ARTICLE

Effects of trans-resveratrol and dimethyl sulfoxide on intermediate metabolism and lipoperoxidation in *Drosophila melanogaster*

Bibiana Kaiser Dutra[1], Felipe Amorim Fernandes[1],
Gilson Luis da Cunha[2] and Guendalina Turcato Oliveira[1*]

ABSTRACT: (Effects of trans-resveratrol and dimethyl sulfoxide on intermediate metabolism and lipoperoxidation in *Drosophila melanogaster*). Many plants contain the phytoalexin trans-resveratrol. Red grapes and peanuts are rich in this antioxidant which is thought to be responsible for the reported protective effect of wine against diseases and also the ability to retard aging. We investigated the effects of trans-resveratrol on the energetic metabolism in Drosophila melanogaster during the adult life. Adult males and females of D. melanogaster were divides in 3 groups: the treated group received 1µM trans-resveratrol dissolved in 3% dimethyl sulfoxide (DMSO), whereas the control groups received only DMSO or water. For the analyses of energy metabolism (lipids, triglycerides, cholesterol, glycogen and proteins) and lipoperoxidation the flies were treated during 21 days. All these experiments were repeated five times in five different cohorts. Results for the metabolism in control group and treated groups with trans-resveratrol or DMSO showed that both sexes flies showed different pattern responses to the different treatments. Both sexes of flies treated with trans-resveratrol or DMSO showed different values for cholesterol, glycogen and total protein as compared to control flies. These results indicate that both treatments can modulate energy metabolism and lipoperoxidation in this strain of D. melanogaster in both sexes.
Key words: phytoalexin, energetic metabolism, fruit flies, oxidative stress.

RESUMO: (Efeitos do trans-resveratrol e dimetil sulfóxido sobre o metabolismo intermediário e lipoperoxidação em *Drosophila melanogaster*) Muitas plantas contêm a fitoalexina trans-resveratrol. As uvas vermelhas e os amendoins são ricos neste antioxidante, o qual é responsável pelo efeito protetor contra doenças que tem sido descrito para o vinho, além de ser considerado por muitos autores ter a habilidade de retardar o envelhecimento. Nós investigamos os efeitos do trans-resveratrol no metabolismo energético de adultos de *Drosophila melanogaster*. Machos e fêmeas adultos de *D. melanogaster* foram divididos em 3 grupos: o grupo tratado recebeu 1µM de trans-resveratrol dissolvido em dimetil sulfóxido (DMSO) a 3%, enquanto os grupos controles receberam apenas DMSO ou água. Para as análises do metabolismo energético (lipídios, triglicerídeos, colesterol, glicogênio e proteínas) e lipoperoxidação, as moscas foram tratadas durante 21 dias. Todos os experimentos foram repetidos cinco vezes em cinco coortes diferentes. Os resultados do metabolismo do grupo controle e dos grupos tratados com trans-resveratrol ou DMSO mostraram que ambos os sexos apresentaram padrões de resposta diferenciada aos diferentes tratamentos. Moscas de ambos os sexos tratados com trans-resveratrol ou DMSO apresentaram diferentes níveis de colesterol, glicogênio e proteínas quando comparados ao grupo controle. Estes resultados indicam que ambos os tratamentos podem modular o metabolismo energético e a lipoperoxidação em adultos de ambos os sexos nesta linhagem de *D. melanogaster*.
Palavras-chave: fitoalexina, metabolismo energético, mosca das frutas, estresse oxidativo.

INTRODUCTION

Stochastic theories indicate that aging is caused by random accumulation of biological damage and not throughout pre-determined mechanisms (Harman 1956). Oxidative stress is caused by free radicals, commonly called reactive oxygen species (ROS), produced during aerobic respiration, which are thought to be the main factor involved in aging. It is also believed that genetic, biochemical and metabolic factors also exert a great influence on aging, with larger animals having longer lifespans than smaller animals because metabolic rate is inversely proportional to body weight. There appears to be a causal relationship between longevity and metabolism and some studies have demonstrated that in some organisms metabolic alterations induced by environmental factors such as diet and temperature

may produce corresponding changes in longevity as demonstrated by a tendency towards decreased metabolic rate with age (Lints 1989).

