

Morphological and histological effects of gamma radiation on the female reproductive system of cowpea beetle, *Callosobruchus maculatus* (F.)

Habeba Mohamed Omar Abuo El-Enine

Plant Protection Dept. Faculty of Agric., Zagazig Univ., Zagazig, Egypt.

Corresponding author: habebaumar1977@gmail.com

Abstract

This study was carried out under constant conditions at $30\pm 1^\circ\text{C}$ and $65\pm 5\%$ RH. in breeding room of Plant Protection Department, Fac. Agric., Zagazig Univ. The aim is to study the effect of gamma radiation producing from (Cs-137) at doses of (1-5 Gy) on the morphology and histology of female reproductive system of cowpea beetle, *Callosobruchus maculatus* (F.) and their malformations. The obtained data are summarized as follows: The normal female reproductive system was containing two ovaries each consist six ovarioles, bursa copulatrix, two Calyx, two lateral oviducts, spermatheca, spermathecal gland and vagina. The irradiated females ovaries from 1-day old eggs were consists of bursa appear, bursa enlarged, ovarioles reduced, no calyx (empty) with 1, 2, 3 and 5 Gy. Moreover, the irradiated female ovaries form 3-days old eggs were consists of bursa appear, ovarioles reduced and calyx without mature eggs with 1, 2, 3, 5 and 10 Gy.

Key words: Cowpea beetle, *Callosobruchus maculatus* (F.), gamma radiation, female reproductive system, ovaries, morphological and histological effects.

Introduction

Cowpea (*Vigna unguiculata* (L.) Walp.) is one of the most popular leguminous crops used as food in Egypt. This crop is susceptible to infested by several insect pests. Cowpea beetle, *Callosobruchus maculatus* (F.) is one of the most damaging pests of cowpea (Decella, 1981; Jackai and Daoust, 1986). Nearly all the species of leguminous seeds are also attacked by *C. maculatus*. This insect pest causes weight loss, decrease germination potential and diminishes the market value of the crop. It causes substantial quantitative and qualitative losses manifested by seed perforation, and reductions in weight, market value and germ inability of seeds (Sekou *et al.*, 2001). Insects not only consume grain, but also contaminate it with their metabolic products, body parts, cast skin, excrement and fragments of immature insects (Smith *et al.*, 1971 and Pedersen, 1994).

Gamma irradiation is an effective disinfestations treatment for stored product pests. Disinfestation by irradiation is a promising method that has been studied extensively in many countries (Ahmed *et al.*, 1977; Begum *et al.* 1980). In addition, gamma radiation has been suggested as a mean of controlling insects in grain and other stored commodities. The degree of damage due to radiation in the reproductive system of pests depends on the age of female and the radiation dose. Morphological and histological effects of gamma radiation on the ovarioles of different insect species were observed. After gamma radiation, the ovarioles of *C. maculatus* were reduced in size and the ovary appeared as an indifferentiated mass (Ahmed *et al.*, 1976).

So, the present work was carried out to study the morphological and histological effects of gamma

radiation on female reproductive system of cowpea beetle, *C. maculatus*.

Materials and Methods

This study was carried out in Plant Protection Department, Faculty of Agriculture, Zagazig University under cooling incubator temperatures. Meanwhile, radiation experiments were conducted at The National Center for Radiation Research and Technology (NCRRT), Atomic Energy Authority, Nasr City Cairo, Egypt.

1. Rearing technique of the tested insect:

Stock culture of cowpea beetle, *Callosobruchus maculatus* (F.), was obtained from the infested cowpea seeds, *Vigna unguiculata* (L.). The cultures of insect were reared on cowpea seeds for many generations under laboratory conditions at $30\pm 1^\circ\text{C}$ and $65\pm 5\%$ R.H.

2. Radiation Technique:

The source of radiation was Cs-137 irradiator with a dose rate of 0.723 rad/sec. all radiation experiments were conducted using the gamma irradiator (Gamma Cell -40, Cesium-137 irradiation unit, Atomic Energy of Canada limited commercial products) located at The National Center for Radiation Research and Technology (NCRRT), Atomic Energy Authority, Nasr City Cairo, Egypt.

