very-low-density-lipoproteins (VLDL) (Ipsen et  
156 al., 2018; Sanyal, 2019), which together affect the serum levels of TAG and cholesterol and may  
157 provoke instability of body homeostasis. Conversely, these changes in lipid metabolism, which are in  
158 part connected to insulin resistance in the hepatic tissue, reinforce the persistence of liver  
159 dysfunction, indicating that cellular metabolism is integrated at the organismal level (Chao et al.,  
160 2019; Sanyal, 2019). Under a normal and healthy homeostatic environment, hepatic cells use  
161 pyruvate to produce energy through the citric-acid cycle in mitochondria. During this process the  
162 exceeding energy is exported from the mitochondria as citrate to generate palmitic acid that can be  
163 used as a precursor of membrane lipids or esterified to TAGs and stored in cytosolic lipid droplets  
164 (LDs) (Currie et al., 2013; Ramos et al., 2021). These hepatic lipid stores can be remobilized through  
165 the  $\beta$ -oxidation pathway in response to energy demands, or exported from the liver in water-soluble  
166 VLDL particles comprising apolipoproteins, cholesterol and phospholipids (Ipsen et al., 2018).  
167 However, under conditions of metabolic dysfunction, excess LDs may accumulate in the liver and

168 trigger the NAFLD, which favors lipotoxicity and subsequently the development of inflammatory  
169 processes and an increase in the levels of reactive oxygen species. Together, these perturbations  
170 provoke mitochondrial dysfunction and disease progression, leading to insulin resistance, T2DM and  
171 metabolic syndrome (Rosato et al., 2019).

172 Binding of insulin to its cognate receptor activates the mTOR signaling network (Meng et al., 2021;  
173 Yoon, 2017), which acts through a feedback loop to dampen the insulin response by inactivating  
174 insulin-receptor-substrate-1 (IRS1) and repressing the expression of the insulin receptor (InR) and  
175 insulin-like-growth-factor-1 (IGF1). Therefore, in the context of metabolic dysfunction, the mTOR  
176 signaling network is poorly activated and the subsequent absence of the negative feedback loop  
177 sustains an auto-amplification of the insulin resistance process. In this pathological condition, a  
178 number of regulatory proteins are synthesized by the hepatocytes to counteract homeostasis  
179 disruption, including Notch-receptor-2, insulin-like-growth-factor-binding-proteins (IGFBPs), the  
180 potassium-inwardly-rectifying-channel-subfamily-J-member-11, chemokines and kinases (Tamarai et  
181 al., 2019).

182 The PPAR nuclear receptors that can act as lipid sensors to regulate metabolic homeostasis are  
183 potential targets for the development of drug therapy to NAFLD (Liss et al., 2017). The human  
184 genome encodes three PPAR members ( $\alpha$ ,  $\beta/\delta$  and  $\gamma$ ), which are differentially expressed in tissues,  
185 although each of them may impinge on liver metabolism. PPAR $\alpha$  is largely expressed in the liver  
186 where it controls several aspects of FA homeostasis; it is frequently found at low level in patients  
187 with steatosis. PPAR  $\beta/\delta$  is mainly expressed in muscle cells, but at lower levels in adipocytes and  
188 hepatocytes. PPAR  $\gamma$ , which is mainly expressed in adipose tissue, is upregulated in the NAFLD  
189 pathological condition to dampen dysfunction of lipid and glucose metabolism. However, these  
190 unbalanced production of PPARs may contribute to insulin resistance and metabolic syndrome (Liss  
191 et al., 2017; Corrales et al., 2018), supporting the use of PPAR agonists to treat NAFLD {Seo, 2008  
192 #1332; Yoo, 2021 #1331}. The utilization Glucagon-likepeptide-1 (GLP-1) receptor agonists is  
193 another therapeutic strategy currently explored in clinical trials { Dai, 2020 #1334; Lv, 2020 #1333}.  
194 GLP-1 is secreted by enteroendocrine cells after meal ingestion to potentiate insulin secretion and  
195 suppress glucagon production. The secreted bioactive forms act systemically, since the GLP-1  
196 receptor (GLP-1R) is expressed in a number of tissues {Dai, 2020 #1334}. The utilization of GLP-1R  
197 agonists exhibiting a longer half-life compare to that of the genuine hormone has been approved for  
198 T2DM treatment {Andersen, 2018 #1335}. These agonists have positive effects on steatogenesis in  
199 T2DM patient {Teshome, 2020 #1336}, but controversial results about the benefit yet restrain their