Antioxidant defenses against ROS produced during cellular respiration can be endogenous (enzymatic or non-enzymatic) or the result of dietary factors such as vitamins, carotenoids, flavonoids and others (Harman 1994). Increased dietary input of antioxidants help in the maintenance of an appropriate antioxidant equilibrium, defined as the balance between the quantity of oxidizing agents and antioxidants occurring in the organism (Halliwell 1995). Some epidemiological studies appear to confirm the protective effects of a diet rich in antioxidants, by showing that increased consumption of fruits and vegetables appears to be directly related to a reduction in cardiovascular diseases and certain types of cancer (Kohlmeier *et al.* 1995, Steinmetz & Potter 1996,

1. Department of Morphophysiological Science, PUCRS. Av. Ipiranga 6681, Pd. 12, Bloco C, Sala 270, Caixa Postal 1429, CEP 90619-900, Porto Alegre, Rio Grande do Sul, Brazil.
2. Laboratório de Drosophila, Universidade Federal do Rio Grande do Su, Porto Alegre, Rio Grande do Sul, Brazil.
* Autor para contato. E-mail: guendato@pucrs.br

Hininger *et al.* 1997, Ness and Powles 1997).

Trans-resveratrol (trans-3,4',5-trihydroxystilbene) was first detected in grapes (*Vitis vinifera*) by Langcake and Pryce (1976) who suggested that this compound was produced as a protective mechanism against phytopathogenic fungi (principally *Botrytis cinerea*) or ultraviolet (UV) light. The first study on the biological effects of trans-resveratrol was carried out by Siemann and Creasy (1992) who detected this compound in wine and suggested that trans-resveratrol was responsible for the biological properties of red wine and marking the start of scientific studies on the biological properties of this compound. Studies indicate that polyphenols are the largest dietary source of protective antioxidants, with trans-resveratrol being the main active antioxidant (Hertog 1996, Hertog *et al.* 1994, 1995, 1997).

It is already known (Fremónt 2000) that trans-resveratrol has the following functions in biological systems: inhibition of lipid peroxidation in membranes and low-density lipids; copper chelating, antioxidant blocking of free radical reactions; modification of eicosanes synthesis; inhibition of platelet aggregation; modulation of lipid metabolism; inhibition of angiogenesis; inhibition of cyclo-oxygenase activity; and possesses anticarcinogenic, estrogenic, vasodilatory and potent anti-inflammatory activity. The structure of trans-resveratrol is similar to that of the estrogen diethylstilbestrol and could therefore substitute estradiol (Bitterman *et al.* 2003) and have important effects on the cardiovascular system (Fremónt 2000). Work development by Howitz *et al.*(19) showed that cells treatment with a low resveratrol concentration (0.5 μM) increased cell survival after ionizing irradiation and higher concentrations (>50 μM) had the opposite effect.

The research described in this paper was carried out to assess the effects of trans-resveratrol on the lifespan of *Drosophila melanogaster* (Oregon-R strain), by investigating the effects of this agent on longevity, as well as energy metabolism and lipoperoxidation in different ages (0, 7, 14 and 21 days).

MATERIALS AND METHODS

The animals were collected and used with the permission of the Ethics Committee of the Pontifícia Universidade Católica do Rio Grande do Sul (PUCRS) (License 0001/03) and Brazilian laws.

Exposure to trans-resveratrol

Pairs of newly eclosed male and female *Drosophila melanogaster* (strain Oregon-R) were placed in glass vials containing a standard *Drosophila* culture media and a filter-paper containing the yeast *Saccharomyces cerevisiae* and the vials divided in three groups of 45 vials. The media of standard culture contained 10% rye flour, 5% dark sugar, 1% nipagin fungicide and 1% agar-agar. Details on the origin and maintenance of these populations have been provided by Oliveira & Cordeiro

(1981). For the trans-resveratrol experimental group 65 μl of a 1 μM trans-resveratrol solution dissolved in 3% (v/v) dimethyl sulfoxide (DMSO) was added to the filter-paper in each vial, being this solution replaced by 65 μl of DMSO 3% in the DMSO control group or by 65 μl of distilled water in the water control group. All these experiments were repeated five times in five different cohorts. The low resveratrol dose was chosen since different works reported that the minor trans-resveratrol concentration that shows biological effects in different animals was 1μM (Hertog *et al.* 1994, 1996, 1997).