3- Morphological and histological studies on the female reproductive system:

After emergence of parent generation, three females seem to be as normal which produced from each 1 and 3-days old eggs treated with different doses (0, 1, 2, 3, 5 and 10 Gy of gamma radiation) were

dissected. The reproductive system was photographed and morphology of each was conducted under dissecting microscope and described. The reproductive system then stained by methylene blue and fixed in aqueous Bouin's solution (picric acid, saturated aqueous 75 ml + formalin 25 ml + glacial acetic acid 5 ml) for 24 h. (Gurry, 1962), then transferred to ascending dilutions of ethyl alcohol (50 %, 70 %, 80 %, 90 %, 100 %) and then in xylene (50-100 %). The specimens were then embedded in paraffin wax. Many sections were obtained and stained with haematoxylin and eosin stains, according to the method of (Drury and Wallington (1980). Serial longitudinal sections (5 μ) of each specimen were made (Mc Manus and Mowry, 1960) and covered with a glass cover slip (Kamelia, 1983). Stained sections were examined. (Hedaya, 1990).

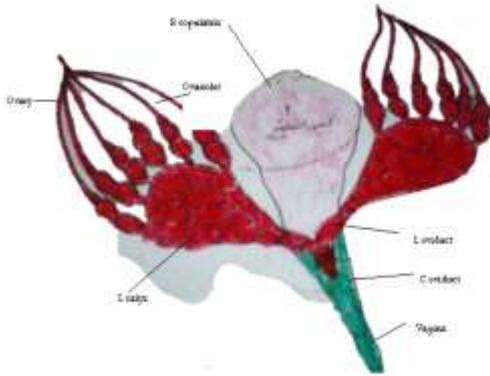
Results and Discussion

Morphological and histological finding of ovaries of cowpea beetle female, *Callosobrochus maculatus* (F.):

The normal female reproductive system was containing two ovaries each of them consist six ovarioles, bursa copulatrix, two calyx, two lateral oviducts, spermatheca, spermathecal gland and vagina (Fig. 1 and 2). While, the irradiated females ovaries form 1-day old eggs were consists of bursa appear, bursa enlarged, ovarioles reduced, no calyx (empty) with 1,2,3 and 5 Gy as shown in Fig. (3, 4, 5 and 6). Moreover, the irradiated females ovaries form 3-days old eggs were consists of bursa appear, ovarioles reduced, calyx without mature eggs, no calyx, bursa exist enlarged, no ovarioles appeared with 1,2,3,5 and 10 Gy as shown in Fig. (7, 8, 9, 10 and 11).

1. Female reproductive system from normal eggs

A: Normal of female reproductive system in Cowpea weevil (After Amir and El-Deeb, 1987)



B: Normal dissected female to appear female reproductive system of *Callosobrochus maculatus* (B: Bursa copulatrix Ovar: ovary Ovariol (6 in each), Calyx, Oviduct, vagina)



Fig. (1): Normal female reproductive system of cowpea beetle, *Callosobrochus maculatus* (F.). A: drawing, B: dissected.

