

# EFFECT OF *Chenopodium ambrosioides* L. (CHENOPODIACEAE) AQUEOUS EXTRACT ON REPRODUCTION AND LIFE SPAN OF *Drosophila melanogaster* (Meigen) (Diptera: Drosophilidae)

## EFEITO DO EXTRATO AQUOSO DE *Chenopodium ambrosioides* L. (CHENOPODIACEA) NA REPRODUÇÃO E TEMPO DE VIDA DE *Drosophila melanogaster* (Meigen) (Diptera: Drosophilidae)

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**ABSTRACT:** Several plants used in folk medicine has bioactivity that depends on dosage. *Chenopodium ambrosioides* is a plant with anti-helminthic and insecticide activities. The objective of this work was to observe effects of *C. ambrosioides* on life span and reproduction of a model-organism, *Drosophila melanogaster*. Larval and adults were fed on two kinds of medium: a standard nourishing medium and standard nourishing medium with 0.1 % of aqueous extract of *C. ambrosioides*. The flies reared on medium with plant extract had less offspring and significative difference in sex ratio. The life span of both sex were affected by the plant aqueous extract: females seems to die early than males. Larval phase survivor females reared on medium with extract had a longer life span, which can be attributable to shorter life span of males reared on medium with plant extract.

**KEYWORDS:** Folk Medicine. Plant Extract. Model-Organism. Botanical Insecticide.

### INTRODUCTION

*Chenopodium ambrosioides* is popularly called as “epazote” in Mexico and Central America, “Mexican tea in the U.S.A” and “erva-de-santa-maria” in Brazil. This plant is one of the most used plants in folk medicine in Latin America, because it has anti-helminthic activity. Besides of its phytoterapic use, this plant is toxic for insects and has potential use as botanical insecticide (GADANO et al., 2002; TAVARES; VENDRAMIN, 2002). The most bioactive substance of *C. ambrosioides* is an essential oil, the ascaridole, a well-known worm repelent substance (GADANO et al., 2002).

The utility of bioactive substances of plants ranges from pharmaceutical industry to the agricultural pest control. In researches seeking plant extracts bioactivity; it is necessary using a model-organism that displays how the compounds reveal this bioactivity. Models organisms can be used in investigation research concerning plant extracts bioactivity. The fruit fly *Drosophila melanogaster* is considered one of the best models, because is widely used to research problems requiring multidisciplinary approach (RUBIN, 1988). *D. melanogaster* is easy to handle, requires small space and allows large number of repetitions, which assure good statistical precision (GRAF et al., 1984). *D. melanogaster* has been an organism used for bioassays to assess genotoxic and anti-genotoxic

effects of medical plants (OLIVEIRA; NEPOMUNCENO, 2004)

This work has the aim to observe effects of aqueous extract of *C. ambrosioides* leaves on longevity and offspring production of *D. melanogaster*. The fertility and duration of life span are good parameters indicators of viability of organism. They are influenced not only by genetics factors, but also by many environmental factors (ZIVANOV-CURILS et al., 2004). Life span and fertility of *D. melanogaster* are smaller under effects of toxic substances (HO; CLIFFORD, 1997).

### MATERIAL AND METHODS

The *D. melanogaster* stock population was obtained from colonies of Genetics Laboratory at Santa Maria Federal University (UFSM). This starter culture was kept into BOD at 25±0.5 °C with 75 % of air humidity, inside of glass tubes with 200mL of volume. The adults and larvae of flies were fed on standard nourishing medium, which stock was prepared with 2 kg corn flour, 280g wheat germ, 150g sucrose, 60g milk powder, 20g NaCl, 10 mL ethanol, 1g nipagim and 600mL water distilled. In order to prepare the medium with plant extract, about 1.2g of fresh leaves was smashed with 240 mL of distilled water of medium. This solution was mixed with 1 kg of components of standard medium. Thus, we obtained a

concentration about 0.1 % of plant extract in flies's standard nourishing medium. Part of stock population was transferred for a new glass tube containing standard medium. We selected 64 males and 64 females from F1 of transferred flies, which form 32 couples for control-group (reared on standard medium) and 32 couples for test-group (reared on standard medium with plant extract). Four couples were put inside 15 mL glass tubes. Daily, we observed dead and alive flies. In order to quantify adult emergence (offspring viability), the survivor flies were transferred for new tube each three days at most for prevention of mixture between parents and their progenies in the same tube. The assessment of offspring production was finished a week after death of last female. The biological parameters of two groups was compared by Student's *t*-test ( $p < 0.05$ ). This experimental design was replicate at twice.