200 usage in NAFLD treatment {Lv, 2020 #1333}. Finally, a number of molecules secreted by the  
201 adipose tissue may affect inflammatory processes, insulin resistance and NAFLD progression.  
202 Adiponectin is one such factor that is down-regulated in response to hepatic stress and constitutes a  
203 promising target to treat steatosis, since studies demonstrated that increased levels of this adipokine  
204 ameliorate NAFLD (Scherer et al., 1995; Shabalala et al., 2020). Given the diversity of signals that  
205 may induce metabolic deregulations in hepatic cells, genetic investigations using alternative *in vivo*  
206 models are needed to decipher the organ interconnections that elicit the deleterious condition of  
207 NAFLD.

208

### 209 **Homeostasis and metabolism in *Drosophila***

210 In *Drosophila*, the FB synthesizes and stores glycogen and TAG-containing LDs (Figure 1A)  
211 (Garrido et al., 2015; Gronke et al., 2003; Yamada et al., 2018). In the gut, dietary lipids are  
212 hydrolyzed to glycerol, free FAs and monoacylglycerols and taken up into the enterocytes through a  
213 molecular mechanism yet poorly characterized (Holtof et al., 2019; Miguel-Aliaga et al., 2018;  
214 Toprak et al., 2020). In the enterocytes, these molecules are converted to diacylglycerols that are  
215 loaded in the haemolymph on apolipoprotein particles, functioning as lipid vehicles to the FB (Palm  
216 et al., 2012). However, FB cells deficient for fatty-acid-synthase fail to accumulate LDs, indicating  
217 that *de novo* FA synthesis is essential for lipid storage in these cells (Garrido et al., 2015).  
218 Conversely, fasting induces mobilization and consumption of lipid stores from the FB. The perilipins  
219 Lsd1 and Lsd2 envelop LDs and protect them from lipolysis, whereas the lipase, Brummer, catalyzes  
220 TAG hydrolysis (Bi et al., 2012; Gronke et al., 2005; Gronke et al., 2007). Concurrent to fasting-  
221 induced lipid hydrolysis, oenocytes accumulate LDs (Figure 1B), suggesting that remobilized lipid  
222 stores efflux from the FB and are taken up and oxidized in the oenocytes (Gutierrez et al., 2007). This  
223 process is similar to the remobilization of lipid stores from adipocytes to hepatocytes induced by  
224 fasting in mammals (Iredale, 2010; Yamada et al., 2020). Congruently, enzymes responsible for  
225 ketone body biogenesis are highly expressed in oenocytes (Huang et al., 2019), although it has not  
226 been formally demonstrated whether other tissues (FB, muscles) may also perform  $\beta$ -oxidation to  
227 supply energy demand (Parvy et al., 2012). This energy mobilization process is regulated by target-  
228 of-rapamycin (TOR), which is present in two distinct complexes, TORC1 and TORC2; the former  
229 directly responds to nutrients, whereas the latter is a component of the insulin signaling pathway. The  
230 intermediates of this signaling network are conserved in the fruit-fly (Table 1), although the TORC1

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231 and insulin/TORC2 signaling branches can work independently in most *Drosophila* tissues, including  
232 the FB (Devilliers et al., 2021).