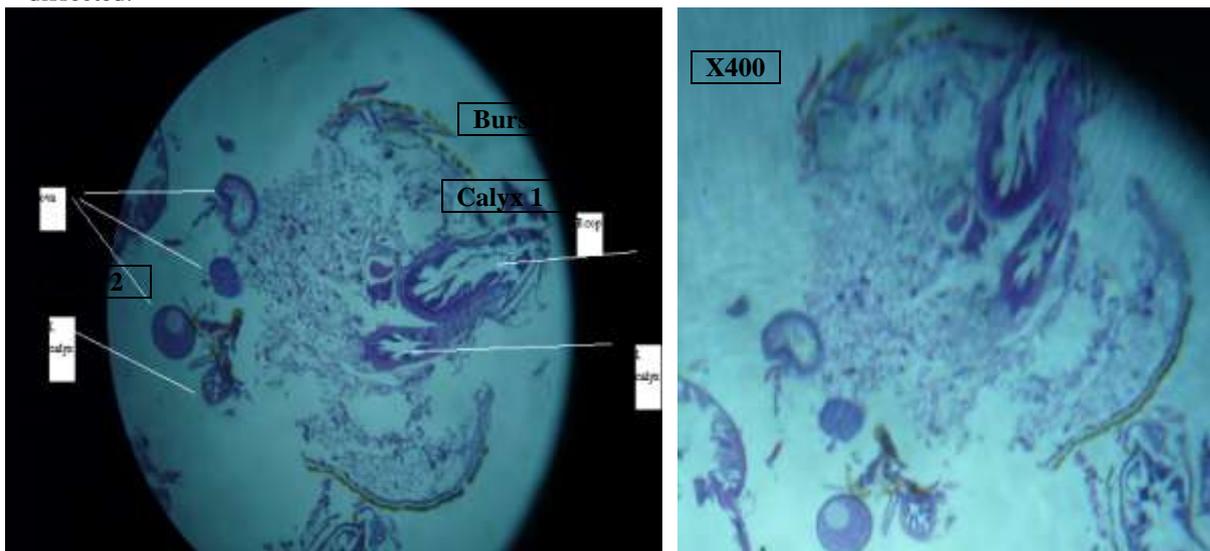


Fig. (2): Showed histology of normal adult female reproductive system of cowpea beetle.

2. Female reproductive system from irradiated eggs

2.1. Females produced from irradiated 1 day old eggs:

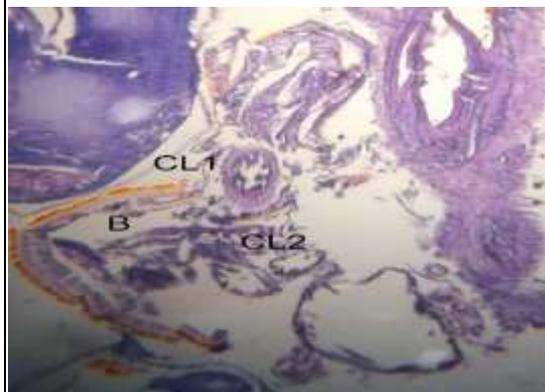
2.1.1. 1Gy of gamma radiation



Females ovaries dissected after treated eggs 1 day old with 1Gy (Bursa appear, ovarioles reduced, no calyx).



Females ovaries dissected after treated eggs 1 day old with 1 Gy (Bursa exist no ovarioles appeared).



(X400)
Calyx and Bursa empty, ovarioles reduced



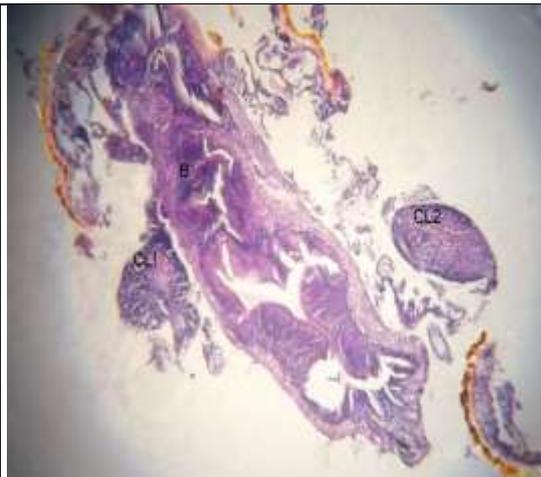
(X400)
Calyx and Bursa empty ovarioles reduced

Fig. (3): Female reproductive system of cowpea beetle, *Callosobrochus maculatus* (F.) arised from 1 day old eggs irradiated with 1 Gy.

2.1.2. 2Gy of gamma radiation



Dissected females after treated eggs 1 day old with 2Gy (Bursa appear, ovarioles reduced, no calyx).



(X400) Enlarged bursa, Calyx empty, no ovarioles

Fig. (4): Female reproductive system of cowpea beetle, *Callosobrochus maculatus* (F.) arised from 1 day old eggs irradiated with 2 Gy.