## RESULTS AND DISCUSSION

No statistical differences were found between two replicates of control and between two replicates of test groups, which assured good experimental conditions for assessment of biological parameters. The offspring production of treated and control flies was low in comparison with that was found by Zivanov-Curils et al. (2004) (Table 1). We assume that low fertility is a characteristic of this experimental population .

The plant extract reduced about 44.5 % of offspring production per female (Table 1). It indicate existence of negative effect of aqueous extract of *C. ambrosioides* on reproduction. Less reproduction can be an indicative of toxic effect from a substance (ZIVANOV-CURILS et al., 2004) Some secondary plants compounds can reduce fecundity of insects, in both sexes (MORDUE; NISBET 2000), because bioactive compounds can interfere directly on oogenesis process. In this research was not possible determine if males or females, or both had formation of germ cells impaired by plant extract.

In both groups, male number was greater than females (Table 1). The sex ratio male/female was not statistically different for control group ( $X^2 = 0.75$ , d.f.=1  $p = 0,386$  ). For experimental group, the sex ratio was different ( $X^2 = 3.893$ , d.f.=1  $p = 0,048$ ). As hypothesis, larval survivorship can be different according to their sex, females can be more sensible to plant extract, which implies in differences of sexual ratio. Choudhary (2003) observed that nicotine (tobacco plant extract) affects adult emergence of males and females of mutant form yellow of *D. melanogaster*. We suggested that there was an evident selection effect on larval phase of females.

As expected, sex had effect on life span duration. Males had greater longevity on comparison with females in the control group (Table 2).

**Table 1.** *Drosophila melanogaster*. Offspring produced by female (average  $\pm$  sd) and proportion of female/male in control group and group treated with 0.1% of *Chenopodium ambrosioides* leaves aqueous extract .

Group	Offspring produced per female	Male/female proportion	Student's <i>t</i> -test	p
Control	198.10 $\pm$ 172.9	1.058:1.000	2.98	0.006
Treated	110.0 $\pm$ 69.0	1.133:1.000		

**Table 2.** Longevity of control and treated groups of *Drosophila melanogaster* (reared on diet containing 0.1 % *Chenopodium ambrosioides* extract according to the gender within group (male x female)

Group	Sex	Gender Longevity (means $\pm$ sd)	Student's <i>t</i> -test	p
Control	male	14.81 $\pm$ 7.10	3.85	< 0.0001
	female	9.66 $\pm$ 4.42		
Treated	male	11.07 $\pm$ 4.56	1.15	0.258
	Female	13.12 $\pm$ 5.31		

