

IMPACT OF GAMMA IRRADIATED HOSTS ON TOTAL LIPIDS AND FATTY ACID COMPOSITION OF KHAPRA BEETLE, *Trogoderma granarium* (EVERTS).

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ABSTRACT

Chemical analysis of total lipids and fatty acid composition of *Trogoderma granarium* was carried out to envisage the effect of irradiated hosts diet on these constituents. Total body lipids were affected indirectly by feeding of larvae on the irradiated diet. The results showed that the fatty acids changed according to the host. There were qualitative and quantitative differences in the individual fatty acid composition between male and female.

Radiation was effective in changing the saturated and unsaturated fatty acid concentrations especially for 18 : 2 and 18 : 3 which are essential for normal growth, development and adult emergence.

In general, it could be concluded that fatty acid composition affected indirectly by feeding of larvae on the irradiated hosts diet, and consequently affect the biological activity of insects.

INTRODUCTION

Khapra beetle *Trogoderma granarium* larvae are known to feed on several different hosts.

The common method that is widely applied for disinfestations is by chemical insecticides. The repeated use of fumigants might involve a serious health hazard to human beings and environment. Therefore, irradiation looks more effective, safe and cheaper (Ahmed *et al.*, 1993, Souka and El-Degwi, 1994, El-Degwi, 1997).

Lipids are important constituents of membranes, play a key role in insect energy metabolism and comprise some of the most important control factors in the insect growth and morphogenesis. Also, insect growth hormones, pheromones and sex attractants are lipoidal in nature (Gibert, 1967). The lipids found in insects are usually common to all higher organisms.

Fatty acids of insect lipids are derived in part from dietary sources and in part from biosynthetic pathway similar to those operative in higher animals (Worthing and Payne, 1974). In general diet influences the synthesis of fatty acids in several species (Turunen, 1983, Subramanyam and Culkowp, 1987, Afify *et al.*, 1997).

Data concerning the effect of ionizing radiation on the composition metabolism of insect lipids are scarce (Afify *et al.*, 1995, El-Orabi and Gareib, 2000, Haiba and Mohamed, 2001). Therefore this study was conducted to determine the effect of the larvae irradiated and non irradiated diet upon the total lipids and fatty acid composition of khapra beetle adults.

MATERIALS AND METHODS

Stock cultures and irradiation treatment:

The strain of khapra beetle *Trogoderma granarium* (EVERTS) has been reared in the Insect and Pest Control lab (NCRRT). Larvae were reared in half pound jar half filled with mixture of whole and crushed host seeds, wheat (*Triticum vulgare*), cowpea (*vigna unguiculata*) and peanut (*Arachis hypogaea*).

Irradiation was conducted by Gamma Cell 220 Irradiation Unit (Co source) located at the (NCRRT) with a dose rate of 2 : 1 rad/sec. Host seeds were irradiated with 100 and 300 Gy, and their respective control. The cultures were kept at $30 \pm 1^\circ\text{C}$ and $70\% \pm 5\%$ R.H. Insects were removed as pupae then sexed and held in (9cm. diameter), petri dishes lined with filter paper. Adults of the first generation were collected from each culture.

Extraction of insects' lipids:

Total crud lipids were extracted from 1gm. Of body weight of freshly adult male and female of *T.granarium* with chloroform: methanol (2:1 v/v) mixture, using soxhlet apparatus (AOAC, 2000). The combined filtrates were evaporated to dryness, extracted with petroleum ether, dried and weighted.

Separation of fatty acids :

Total lipids of insects were saponified with methanolic KOH (20%) for 24 hrs at room temperature. The unsaponified matter was extracted several times with petroleum ether at $40-60^\circ\text{C}$ and removed. Fatty acids were freed from their alkaline salts by acidification with HCL and then extracted with petroleum ether and methylated with diazomethane (Vogel, 1980).

Gas chromatographic analysis of Fatty acids :

Fatty acids were identified by gas liquid chromatographic (GLC) technique. GLC (Unicam model, UK) was used to characterize the fatty acids based on authentic fatty acids. Chromosorb R packed column (1.5m x 4mm) was used to separate fatty acids. GLC equipped with dual flame ionization detector. The temperature was programmed from 60°C / min for 250°C at a rate of $12^\circ\text{C}/\text{min}$ then held at 250°C for 15 min (Hamilton *et al.*, 1992).