2.1.3. 3Gy of gamma radiation

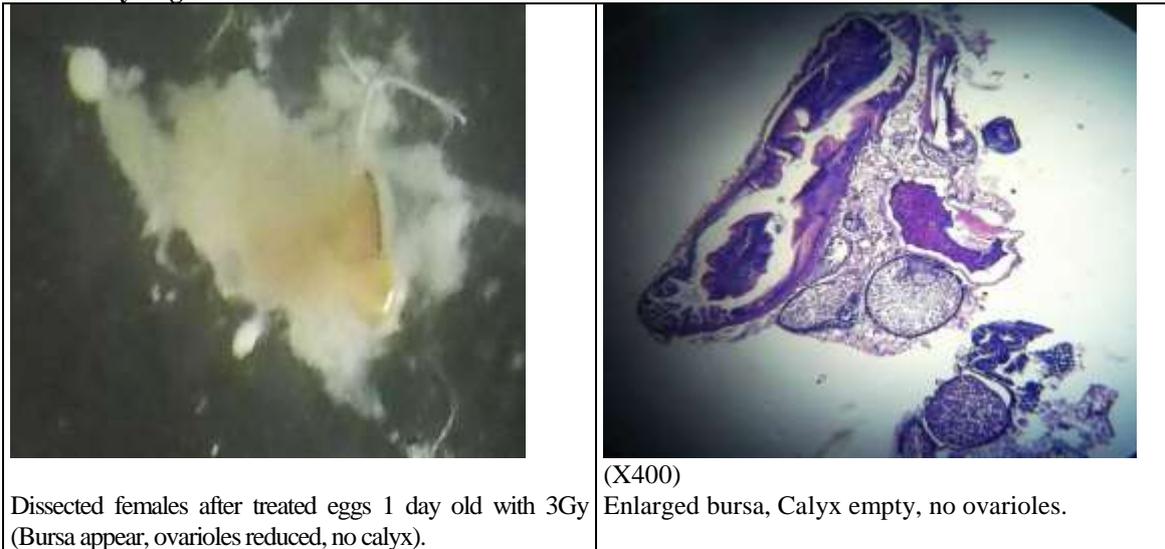


Fig. (5): Female reproductive system of cowpea beetle, *Callosobrochus maculatus* (F.) arised from 1 day old eggs irradiated with 3 Gy.

2.1.4. 5 Gy of gamma radiation

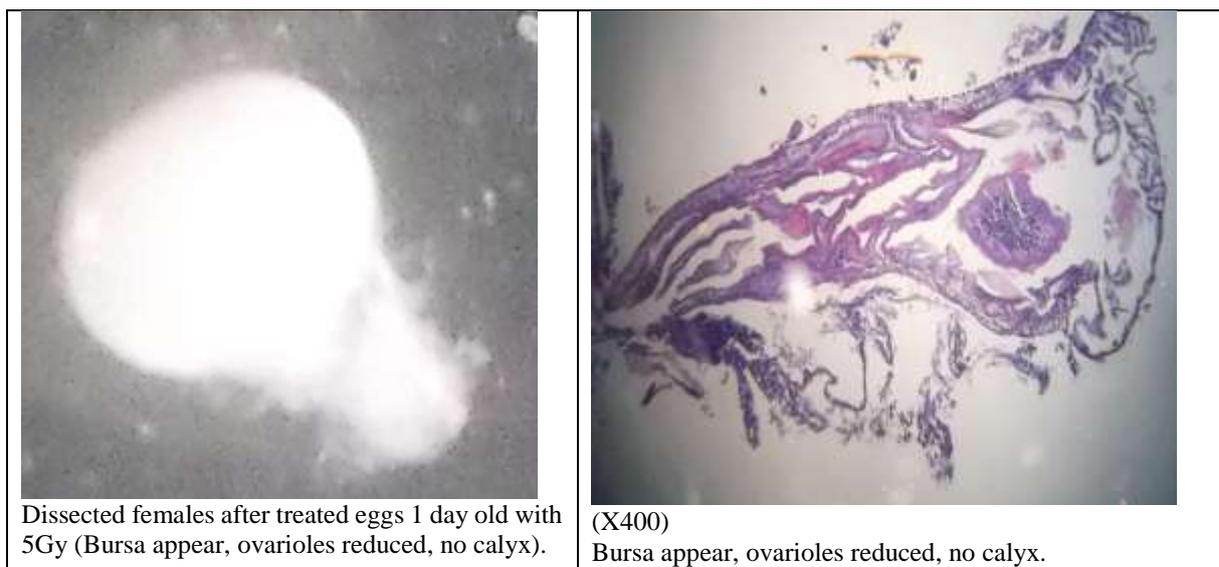


Fig. (6): Female reproductive system of cowpea beetle, *Callosobrochus maculatus* (F.) arised from 1 day old eggs irradiated with 5Gy.