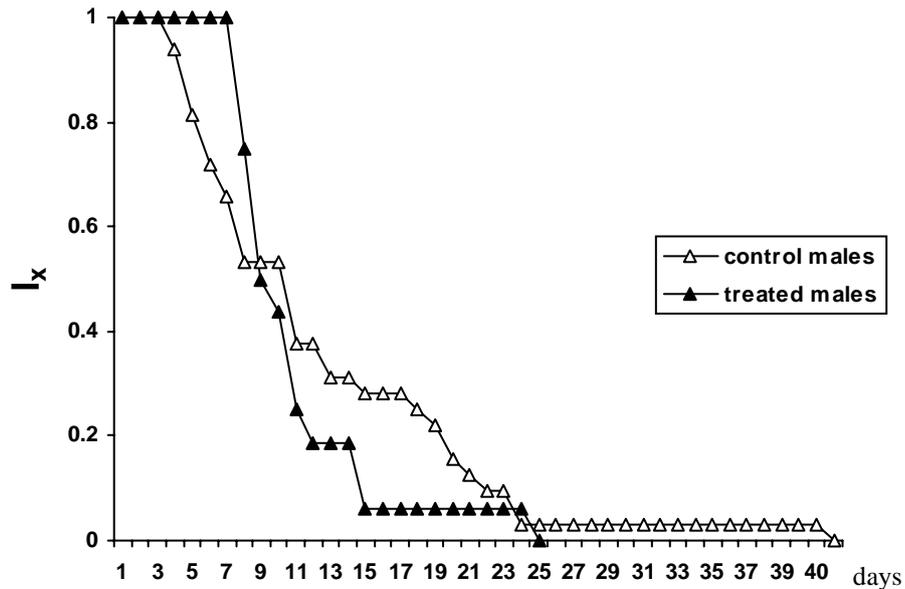
According to Barnes et al. (2008) the *Drosophila* female longevity is shortened after successive matings, which could explain their

shorter longevities than males longevities in control group. Differently, there is no difference on life span of males and females when they were reared

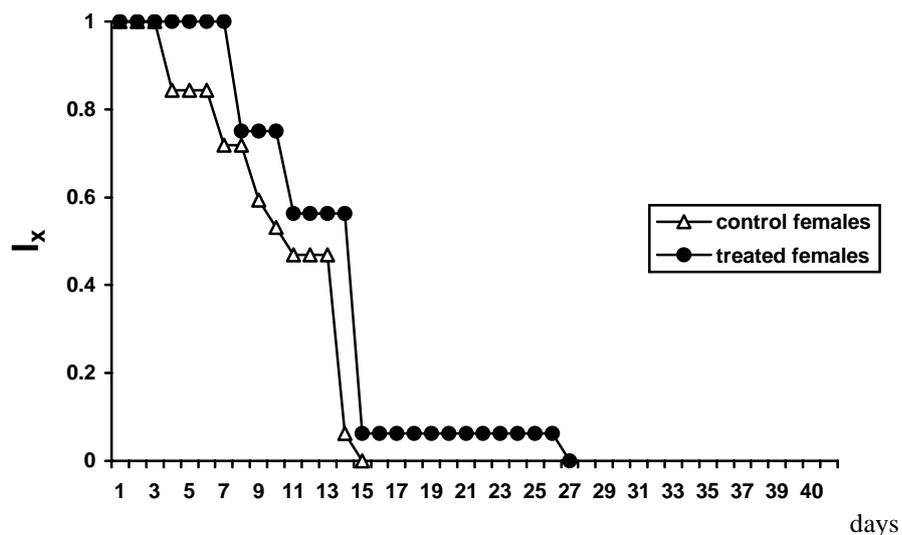
on medium containing *C. ambrosioides* extract (Table 2). The male longevity seems to be affected by extract plant. The males, which larval phase fed on diet with plant extract, had a shorter life span than males reared on standard diet in average ( $p < 0.05$ ) (Table 2, Figure 1).

These results suggest that fertility of female decreased in consequence of shorter male life span in test group. In consequence, females of test group survived longer than those in control group ( $p =$

0.021) (Table 2, Figure 2). We concluded that experimental dosage of aqueous extract of *C. ambrosioides* affect negatively both sex of *D. melanogaster*, but in different way, males were affected lately in their life span when in comparison with females. But, direct effect of plant extract on fertility of *D. melanogaster* females needs an appraisal based on careful analytical evaluation.



**Figure 1.** Survivorship of *Drosophila melanogaster* males ( $l_x$ ) reared on standard medium (control males) and on standard medium containing *Chenopodium ambrosioides* extract at 0.1% concentration (treated males).



**Figure 2.** Survivorship of *Drosophila melanogaster* females ( $l_x$ ) reared on standard medium (control females) and females reared on standard medium containing *Chenopodium ambrosioides* extract at 0.1% concentration (treated females).