RESULTS AND DISCUSSION

The effect of gamma irradiated and non irradiated diet on the percentage of total lipids in females and males of *T.granarium* is shown in Table (1) No appreciable change in the total lipid contents was noticed in males reared (as a larvae) on non-irradiated and irradiated wheat with 300 Gy (49.5 % and 49.7%) while , lipid content was reduced when wheat irradiated with 100 Gy (37.9%). Total lipid content of males fed on peanuts followed the similar trend. There was highly reduced of total lipid content in males resulted from the irradiated cowpea.

Females fed on irradiated wheat with 300 Gy showed relatively increase of lipid content compared to the non-irradiated ones, when females

were fed on cowpea and peanuts irradiated with 300 Gy. In contrary (23.8% and 38.2%, respectively) when compared with the control (36.5% and 43.2%). Female showed higher lipid contents than males except in case of irradiated peanut with 300 Gy were males contained higher total lipid than females, (45.5%). Generally, total body lipids were affected indirectly by feeding of larvae on the irradiated diet.

Table (1) : Effect of gamma irradiated and non – irradiated host diet on total lipids on *T. granarium*.

Host Diet	Dose in Gy.	Total lipids (%)					
		Control		100		300	
		male	female	male	female	male	Female
Wheat		49.5	51.3	37.9	52.5	49.7	58.6
Cowpea		27.2	36.5	21.5	39.4	10.5	23.8
Peanut		41.6	43.2	38.5	46.5	45.5	38.2

Data presented in Table (2) show the effect of irradiated and non-irradiated hosts diet on the relative percentages of fatty acids in males. The fatty acid 22 : 0 represent the major one when males were fed as a larvae on non-irradiated wheat (26.93%) followed by 18 : 1. It is worthy to note that this major one 22:0 decreased heavily (3.44%) at 100 Gy and disappeared at 300 Gy treatment while the concentration of 18:1 increased (42.14%) at 100 Gy and decreased to (15.49%) by increasing the irradiation dose. 24 : 0 was in the contrary with this trend. The fatty acid 23 : 0 was found in males reared on wheat irradiated with 300Gy (26.30%). Saturated C16 increased (21.24%) at 100 Gy comparing with control.

Table (2) : Effect of gamma – irradiated and non – irradiated host diet on relative percentages of fatty acid composition of *T. granarium* males.

Fatty acid	Dose in Gy.	host diet								
		wheat			cowpea			peanuts		
		Control	100	300	control	100	300	control	100	300
Myristic	14 : 0	—	—	—	—	—	5.82	—	—	—
Palmitic	16 : 0	11.78	21.24	8.30	—	18.81	30.55	20.18	14.37	34.94
Palmitic	16 : 1	—	2.06	—	—	1.58	—	—	—	—
Stearic	18 : 0	—	4.68	—	—	7.48	28.12	—	6.59	—
Oleic	18 : 1	26.70	42.14	15.49	53.27	54.8	27.38	33.94	51.50	47.88
Linoleic	18 : 2	4.98	16.04	9.81	14.65	11.71	8.31	13.79	20.66	17.18
Linolenic	18 : 3	—	2.50	—	—	1.33	—	—	—	—
Arachidic	20 : 0	—	—	—	—	1.15	—	—	—	—
Behenic	21 : 0	17.75	—	—	—	2.97	—	—	—	—
Docosanoic	22 : 0	26.93	3.44	—	—	0.81	—	—	6.89	—
Tricosanoic	23 : 0	—	—	26.30	—	—	—	18.35	—	—
Tetracosanoic	24 : 0	11.95	7.85	40.08	28.05	—	—	13.76	—	—
Unsaturated		31.68	62.74	25.30	67.92	68.8	35.51	47.73	72.16	65.06
Saturated		68.32	37.26	74.70	32.08	31.20	64.49	52.27	27.84	34.94
Unsaturated/saturated		46.4	168.4	33.9	211.7	220.5	58.2	91.3	260.0	186.2

Three fatty acids were found in males fed as a larvae on non irradiated cowpea, it is very important to note that the most abundant one was 18 : 1 (53.27%). Nine fatty acids detected in while, males fed on irradiated host with 100 Gy. 18 : 1 was the most dominant (54.18%) and decreased to (27.38%) with 300 Gy. Saturated fatty acid 16 : 0 was found in males (18.8%) and increased with 300 Gy (30.55%), also 18: 0 followed the similar trend. In addition the irradiation dose effectively in reduced 18 : 2 and 24 : 0 was disappeared.

