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Effect of Laser Radiation on the Phenotypic Mutations of Drosophila melanogaster (Diptera:Drosophilidae)

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Article History	Abstract
Received: 11 Feb 2022 Revised: 26 May 2022 Accepted: 10 August 2022 CC License CC-BY-NC-SA 4.0	This study was conducted to find out the effect of 5 periods of exposure to laser rays (0,15,10,5,20) minutes on the phenotypic mutations of Drosophila melanogaster, as well as calculating the percentage of mortality and the percentage of larval failure. The results showed that laser beams had significant effects in causing mutations, especially in the 15-minute period, which amounted to 0.33 and thus formed significant differences compared to the control group. The periods of exposure to laser rays also caused clear effects in the rate of larval mortality, as the death rate was 100% for the period of 20 minutes, while this rate decreased to 27% and 34% for the periods 5 and 10 minutes, respectively, while the lowest failure rate was 0% at the period 20, which led to the death of all the larvae. <i>key wards: - drosophila, laser radiation, mortality percentage, phenotypic mutation.</i>

Introduction

A mutation is defined as any sudden change that occurs in the quantity or quality of genetic material due to natural factors and others that were manufactured by man, and the sum of these factors is called mutagen, although some mutations occur automatically, but geneticists used the intensification of mutations in order to obtain a second type. These are the induced mutations, after treating the organism with one of the well-known mutagens (Wolf & Reddy, 1993; Oladosu et al., 2016).

Induced mutations may occur as a result of physical or chemical factors from physical factors such as radiation of all kinds. As for chemical mutants, for example, food and pesticides used to eliminate types of living organisms and medicines. These mutants vary in intensity of exposure and fall between acute exposure or chronic exposure. Acute exposure may create direct changes or effects on the organism, unlike chronic exposure, whose symptoms do not appear until after a long period of time (Organisms et al., 2021).

It has used radiation in practical applications and has achieved impressive successes that facilitated its entry into the field of industry, medicine, agriculture and scientific research. It entered the military field and was present in nuclear reactors. Because of the modern use of radiation, it has become today a serious challenge to the vital environment until it has become a matter of concern to man and a threat to his life on the globe. This prompted researchers all over the world to submit studies and scientific research in which they talk about the dangers of radiation and its negative effects on humans and living creatures (Ojovan et al., 2019).

Those concerned with genetics relied in their studies permanently on organisms that were subjected to natural or created mutations. They used them in the early genetic study, which was represented by the morphological changes that can be seen on the organism, such as the shape of the wings, the color of the eyes, and the antennae in Drosophila fly. One of the types specialized in mutations (Hales et al., 2015; Meneely, 2020).

Drosophila is one of the most widely used species in causing mutations, as it is typical and most suitable for genetic experiments for several advantages mentioned previously, and this fly was used for the purpose of studying human diseases, where scientists noted that there is a clear similarity in the process of crossing over chromosomes, and there is a clear similarity In the genetic material of humans and Drosophila, many studies have been presented on Drosophila (Charlesworth, 2015). Where he (Zhikrevetskaya et al., 2015) created genetic mutations when exposing the insect to doses of ionizing rays of the type of gamma. Shaffer & Wuller,(1994) have studied the evolution and competition between different Drosophila species, and several studies have been presented on some sexual mutations of the insect.

In this study, laser beams were used to create mutations in Drosophila to study the extent of the effect of these rays. The laser is an amplification of light that stimulates radiation, and the laser is a source for producing visible or invisible light energy and has specifications that are specific to it without the rest of the types of light produced from other sources, whether natural or Industrial (Hrabina et al., 2003). There are wide applications for the use of laser in the fields of medicine, industry, biological research and agriculture (Abu- Elsaoud, 2013). Studies have shown that laser has clear effects on cell components of proteins, amino acids, and the rate of intracellular enzymes (Perveen et al, 2012).

The laser has the ability to kill microorganisms and fungi because of its thermal effects due to the absorption of these rays by water molecules, as water constitutes the high percentage of the components of the living cell, and the laser is sent in the visible and ultraviolet regions, so its effect here is a chemical effect because the photons that make up visible and ultraviolet rays It has the ability to cause a chemical reaction to the molecules of living matter, and these rays cause inhibition of the components of the living cell due to a genetic change in the DNA of the living cell, even if it is for small doses, unlike infrared rays. The effect on it is thermal (physical) because the energy of the photons in it is low. A field for chemical reaction events (Wang et al., 2017; Tuchin et al., 2022).