2.2. Females produced from irradiated 3 days old eggs:

2.2.1. 1Gy of gamma radiation



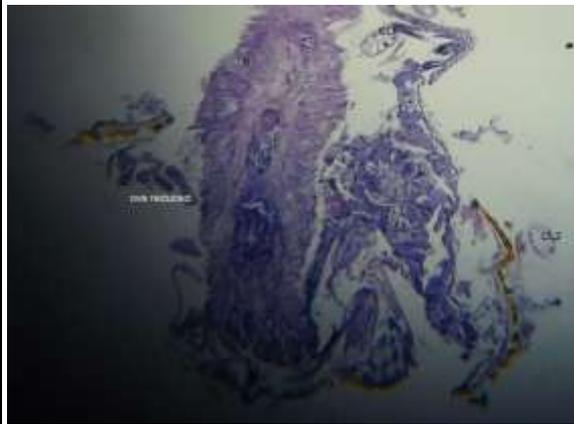
Dissected females after treated eggs 3 day old with 1 Gy (calyx without mature eggs).



Dissected females after treated eggs 3 day old with 1Gy (Bursa appear, ovarioles reduced, no calyx).



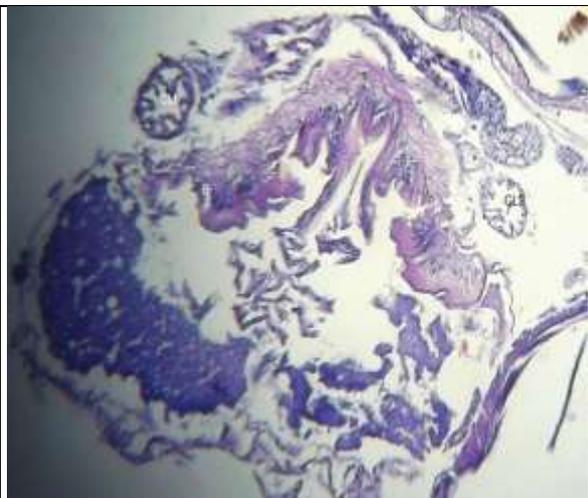
(X400)
Calyx without mature eggs



(X400)
Bursa appear, ovarioles reduced, no calyx.

Fig. (7): Female reproductive system of cowpea beetle, *Callosobrochus maculatus* (F.) arised from 3-days old eggs irradiated with 1Gy.

2.2.2. 2Gy of gamma radiation



(X400)