233 The FB also acts as a nutrient sensor to coordinate overall body growth and homeostasis through the  
234 production of insulin-like-peptides (Dilps) by a cluster of neurosecretory cells (NSCs) (Colombani et  
235 al., 2003). Moreover, similar to mammals, sugar metabolism in flies is modulated by an  
236 insulin/glucagon-like axis (Figure 1A-B) (Oh et al., 2019). The *Drosophila* genome encodes eight  
237 Dilps (Brogiolo et al., 2001), Dilp2 and Dilp5, which are produced in the NSCs, are the major  
238 regulators of sugar homeostasis (Rulifson et al., 2002). Genetic ablation of the NSCs results in an  
239 overall reduced body size and in an increase in the levels of glucose and trehalose (disaccharide of  
240 glucose) that is the main circulating sugar in insects. This phenotype resembles the metabolic defects  
241 provoked by insulin deficiency in Type-I-diabetic patients. Conversely, the *Drosophila* adipokinetic  
242 hormone (AKH) produced by the neuroendocrine corpora cardiaca (CC) controls circulating sugar  
243 levels in a glucagon-like manner (Kim and Rulifson, 2004). Both NSCs and CC secrete their  
244 products into the haemolymph, close to the *Drosophila* heart equivalent, thereby favoring hormone  
245 distribution throughout the entire body. Secretion of AKH in *Drosophila* by the CC depends on an  
246 ATP-K<sup>+</sup>-dependent channel that directly responds to circulating sugar levels (Kim and Rulifson,  
247 2004). A pair of glucose-sensing neurons plays a pivotal role in coordinating NSC and CC functions  
248 by activating Dilp secretion, while inhibiting AKH secretion (Figure 1A-B)(Oh et al., 2019).

249 Furthermore, secretion of Dilps by the NSCs strongly relies on FB messengers that relay nutritional  
250 cues (Colombani et al., 2003; Delanoue et al., 2016). The JAK-STAT ligand Unpaired2, is one such  
251 messenger, which can be functionally replaced using molecular genetic tools by human leptin,  
252 showing that as in mammals, fat cells produce an hormone in response to nutrient load to control  
253 feeding physiology (Rajan and Perrimon, 2012). Therefore, although the mammalian liver equivalent  
254 is not a discrete organ in *Drosophila*, hepatic functions and dysfunctions are closely conserved: the  
255 FB appears to be in charge of hepatic functions related to feeding, whereas the oenocytes accomplish  
256 functions related to fasting. Further, both in mammals and flies, integration of the TOR signaling  
257 network at the organismal level is central in controlling lipid and sugar metabolism in response to the  
258 nutritional status (Sanguesa et al., 2019; Schmitt et al., 2015).

259 Most of the mammalian transcriptional regulators involved in NAFLD are conserved in *Drosophila*  
260 (Table 1). Mondo, the ChREBP/MondoA homologue, regulates several metabolic routes in response  
261 to dietary sugar {Mattila, 2015 #869}. The *Drosophila* SREBP regulates the expression of genes  
262 required for FA synthesis (Kunte et al., 2006). However, consistent with insect sterol auxotrophy

263 (Clark and Block, 1959), SREBP activity is not regulated by sterols, but by  
264 phosphatidylethanolamine (Dobrosotskaya et al., 2002). The best homologues of LXRs and PPARs  
265 are the Ecdysone receptor (EcR) and the nuclear receptor Eip75B, respectively (Parvy et al, 2014).  
266 Ecdysone is a steroid hormone that controls developmental transitions, whereas Eip75B is an  
267 intermediate of the ecdysone signaling, whose activity depends on nitric-oxide-synthase.  
268 Surprisingly, manipulating this signaling pathway in the ecdysone-producing gland, results in a  
269 dramatic changes in LD accumulation in FB cells, indicating that this signaling pathway also impinge  
270 on lipid homeostasis (Caceres et al., 2012). In summary, the metabolic routes and most of the  
271 regulatory genes that play a critical role in NAFLD are conserved in *Drosophila*. The activity of  
272 some regulatory gene products varies as compare to that of their mammalian counterparts, but  
273 remains connected to basal metabolism. Importantly, the phenotypes induced by loss-of-function of  
274 these genes can be used as reference criteria to monitor the efficiency and the adverse effects of drug  
275 compounds in preclinical trials.