The interaction of laser with living tissues provides important information about what are the characteristics of living tissues and their biological behaviors, and that the interactions of laser and living matter are very important, as it was used several decades ago as a blood irradiation technique, and the method was important to stimulate living cells (Jawad et al., 2011). Moreover, the phenotypic traits are highly affected by external conditions, and these effects are transmitted through genes to future generations, so this study aimed to know the effect of laser rays on causing phenotypic mutations, as well as the effect of these rays on the rate of death of Drosophila larvae and the rate of formation of pupae (Zadeh et al., 2022.

Materials and Methods

Collection and Identification of Drosophila Melanogaster

Samples of adult Drosophila insects were collected from Al-Diwaniyah governorate using the (Shaffer et al., 1994) method by placing traps consisting of glass containers 7 cm high and 2.5 cm in diameter containing fermented fruits (apples - bananas - oranges) for the purpose of attracting insects after leaving them for four Continuous days, then closed the mouth of the traps with pieces of thin cloth to prevent insects from flying and to give adequate ventilation to the insect. Insects were transferred to the laboratory and placed in the refrigerator for ten minutes to immobilize them, then they were transferred to prepared media for insect rearing. The insect was identified by the taxonomic key of (O'Grady & Markow, 2009) of Drosophila fly as Drosophila melanogaster.

Preparing the breeding media for raising the Drosophila

The medium used for Drosophila fly cultivation was prepared by (Riechmann et al., 1997) using laboratory prepared medium consisting of (2 g of agar, 10 g of sugar, 10 g of bread yeast, 10 g of

maize flour and 100 ml of distilled water), these materials were mixed well, then boiled for 5-10 minutes for the purpose of sterilization. This mixture was poured into tubes prepared for breeding, then the nozzles of the tubes were covered with cotton, then the tubes were transferred with the media for sterilization by an Autoclave device. After the sterilization process was completed, the yeast suspension was added to the medium. The prepared live snake was prepared by dissolving (10 g of yeast in 100 ml of distilled water) in the form of drops and leaving the medium for 48 hours before the insects were transferred to it. A pair of insects (male, female) was placed in the medium prepared for breeding. The insects were left for five days until the eggs were laid, which hatched to give larvae, then pupae, then adults, and the larvae were reared for many generations in cages.

A group of Drosophila adults were collected in Petri dishes containing the medium prepared for this purpose, and were kept in those dishes for the purpose of obtaining larvae not exceeding 24 hours old, and then placed in incubators and constant environmental conditions of light, heat and humidity for the purpose of obtaining larvae As a result of breeding for several generations, the larvae were divided into five groups, and after obtaining the larvae, they were placed in 250 ml glass bottles at a rate of 20 larvae in each bottle and at a rate of five bottles, and they were exposed to laser beams for periods of time (0, 5, 10, 15, 20) minutes and we denote the time periods with the symbols (T20, T15, T10, T5, T0) respectively. After the irradiation process was completed, the larvae were transferred to the same incubator with the same environmental conditions. As for the comparison treatment, the larvae were left without irradiation and placed after isolation in the optimal conditions for the insect's life. The percentage of mortality was calculated according to the Schneider and Orell Formula equation (Al-Ameri et al., 2020; Al-Ameri and Alhasan, 2022; Qasim et al., 2022).

Statistical analysis

The results were statistically analyzed by using the statistical analysis program (SPSS-V18), and using the Anova table, then they were compared according to the LSD test at a probability of P \leq 0.05 (Al-Rawi & Abdulaziz, 2000; Al-Ameri and Alhasan, 2020).