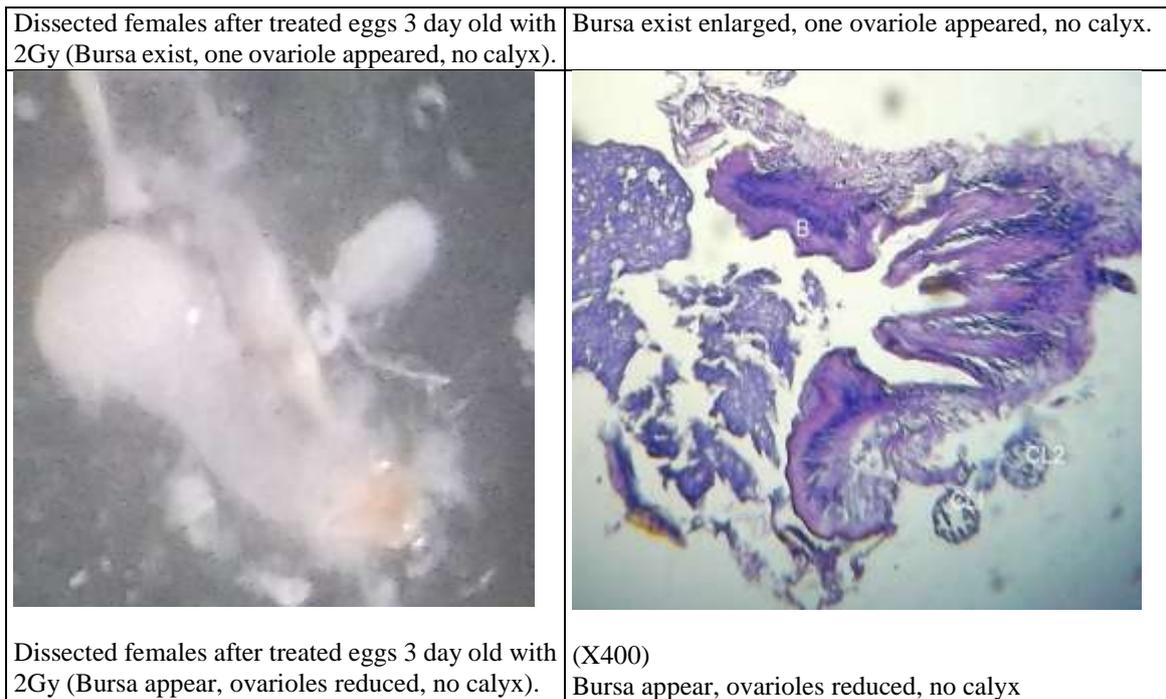


Fig. (8): Female reproductive system of cowpea beetle, *Callosobrochus maculatus* (F.) arised from 3-days old eggs irradiated with 2Gy.

2.2.3. 3Gy of gamma radiation



Fig. (9): Female reproductive system of cowpea beetle, *Callosobrochus maculatus* (F.) arised from 3-days old eggs irradiated with 3Gy.