276

## 277 **Perspectives**

278 Alternative models have largely contributed to our understanding of biological processes. Recent  
279 studies have shed light on potential targets for drug therapy, which need physiological validation  
280 prior to clinical trials. Thanks to a plethora of genetics tools to direct in a tissue-specific manner  
281 either over-expression or RNAi-inactivation of a gene of interest (Ugur et al., 2016) {Senturk, 2018  
282 #1062}, the fruit-fly model emerges as a powerful alternative for large-scale analyses. Collection of  
283 transgenic lines targeting a vast majority of the *Drosophila* genes, with a particular focus on the  
284 orthologues of disease-related-human genes, are available from Stock centers (Heigwer et al., 2018;  
285 Dietzl et al., 2007). This approach validated the functions of relevant proteins, such as CDGSH-iron-  
286 sulfur-domain-containing-protein-2, whose haplo-insufficiency causes NAFLD and promotes HCC  
287 development (Shen et al., 2017). This approach also identified genes that contribute to fat deposition  
288 (Hoffmann et al., 2013; Pospisilik et al., 2010) and whose dysregulation in humans can lead to  
289 obesity, diabetes, and NAFLD (Graham and Pick, 2017). Liver diseases are a burden in the modern  
290 society, especially NAFLD and HCC; *in vivo* investigations using *Drosophila* in translational  
291 approaches will be useful to validate enzymes and other molecules crucial for body homeostasis,  
292 increasing the chance to develop innovative therapeutic strategies.

293

294 **Conflict of Interest**

295 The authors declare that the research was conducted in the absence of any commercial or financial  
296 relationships that could be construed as a potential conflict of interest.

297

298 **Author Contributions**

299 The authors (K.C.M.M. & J.M) contributed equally to the content of the work.

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309

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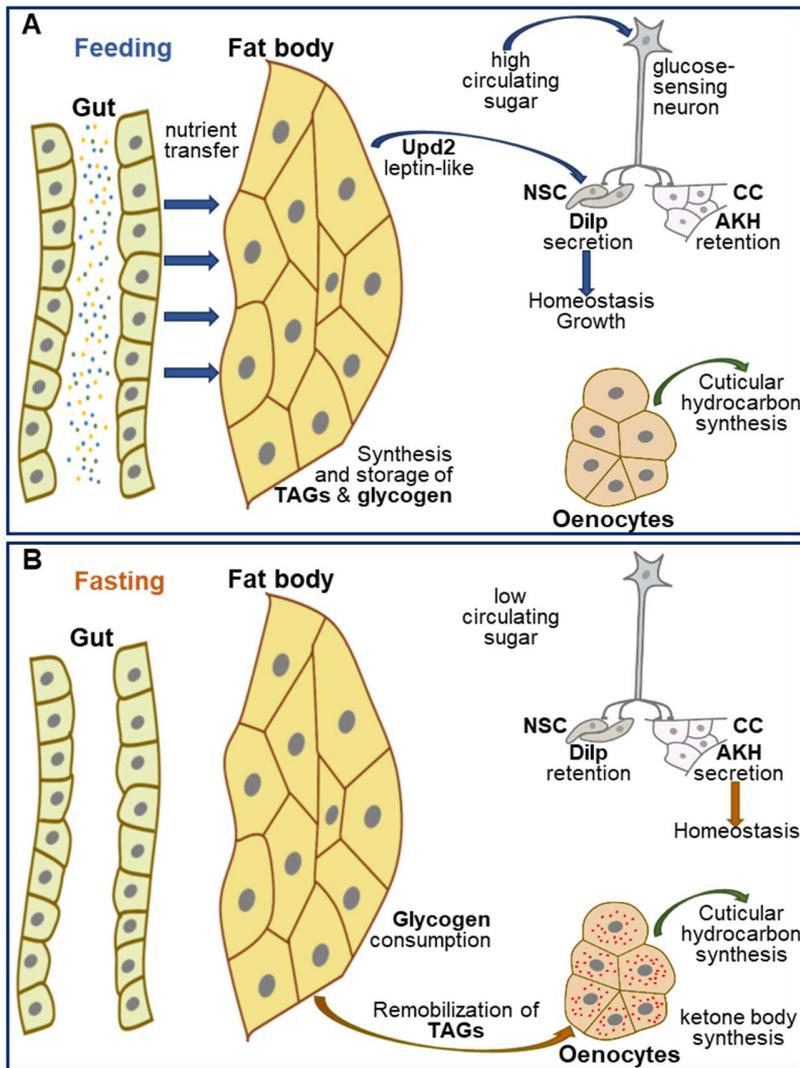
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654 **Figure 1: Hepatic-like homeostasis in *Drosophila*.** (A) In fed *Drosophila*, nutrients are transferred  
 655 to the appropriate organs, in particular the FB, which acts as a nutrient sensor to coordinate  
 656 organismal growth and homeostasis. The FB synthesizes and stores glycogen and TAGs, and secretes  
 657 several proteins, including Upd2, which can be replaced experimentally by mammalian leptin; Upd2  
 658 and glucose-sensing neurons contribute to regulating the secretion of Dilps and AKH to control  
 659 growth and homeostasis. (B) In fasting *Drosophila*, FB glycogen and TAGs are remobilized, the  
 660 latter being transferred to the oenocytes to produce ketone bodies, while glucose sensing neurons  
 661 potentiate AKH secretion and Dilp retention in neurosecretory cells to maintain homeostasis.