The fatty 18 : 1 was the major component among the total fatty acids of males fed on peanuts (33.94%) and increased to (51.50% and 47.88%) in case of irradiated seeds with 100 and 300 Gy. The same trend was observed with 18:2, while 16 : 0 decreased in males with 100 Gy (14.37%) and increased with 300 Gy (34.94%), The fatty acids 23 : 0 and 24 : 0 were presented in males and disappeared when host diet irradiated. The fatty acids 18 : 0 and 22 : 0 detected in males with 100 Gy.

The composition of fatty acids of the adult females reared as a larvae on different irradiated and non-irradiated hosts diet are shown in Table (3).

Table (3) : Effect of gamma – irradiated and non – irradiated host diet on relative percentages of fatty acid composition of *T. granarum* females.

Fatty acid	Dose in Gy.	host diet								
		wheat			cowpea			peanuts		
		control	100	300	control	100	300	control	100	300
Lauric	12 : 0	—	0.52	1.32	—	—	—	—	—	—
Myristic	14 : 0	0.51	0.74	—	—	—	0.84	—	—	3.41
Palmitic	16 : 0	31.80	34.30	23.45	29.48	32.08	15.82	18.77	20.46	4.30
Palmetoleic	16 : 1	1.48	1.22	—	—	—	2.11	2.04	—	—
Stearic	18 : 0	4.45	5.47	6.01	8.70	7.08	7.17	5.35	7.70	4.81
Oleic	18 : 1	46.03	44.91	49.64	51.42	50.30	54.00	45.96	52.08	28.99
unknown		—	—	—	—	—	2.91	—	—	—
Lionoleic	18 : 2	13.97	13.55	17.00	10.40	9.41	10.82	20.94	19.76	11.94
Linolenic	18 : 3	0.32	0.52	—	—	1.39	1.68	—	—	—
Arachidic	20 : 0	0.54	0.79	—	—	—	1.08	—	—	—
Behenic	21 : 0	—	0.61	—	—	—	—	—	—	—
Docosanoic	22:0	0.52	0.74	—	—	—	1.16	—	—	—
Triconsanoic	23:0	—	—	—	—	—	—	—	—	37.36
Tetracosanoic	24:0	—	—	2.57	—	—	2.41	6.96	—	4.19
Hexacosanoic	26:0	0.39	—	—	—	—	—	—	—	—
Unsaturated		61.8	60.2	66.69	61.82	60.83	68.44	68.49	71.84	40.93
Saturated		38.2	39.8	33.36	38.18	39.17	31.39	31.06	28.16	59.07
Un Saturated/Saturated		161.8	151.3	199.8	161.9	155.3	218.6	230.0	255.1	69.29

The fatty acids 18:1 and 16:0 occurred as major components among the other acids of females reared on untreated wheat (46.03% and 31.80%). The level of unsaturated acids 18 : 1 and 18 : 2 increased by increasing the irradiation dose (49.64% and 17.00%) on the contrary, 16 : 0 level increased in females with 100 Gy (34.30%) and decreased with 300 Gy (23.45%). The fatty acids 12 : 0 was found in small concentration with irradiated host diet although it was not in present in control. It is clear from the data that concentration of most fatty acids increased when females were fed on irradiated diet.

The major fatty acids 18 : 1 (51.42%) in female reared on cowpea increased when host irradiated with 300 Gy (54.0 %). However, the level of 16:0 increased with 100 Gy (32.08 %) and decreased to (15.82 %) at 300 Gy . The radiation dose effectively reduced the content of 18 : 0. In females fed as a larvae on irradiated cowpea, the fatty acids 16 : 1, 20 : 0, 22 : 0, and 24 : 0 were presented in small concentrations.