Results and Discussion

Different deformations appeared for emerging adults from irradiated larvae, and the deformations varied between (small insects, body curvature, and a very clear reduction of antennae and wings), the highest percentage of deformations was at the T20 period, and the highest percentage of adults not coming out of the shed skin was 0% at the T20 period. The reason for these abnormalities is due to the effect of radiation on somatic cells and the occurrence of mutations in chromosomes and dominant lethal mutations (Al-Baldawy et al., 2020; Alhasan and Hussein, 2022). The highest mortality rate appeared in the period T20, where there were no insects that reached the adult stage, while the period T0 was the standard sample, as it did not witness any deformations or morphological mutations, and the highest percentage of reaching the adult stage was recorded. The percentage of emergence of adults to the treatment T5 = 73%, and a number of phenotypic mutations appeared, such as (body curvature of some of them and reduction of antennae), while the T15 group recorded 56% arrival of the adult stage, and the mutations were very clear, all of them appeared (reduction of antennae and some of them showed curvature of the body). As for the T 20 group, it showed the lowest eruption rate. The access rate for the adult females was 0%.

It was also noted that the rate of larval mortality is directly proportional to the increase in the period of exposure to radiation, and the reason may be due to the effect of radiation on somatic cells and mutations that led to the death of larvae (Al-Ameri et al., 2020; Hussain et al., 1994). Or because of the effect of radiation on the cytoplasm of cells and then the death of these cells or damage to the tissues lining the middle intestine, which leads to the prevention of their renewal from time to time, or perhaps the radiation led to a decrease in the resistance of the larvae to microorganisms and diseases, which leads to their rapid death (Al-Bahadli, 2002; Al-Ameri and Alhasan, 2020; Qasim et al.,2022;). The reason for this change in the shape of the body and the reduction of the antennae is due to the fact that the larval stage is the stage of preparation for the entry of the insect to the adult stage. Any influence during the process of division leads to the occurrence of many disorders in cell division, and the reason for this is due to influential structural changes in chromosomes or in protein molecules in the nuclei of cells or in the cytoplasm, and this leads to preventing the development of insects exposed to radiation (Ahmed, 1998; Nijhout et al., 2014; Hamzah, 2018). It is possible that when exposure to

radiation affects the somatic cells while they are in a state of division, which leads to effects on the body systems while they are in the process of development, and that these radiations caused damage to cells because the larval stage is sensitive to stimuli, and explained (Al-Tamah, 2006; Kadhem and Hamzah, 2020) the reason for the appearance of deformations In adults that were produced from a larval stage exposed to radiation, if any imbalance resulted in the genetics of the somatic cells prepared for division and responsible for the growth and integration of the insect.

Treatment	The proportion of normal adults	Percentage of deformed adults
	Mean±SD	Mean±SD
T ⁰	0.90 ±0.11	0.00 ± 0.0
T ⁵	0.74 ±0.12	0.21 ± 0.01
T ¹⁰	$0.48 \pm 0.0.52$	0.28±0.25
T ¹⁵	0.41 ±0.22	0.33±0.34
T ²⁰	0.00	0.00

Table (2). Percentage of larvae of the treated Drosophila flyExposure periodspupa phase ratio (%) T^0 91 T^5 73 T^{10} 64 T^{15} 56 T^{20} 00

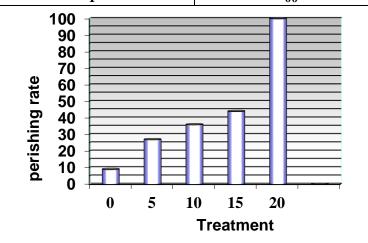


Figure (1). Average mortality rate of Drosophila fly larvae

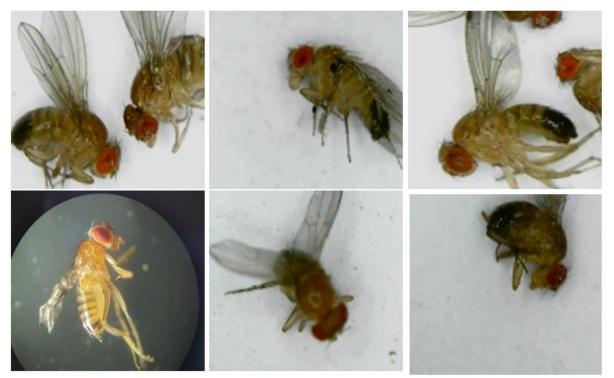


Figure (2). Effect of laser radiation on the phenotypic mutations of Drosophila melanogaster

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