Biochemical analyses

The metabolic determinations were made from homogenized pools of animals. Pools of twenty males or twenty females each were used for determination of glycogen and total proteins, fifty males or fifty females for quantification of total lipids and triglycerides, and fifty males or fifty females for quantification of lipoperoxidation levels for each cohorts. Since the *D. melanogaster* Oregon-R is an inbred strain, the individual variation can be considered as not significant in the pools. All determinations were performed five times in five different cohorts.

a. Glycogen was extracted from tissues following the method described by Van Handel (1965). Glycogen levels in the animals were determined as glucose equivalent (glucose-oxidase method), after acidic hydrolysis (HCl) and neutralization (Na_2CO_3), following the method of Geary *et al.* (1981). Glucose was quantified using a Biodiagnostic kit (glucose-oxidase). Results are presented as mmol/g of tissue.

b. Proteins were measured following the method described by Lowry *et al.* (1951), using bovine albumin (Sigma Chemical Co., St. Louis, Missouri, U.S.A.) as the reference. Results are expressed in mg/ml of homogenate.

c. Lipids were extracted from tissue weighted and homogenized in a 2:1 (v/v) chloroform-methanol solution, according to Folch *et al.* (1957). Total lipids in this homogenate were determined by the sulfophosphovanillin method (Frings & Dunn 1970; Meyer & Walter 1980). This method consists of oxidizing cellular lipids to small fragments after chemical digestion with hot concentrated sulfuric acid. After the addition of a solution of vanillin and phosphoric acid, a red complex is formed, which is measured at 530 nm in a spectrophometer. Triglycerides were measured by the reactions of lipase, glycerokinase, 1-P-glycerol oxidase, and peroxidase enzymes (Biodiagnostic Kit/GPO Trinder). Cholesterol was measured by a Labtest kit (Total Cholesterol Liquiform). Results are expressed as mg/g of tissue.

d. Lipoperoxidation levels were quantified through the method of Buege and Aust (1978), by measuring thiobarbituric acid reactive substances (TBARS), using the extraction method of Llesuy *et al.* (1985). The results were expressed in nmol of TBA/mg of protein.

All metabolic parameters passed the Kolmogorov-Smirnov test for normality and the Levene test for homogeneity. Biochemical data was analyzed using one-way analysis of variance (ANOVA) and the Bonferroni comparison test. The different treatments and sexes were compared using two-way ANOVA. All tests were run using the Statistical Package for the Social Sciences (SPSS) program, version 11.5 for Windows®.

RESULTS

In the present study we found that, seven days after eclosion, female flies of the control group showed a significant ($p<0.05$) decrease in total lipids and triglycerides and that these levels remained more or less constant until the end of 21 days study period (Fig. 1), these results contrasting with those for cholesterol levels which were found to show a light tendency to increase

gradually from eclosion to 21 days (Fig. 1C).

In DMSO treated group (Fig. 1) females at 21 days post-eclosion total lipids rose by about 4 times and triglycerides by about 2 times as compared to the values at eclosion, while cholesterol levels showed increase of 66,67 times at seven days and increase of 50 times at 21 days post-eclosion.

For female flies in the trans-resveratrol treated group (Fig. 1) the lipid levels remained constant up to 14 days post-eclosion but by 21 days post-eclosion had risen by 4 times in relation to the amount present in newly eclosed (day zero) females in the same group or in the control group at 21 days, with triglycerides levels showing the same basic pattern except that in this case the increase at 21 days was 150% as compared with the values at eclosion for females in the same group or the 21 day post-eclosion values for control group females. Cholesterol levels in females in the trans-resveratrol group were found to be increased only at day seven, the values at
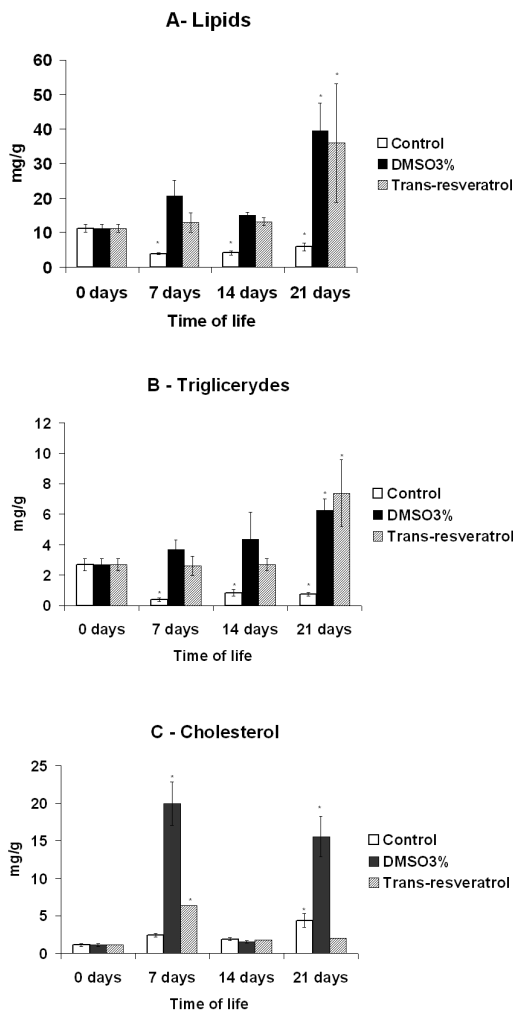