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**RESUMO:** Muitas plantas usadas na medicina popular possuem bioatividade dependente da dosagem. *Chenopodium ambrosioides* é uma planta com atividades anti-helmínticas e inseticidas. O objetivo deste trabalho foi observar efeitos de *C. ambrosioides* na duração de vida e reprodução de um organismo modelo, *Drosophila melanogaster*. Larvas e adultos foram alimentados em dois tipos de meio: um meio nutritivo padrão e um meio nutritivo padrão com adição de 0,1% do extrato aquoso de *C. ambrosioides*. As moscas criadas no meio com o extrato da planta tiveram uma progênie menor e diferença nas razões do sexo. A duração de vida de ambos sexos foi afetada pelo extrato aquoso da planta: as fêmeas morreram mais nos estágios iniciais que os machos. As fêmeas sobreviventes da fase larval, criadas no meio com extrato, tiveram longevidade maior, o que pode ser atribuído como consequência do menor tempo de vida dos machos adultos criados no meio com o extrato da planta.

**PALAVRAS-CHAVE:** Medicina popular. Extrato de planta. Organismo-modelo. Inseticida botânico.

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**REFERENCES**

- BARNES, A. I.; WIGBY, S.; BOONE, J.M.; PARTRIDGE, L.; CHAPMAN, T. Feeding, fecundity and lifespan in female *Drosophila melanogaster*. **Proc. R. Soc. Lond. B.**, London, v. 275, p. 1675-1683, jul. 2008.
- CHOUHARY, S. Effect of nicotine (plant extract) on sex-ratio of *Drosophila melanogaster* (Meigen). **J. Environ. Biol.**, Muzaffarnagar, v. 24, p. 493-494, oct. 2003.
- GADANO, A.; GURNI, A.; LÓPEZ, P.; FERRARO, G.; CARBALLO, M. In vitro genotoxic evaluation of the medicinal plant *Chenopodium ambrosioides* L. **J. Ethnopharmacol.**, London, v. 81, p. 11-16, jun. 2002.
- GRAF, U.; KATZ, F. E.; FREI, A. J.; JUON, H.; HALL, C. B.; KALE, P. G. Somatic mutation and recombination test in *Drosophila melanogaster*. **Mutat. Res-Gen. Tox. En.**, London, v. 6, p. 153-188, jan. 1984.
- HO, Y. K.; CLIFFORD, A. J. Development and survival of *Drosophila melanogaster* fed a diet containing pyrimidine analogs. **Comp. Biochem. Physiol. B**, New York, v. 118, p. 167-172, sep. 1997.
- MORDUE, A. J.; NISBET, A. J. Azadirachtin from the neem tree *Azadirachata indica*: its action against insects. **An. Soc. Entomol. Brasil**, Londrina, v. 29, p. 615-632, dec. 2000.
- OLIVEIRA, A. P.; NEPOMUCENO, J. C. Avaliação dos efeitos genotóxicos e antígenotóxicos do avelós (*Euphorbia tirucalli*) em *Drosophila melanogaster*. **Bioscience J.**, Uberlândia, v. 20, p. 179-186, may/aug. 2004
- RUBIN, G. M. *Drosophila melanogaster* as an experimental organism. **Science**, Washington, v. 240, p. 1453-1459, jun. 1988.
- TAVARES, M. A. G.C; VENDRAMIN, J. D. Bioatividade da erva-de-santa-maria, *Chenopodium ambrosioides* L., sobre *Sitophilus zeamais* Mots. (Coleoptera: Curculionidae). **Neotrop. Entomol.**, Vacaria, v. 34, p. 319-323, mar./apr. 2005.
- ZIVANOV-CURILS, J. ; TOMIN, J.; BOJANIC, V.;BOJANIC, Z.; NAJMAN, S.; KATIC, K.; MRCARICA, E, DJINDJIC, B. Effect of chronic phenol intoxication on fertility and life span of *Drosophila melanogaster*. **Arch. Oncol.**, Sremska-Kamenica, v. 1, sup. 12, p. 17-18, apr. 2004.