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<b>Human</b>	<b><i>Drosophila</i></b>	<b>Function in <i>Drosophila</i></b>	<b>Reference</b>
ChREBP MondoA	Mondo	Regulates sugar usage	<i>Havula et al., 2013</i>
SREBPs	SREBP	Regulates genes of FA synthesis	<i>Kunte et al., 2006</i>
LXRs	EcR	Receptor for the steroid hormone ecdysone	<i>Parvy et al., 2014</i>
PPARs	Eip75B	Intermediate in steroid hormone signaling	<i>Parvy et al., 2014</i>
PNPLA3	Brummer	Performs TAGs breakdown	<i>Gronke et al., 2005</i>
Perilipins	Lsd1 Lsd2	Envelop LDs	<i>Bi et al., 2012</i>
Apolipo- proteins	apoLpp apoLTP MTP	Performs lipid transport throughout hemolymph Performs lipid loading on Lpp in enterocytes Performs lipid loading on Lpp in FB cells	<i>Palm et al., 2012</i>
mTOR	TOR	Regulates growth in response to nutrients	<i>Oldham et al., 2000</i>
Insulin IGF1 IGF2	DILPs	Regulate growth and metabolism	<i>Brogiolo et al., 2001</i>
IGFBP	Imp-L2	Modulates DILP activity	<i>Honegger et al., 2008</i>
InR IGF-R	InR	Receptor for the DILPs	<i>Brogiolo et al., 2001</i>
IRS1	chico	Regulates body growth and aging	<i>Bohni et al., 1999</i>
Glucagon	AKH	Regulates dietary sugar response	<i>Kim and Rulifson, 2004</i>
Leptin	Unpaired2	As for leptin, Udp2 is a ligand for the receptor of the JAK/STAT pathway	<i>Rajan and Perrimon, 2012</i>
ACC1 ACC2	ACC	rate-limiting enzyme for fatty acid synthesis	<i>Parvy et al., 2012</i>
FASN	FASN1 FASN2 FASN3	Ubiquitous FA-synthesis Oenocyte-restricted FA-synthesis Oenocyte-restricted FA-synthesis	<i>Garrido et al., 2015</i>

663 **Table 1: *Drosophila* homologues of human gene products involved in metabolic and liver**  
664 **disease.**