Figure 1. Lipids (A), Triglycerides (B) and Cholesterol (C) in female *D. melanogaster* fruit flies as measured from eclosion to day 21 post eclosion. Columns represent mean values ± the standard error of the mean. Asterisks indicate significant differences between the values for the columns they represent and values in the columns for the same group at eclosion.

Figure 2. Levels of Glycogen (A), Proteins (B) and Lipoperoxidation (C) in female *D. melanogaster* fruit flies as measured from eclosion to day 21 post eclosion. Columns represent mean values ± the standard error of the mean. Asterisks indicate significant differences between the values for the columns they represent and values in the columns for the same group at eclosion.
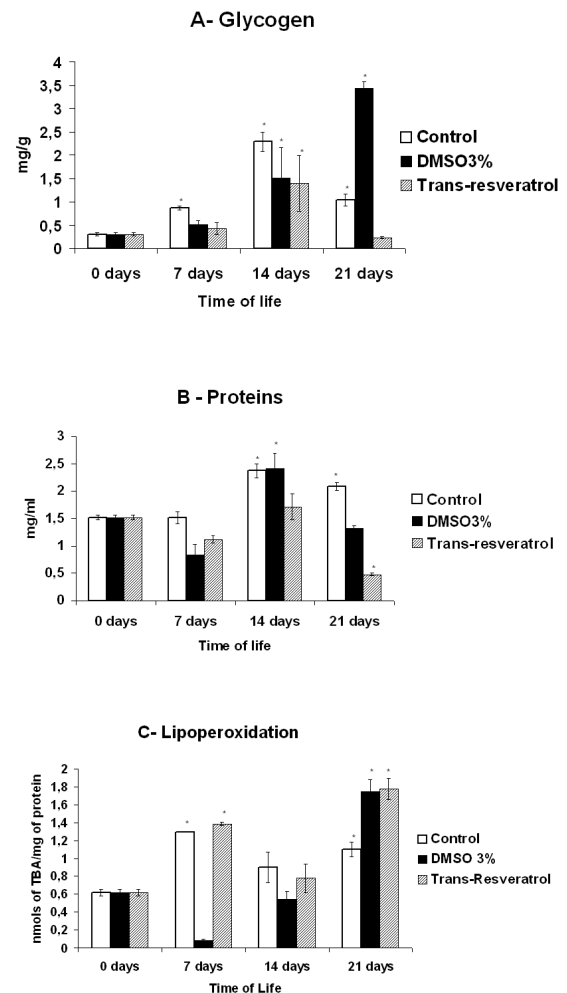
14 and 21 days being similar to those of control group females (Fig. 1C).

In our study we found that seven days after eclosion glycogen levels rose above basal levels in the female flies in the control group and remained high until the end of the experiment at 21 days post-eclosion, although the higher level was measured at 14 days (Fig. 2). In the DMSO-treated group there was a gradual increase in glycogen in the female flies throughout the experiment with 14 day and 21-day post-eclosion glycogen levels being higher than those at eclosion. The glycogen reserves of trans-resveratrol treated females 14 days after eclosion were about 3 times higher than the levels at eclosion (significant at $p < 0.05$) but by 21 days post-eclosion the glycogen reserves of trans-resveratrol treated females had dropped back to the basal levels found at eclosion (Fig. 2A).