2.2.4. 5Gy of gamma radiation

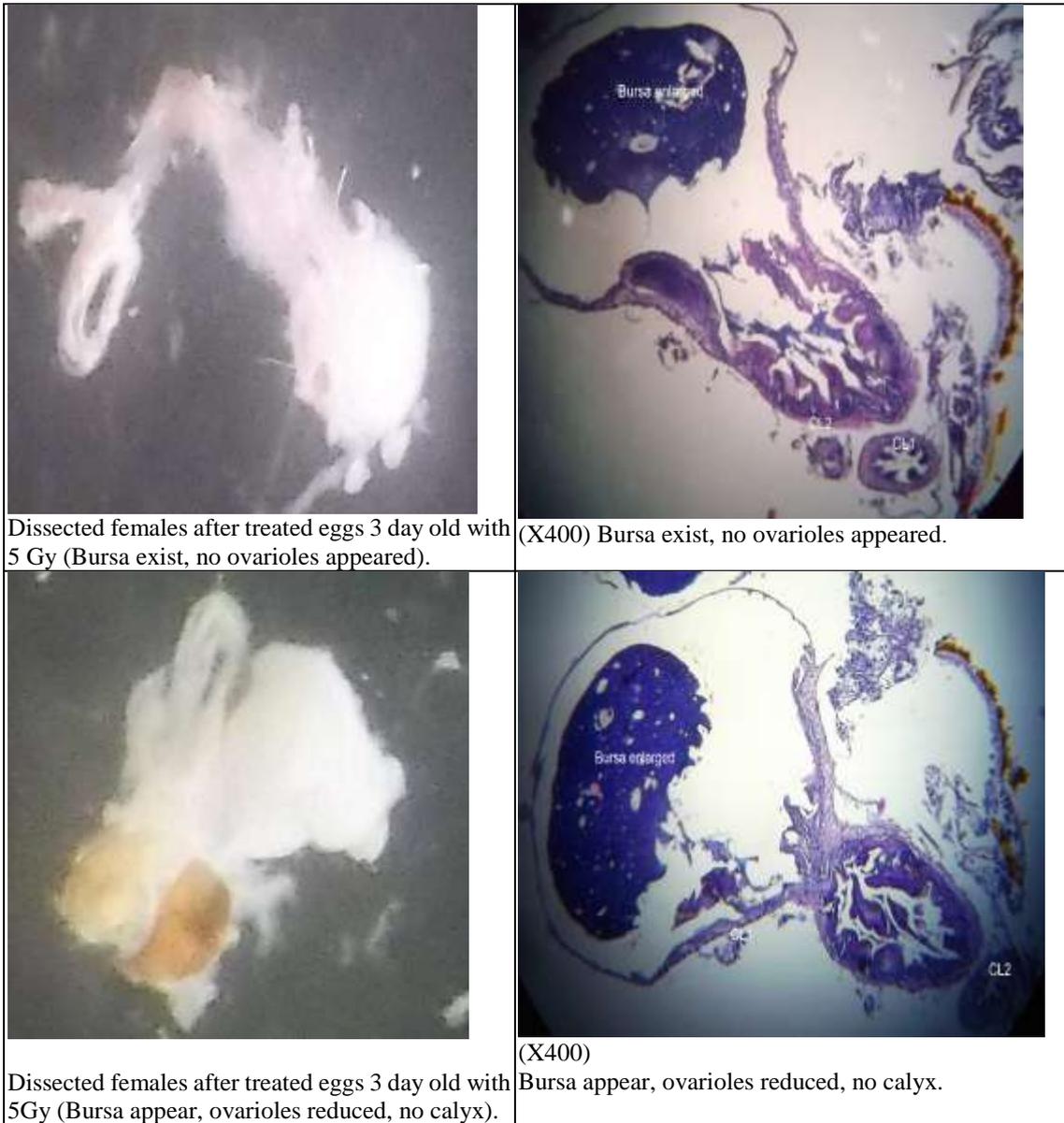


Fig. (10): Female reproductive system of cowpea beetle, *Callosobrochus maculatus* (F.) arised from 3-days old eggs irradiated with 5 Gy.

2.2.5. 10 Gy of gamma radiation



Fig. (11): Female reproductive system of cowpea beetle, *Callosobruchus maculatus* (F.) arised from 3-days old eggs irradiated with 10Gy.

These results are in agreement with those: **Amir and El-Deeb (1987)**, **Hedaya (1990)** who found that the female reproductive system of *C. maculatus*, has two ovaries each consisting of six telotrophic ovarioles. The cowpea beetle, *C. maculatus* has a telotrophic ovariole. Its organization is similar to that of *C. analis* and *C. chinensis* described by **Aggarwal (1967)** and **Tikku et al. (1978)**. The Colorado beetle, *Leptinotarsa decemlineata* has also a pseudotelotrophic ovariole described by **De Loof et al. (1972)**. They also, noticed resemblance in the germarial cells so that the oogonia and trophocyte cannot be differentiated. **Aggarwal (1967)** described that germarium cells as nurse cells, the nucleus is voluminous and spherical.

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التأثيرات المورفولوجية والهستولوجية لأشعة جاما على الجهاز التناسلي لأنثى خنفساء اللوبيا
Callosobruchus maculatus (F.)

حبيبة محمد عمر أبوالعنين

قسم وقاية النبات - كلية الزراعة - جامعة الزقازيق - مصر

أجريت هذه الدراسة تحت الظروف المعملية الثابتة ($30 \pm 1^\circ\text{C}$ ، $65 \pm 5\%$ رطوبة نسبية) بحجرة التربية- قسم وقاية النبات- كلية الزراعة- جامعة الزقازيق. بهدف دراسة تأثير أشعة جاما الناتجة من السيزيوم ^{137}Cs بجرعات اشعاعية من (1-5 جراى) على مورفولوجى وهستولوجى الجهاز التناسلى لأنثى خنفساء اللوبيا والتشوهات الناتجة عنها. ولخصت النتائج كالتالى: وجد أن الجهاز التناسلى فى الأنثى يتكون من مبيضين (6 فروع لكل مبيض)، كيس التلقيح، الكالكس (2)، قناتى بيض جانبيين، المهبل، القابلة المنوية وغدة القابلة المنوية. وجد أن الاناث الناتجة من البيض المعامل عمر 1 يوم حدث اختزال للفروع المبيضية، كيس التلقيح استطال بشكل واضح، الكالكس قد يظهر ولا يوجد به بويضات (فارغ) وذلك فى الجرعات (1، 2، 3، 5 جراى). بينما فى حالة الاناث الناتجة من البيض المعامل عمر 3 يوم يتكون المبيض من كيس تلقيح ظاهر، اختزال فى الفروع المبيضية، الكالكس (بدون بيض ناضج) وذلك للجرعات (1، 2، 3، 5، 10 جراى).