The unsaturated fatty acid 18 : 1 represents the major one (45.96) of females reared on peanuts, it was increased with 100 Gy (52.08%) and decreased (28.99%) at 300 Gy . The same trend was observed with saturated fatty acids 16 : 0 and 18 : 0 Whereas the level of 18:2 was decreased by increasing the irradiation dose. The fatty acid 24 : 0 was found only in female reared on irradiated peanut with 300 Gy as a major component among the other fatty acids.

The ratio between unsaturated and saturated fatty acids reached a maximum in females and males reared on irradiated peanuts with 100 Gy (255.1 and 260.0) and minimum was found in females on irradiated peanuts with 300 Gy (69.29) while, decreased in males (33.9) reared on irradiated wheat with 300 Gy.

Turunen (1983); Subramanyam and Outkomp (1987) mentioned that the unsaturated fatty acids especially 18 : 2 (linoleic) and 18 : 3 (linolenic) are essential for normal growth, development and adult emergence. From the above results the changes in the two unsaturated fatty acids as a results of feeding on irradiated diet of the hosts could be used as additional bioindex for disturbance on the biological activity of insects.

In the present work, the results showed that there was difference between sexes in total lipids, females had higher lipids than males. Also differences in total body lipids and principal fatty acids composition between sexes may be related to their lipid requirements. (Turunen, 1983) reported that the levels of lipids and fatty acids are profoundly influenced by the diet.

The fatty acids of adults from larvae reared on different hosts reflects more closely the fatty acids of their hosts. Thus when fed on diet with a high concentration of fat the adults tends to deposit the fatty acids of its host. However, when fed on diet with low concentration of fat the adults tends to synthesize its fatty acids and deposit fat differed from that of the host. Afify *et al.* (1997) recorded similar conclusion with *C. maculatus*.

The administration of radiation dose for the hosts could alter the composition of fatty acids. The presence of unknown fatty acid in case of irradiated cowpea with 300 Gy in female may be formed from isomerization of 18 : 1 caused by radiation and the chemical composition of cowpea. The

effect of sterilization by gamma radiation on fatty acids synthesis have been reported in several insect species. Radiation influences the biosynthetic pathway of fatty acids and effects fatty acids composition (Afify et al., 1995, El-Orabi and Ghareib, 2000, Haiba and Mohamed, 2001). These effects caused changes in structure of phospholipids and their biological significance as nutritional, prostaglandinogenic and presumed structural role (Downer, 1978, Stanley – Samuelson and Dadd, 1983).

Ionization causes the majority of the immediate chemical changes in living materials. This damage may be a direct result of an ionizing track or it may be due to the indirect action of the free radicals (Coggle, 1977).

Generally it could be concluded that fatty acid composition affected indirectly may be by feeding larvae on the irradiated diet, thus cause disturbance on the biological activity of insects.

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تأثير العوامل المعاملة بأشعة جاما على محتوى الدهون والأحماض الدهنية في حشرة خنفساء الصعيد .

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** قسم الكيمياء الحيوية – كلية الزراعة – جامعة القاهرة .

درس تأثير تربية اليرقات على بعض العوامل (الفمغ – اللوبيا – الفول السوداني) والمعاملة بأشعة جاما بالجرعات ١٠٠ ، ٣٠٠ جراي في تقدير محتوى الدهون والتغيرات التي تحدث في الاحماض الدهنية في أجسام الحشرات الكاملة من الذكور والإناث . أظهرت النتائج تأثير الإشعاع الغير مباشر على محتوى الدهون نتيجة تغذية اليرقات على العوائل الغذائية المعاملة .

حدث تغيير في نوعية وكمية الأحماض الدهنية تبعاً للعامل ، وكذلك ما بين الذكور والإناث للحشرة ، وكان للإشعاع تأثير في تغيير كميات الأحماض الدهنية المشبعة والغير مشبعة وخاصة اللينولينيك (ك١٨:٢) ، واللينولينيك (ك١٨:٣) والمسئولة عن النمو والتطور وخروج الحشرات. وعموماً فإن الاحماض الدهنية تأثرت بطريق غير مباشر نتيجة تنحية اليرقات على العوائل الغذائية المعاملة بأشعة جاما بالمقارنة عند التغذية على عوائل غير معاملة بالإشعاع . والتي أثرت بدورها على الصفات الحيوية للحشرة .