Figure 2B shows the total protein content of untreated control and DMSO or trans-resveratrol treated females during their 21-day lifespan, from which can be seen that 14 days post-eclosion control group females showed an increase in protein levels which remained high until the

end of the study at 21 days post-eclosion, while females treated with trans-resveratrol showed a decrease in protein levels between 14 and 21 days post-eclosion. Treatment with DMSO produced an increase in protein levels in control group females 14 days post-eclosion which was followed at 21 days by a return to protein values similar to the those seen in control group females at eclosion (day zero). Treatment with DMSO produced a significant difference ($p < 0.05$) as regards to control group females but not to trans-resveratrol group females ($p > 0.05$).

The lipoperoxidation values for female are shown in Fig. 2C, from which it can be seen that in control group females the highest lipoperoxidation values occur seven days post-eclosion and these values remained high until the end of the experiment at 21 days. Female flies treated with DMSO showed an increase in oxidative damage only at the end of the experiment 21 days post-eclosion. In trans-resveratrol treated females there was an increase in lipoperoxidation values at seven days post-eclosion but oxidative damage at 14 days was reduced again, although the lipoperoxidation values increased again at 21 days post-eclosion. Two-way ANOVA of the data shown in figure 2C demonstrated that each treatment (control, DMSO and trans-resveratrol) modulated the
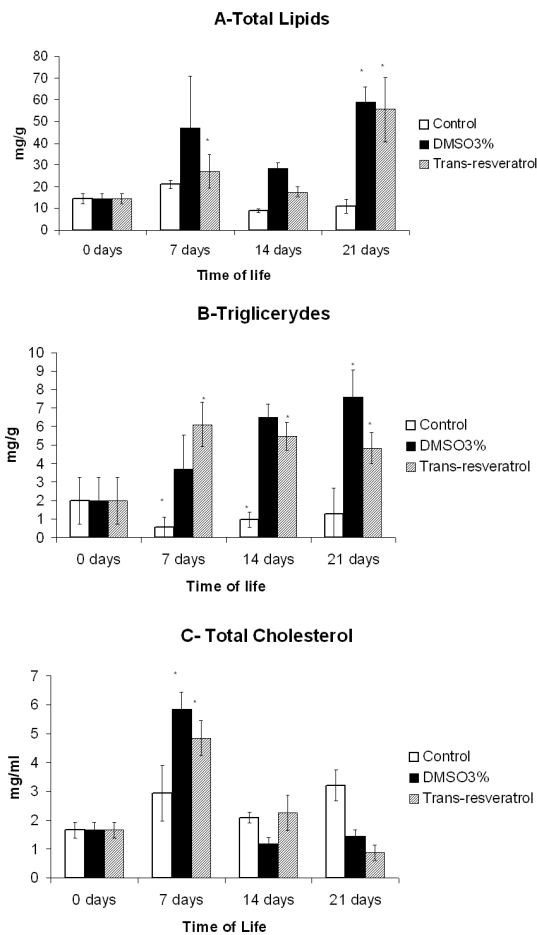
Figure 3. Lipids (A), Triglycerides (B) and Cholesterol (C) in male *D. melanogaster* fruit flies as measured from eclosion to day 21 post eclosion. Columns represent mean values ± the standard error of the mean. Asterisks indicate significant differences between the values for the columns they represent and values in the columns for the same group at eclosion.
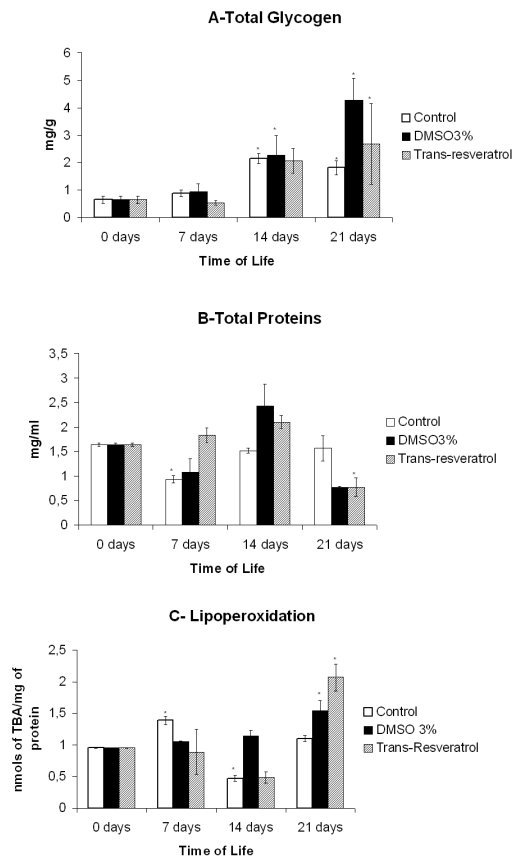
Figure 4. Levels of Glycogen (A), Proteins (B) and Lipoperoxidation (C) in male *D. melanogaster* fruit flies as measured from eclosion to day 21 post eclosion. Columns represent mean values ± the standard error of the mean. Asterisks indicate significant differences between the values for the columns they represent and values in the columns for the same group at eclosion.

lipoperoxidation profile of female flies in a different manner.

In our study the control group male flies showed no significant variation in total lipid levels at any time throughout the 21 days of the experiment, but males treated with DMSO showed two lipid peaks, one seven days after eclosion and another at 21 days, while in males treated with trans-resveratrol there was a two-fold increase in lipids at the end of the adult life cycle (day 21) (Fig. 3A).

We also found that the triglyceride levels of control group male flies were higher at eclosion than at seven days post-eclosion and remained low until the end of the experiment at 21 days post-eclosion, this reduction of triglyceride levels coinciding with the period preceding the reproductive peak of *D. melanogaster* (Fig. 3B). In DMSO treated males there was a gradual increase in triglyceride levels from eclosion to 21 days post-eclosion, while in trans-resveratrol treated male flies there was an increase in triglyceride levels at seven days post-eclosion and these levels remained high until the end of the experiment at 21 days post-eclosion (Fig. 3B).

Our results for cholesterol in control group males were similar to those for total lipids in that no significant variation in total cholesterol levels occurred in control group males during the 21 days of the experiment. However, cholesterol values in males treated with DMSO or trans-resveratrol showed a four-fold increase in total cholesterol seven days after eclosion, although at 14 day and 21 day post eclosion total cholesterol levels returned to values similar to those occurring at eclosion (Fig. 3C).

Males in the control and DMSO groups presented significantly lower ($p<0.05$) glycogen levels at eclosion that at 14 and 21 days post-eclosion, with DMSO group males showing a large increase in glycogen levels at 21 days. Males treated with trans-resveratrol maintained constant glycogen values up to 14 days post-eclosion but these increased significantly at the end of the study at 21 days (Fig. 4A). In the control and trans-resveratrol groups both sexes showed differences in glycogen profile over time (Figs. 2A and 4A) but there were no significant quantitative differences between the glycogen profiles of the two sexes in the DMSO group.

The protein levels in control group males decreased significantly ($p<0.05$) seven days after eclosion but by 14 days post-eclosion these levels had returned to the basal values seen at eclosion and remained at these values until the end of the experiment at 21 days (Fig. 4B). The decrease in protein levels in control group males seven days post-eclosion coincides with the period preceding the *D. melanogaster* reproductive peak but was not seen in the females control group (Fig. 4B). In the DMSO group the protein levels for both sexes were maintained more-or-less constant throughout the 21 days of the experiment, whereas in the trans-resveratrol group both males and females showed a 50% decrease in protein levels 21 days post-eclosion.

The lipoperoxidation data indicates that oxidative damage at eclosion (day zero) is more severe in control group males (Fig. 4C) than control group females (Fig. 3C) and also shows that in this group lipoperoxidation peaks seven days after eclosion but subsequently decreases at 14 days post-eclosion and remains low until the end of the experiment 21 days post-eclosion. Throughout the 21 days of the experiment flies of both sexes in the trans-resveratrol and DMSO groups showed significant modifications in the oxidative damage profile as compared with flies of both sexes in the control group, the trans-resveratrol and DMSO groups showing lipoperoxidation peaks only at the end of the experiment 21 days post-eclosion (Fig. 4C).

DISCUSSION

The use of *D. melanogaster* as an experimental model for the study of aging processes presents certain limitations. The somatic cells of adults of this fly are post-mitotic, i.e., do not divide. In principle, this is a limitation, because mammalian cells do undergo a process of division and, consequently, turnover (although not in neurons). However, it makes possible to evaluate the effects of cumulative molecular damage to somatic cells during aging (Da Cunha & Oliveira 1996). Furthermore, the use of *D. melanogaster* allows to estimate the contribution of epigenetic modifications to the senescence process, and in particular to detect non-enzymatic modifying events such as racemization, deamination and glycosylation of proteins, which may clarify various aspects prior to beginning investigation of the same processes in vertebrates (Miquel & Econômos 1982, Lee *et al.* 2001, Lezhava 2001, Valenzano *et al.* 2006).

The effects of different trans-resveratrol concentrations on the mitochondrial respiratory chain in mice brain cells were studied by Zini *et al.* (1999) who found that when mitochondrial respiratory chain complexes I to V were activated by glutamate and malate combined trans-resveratrol concentrations (1^{-11}M to 1^{-4}M) showed an effective decreasing in respiratory chain. Zini *et al.* (1999) also showed that trans-resveratrol inhibits the respiratory chain complexes I to III and is capable of not only inhibiting the complex III enzyme ubiquinone cytochrome-C reductase (Coenzyme Q) by up to 20%, but also inhibiting mouse brain ATPase activity and acting as a protecting agent in other peroxidation systems as well as during lipid peroxidation in cerebral synaptosoma and the scavenging of superoxide radicals. These results suggest that the trans-resveratrol acts not only if opposing the production of ROS but also scavenges them.

During the experiment (21 days) of metabolic analyzing there was a significant difference ($p<0.05$) between females in the control group and females in the trans-resveratrol and DMSO groups in respect to total lipids, triglycerides and cholesterol (Fig. 1). Figure 1C suggests that DMSO modulated cholesterol metabolism during both the initial part of the lifespan of the female flies

and at 21 days post-eclosion, whereas trans-resveratrol showed an effect only during the initial part of the 21 day period, these differences between DMSO and trans-resveratrol being statistically significant ($p<0.05$).

In insects, lipid reserves produced during larval feeding are important for first flight activities, homeostasis, courtship and reproduction and are especially important for the survival of females which need large amounts of energy for gamete production and vitellogenesis (Chapman 1998, Dutra *et al.* 2007). Our study reinforces this importance because there was a decrease in total lipids and triglycerides in female flies in the control group seven days after eclosion, before the reproductive peak for female *D. melanogaster*, and treatment with trans-resveratrol or DMSO appearing to prevent such decreases by maintaining lipids and triglycerides concentrations for up to 21 days post-eclosion.

Our findings are additionally supported by the study carried out by Borash & Ho (2001) studying female *D. melanogaster* concerning late fertility or starvation-resistance by caloric restriction have found increased levels of lipids and other metabolites. This study demonstrated that when female *D. melanogaster* are maintained at high population density for multiple generations there is an increase in lipid reserves. Da Cunha & Oliveira (1996), working with *D. melanogaster* selected for resistance to the oxidant agent Paraquat and larval starvation, showed that, in order to be resistant to both conditions, the selected individuals should be carriers of the fast electrophoretical variant of the NADP-dependent isocitrate dehydrogenase (IDH-NADP). The activity of this enzyme supplies NADPH for both glutathione reducing and lipogenesis (Cunha & Oliveira 1996).

According figure 2A there was a significant difference ($p<0.05$) in respect to glycogen profile between trans-resveratrol treated female flies and untreated control females and DMSO control females, with the trans-resveratrol treated females maintaining their glycogen levels throughout the course of the experiment, this phytoalexin appeared to modulate glycogen metabolism throughout the lifespan of the female flies principally because the older animals were capable to maintained the glycogen reserves. Trans-resveratrol has been shown to have an inhibitory effect on the respiratory chain in mice neurons (Zini *et al.* 1999). This effect can alter the ATP: ADP ratio and increased glycogen degradation or decreased glycogen synthesis could make carbon skeletons available and promote buffering of alterations in the glycolytic pathway. This hypothesis is reinforced by studies that show that there is up to four-fold increase in the expression of genes coding for the glycolytic enzyme glucose–6-phosphate dehydrogenase (G6PD) and the glycogen synthesis inhibitor Isopentenyl Pyrophosphate (IPP-2) in the muscular fibers of calorie-restricted mice (Lee *et al.* 1999, Voohies *et al.* 2001, Ji Li 2001, 2002). Furthermore, increased expression and / or activity of G6PD have been proposed to represent an important genetic influence on the life span of several organisms, since this enzyme is described as one of the main sources of reducing power (NADPH) for glutathione regeneration from its oxidized to its reduced form (Cheng *et al.* 2004).

In our study, treatment with DMSO appeared to increase the accumulation of glycogen in female flies at 21 days post-eclosion as compared to both control and trans-resveratrol treated females. Glycogen is associated with the production of glycerol and a study of *D. melanogaster* populations selected for increased lifespan and exposed to low temperatures showed that flies lived longer and were more resistant to low temperatures when they possessed higher glycogen reserves and it is possible that high glycogen reserves play a role not only in flight, reproduction and oviposition but also aids *D. melanogaster* in surviving low temperatures (Luckinbill 1998).

In dipteran insects proteins are not only a rich source of amino acids for oocyte production and the formation of mRNA during embryogenesis, but also transferred to the eggs during vitellogenesis, and this may explain the fact that highest protein levels occurred at 14 days which is the reproductive peak for *D. melanogaster* (Chapman 1998). It is interesting to note that the highest lipoperoxidation values in the control group females occurred before the reproductive peak, which may be related to the fact that reproduction requires high levels of energy which means that high levels of oxidative damage are maintained until after the reproductive peak at 14 days post-eclosion.

A theoretical study by Longo & Finch (2003) on the regulation of longevity genes suggests that there are genes associated with the regulation of proteins such as catalase, heat-shock proteins, superoxide dismutases, and serine and threonine kinases. The conserved nature of longevity genes is also supported by homologous genes present in yeasts and the nematode *C. elegans* that provide increased protection against oxidative damage and other forms of stress, while conserved genes possibly involved in longevity have also been observed in *D. melanogaster* (Longo & Finch 2003). Rissanen *et al.* (2003) have also pointed out that mutations that decrease the activity of the insulin/insulin-like growth factor 1 (IGF-I) route not only increase the storage of nutrients and the expression of enzymes such as superoxide dismutase but also increase longevity. The similarity of these metabolic routes suggests that genes associated with the regulation of longevity have developed from a common ancestral form.

Our results show that male *D. melanogaster* treated with trans-resveratrol or DMSO showed similar responses regarding lipids, triglycerides and total cholesterol but there was a significant difference ($p<0.05$) between male flies in these two groups and males in the control group (Fig. 3, A, B and C). As was observed for female *D. melanogaster*, treatment with trans-resveratrol or DMSO increased lipid reserves of post-eclosion male *D. melanogaster*. As was discussed above in relation

to female flies, lipid levels are very important for the survival of insects such as fruit flies (Lee *et al.* 1999). We found that trans-resveratrol and DMSO appeared to have the same effect on total lipid and triglyceride values except that the cholesterol levels of females treated with DMSO were different to the cholesterol levels of females treated with trans-resveratrol, possibly because of the estrogenic effects of trans-resveratrol (Fremónt 2000).

Although a literature search failed to find any reference to DMSO effects on intermediary metabolism, work has been published in relation to the effects of ROS in *Microcystis aeruginosa* where DNA was damaged by the hepatotoxic cyclic heptapeptide microcystin (a potent inhibitor of protein phosphatase type I and 2A), the solvent for which was 5% (v/v) DMSO which was described as a hydroxyl radical scavenger (Zegura 2004). These data support results from our laboratory which show that in female *D. melanogaster* treated with either resveratrol or DMSO lipoperoxidation rates are at a minimum 14 days post eclosion, coinciding with the period immediately after the reproductive peak of this species. The reproductive peak in *Drosophila* requires high levels of energy and a probable concomitant increase in lipoperoxidation, such an increase having been noted in control flies (i.e. those not exposed to DMSO or trans-resveratrol) in this work. The data in figure 4B suggests that both DMSO and trans-resveratrol may be acting as antioxidants which reduce oxidative damage and cause peak mortality for the DMSO and trans-resveratrol groups to occur later in relation to the control group.

In summary, our results indicate that treatment with trans-resveratrol modulated both energy metabolism and lipoperoxidation in female and male *D. melanogaster*. We also suggest for the first time that DMSO can have an effect on energy metabolism and lipoperoxidation.

ACKNOWLEDGEMENTS

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