

## Effect of irradiation treatments on food biosafety.

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### Abstract

The aim of the current study clarifies the effect of food irradiation as a preservation method on microbiological analysis and growth performance as well as genetic processes of *Clarias garipinus* by estimating chromosomal aberration test in somatic cells and compare it with the effect of other food preservations such as preservation by freezing and preservation by thermal treatment on food. The results indicate that 25 and 35 kGy doses of gamma irradiation showed there were no growth in the total viable (mesophilic aerobic) plate counts and increased the shelf-life of samples compared with other preservation methods which used. There were no significant differences between irradiated and non-irradiated sample ingrowth Parameters. Higher dose of gamma irradiation was apparent that there were a significant ( $P < 0.05$ ) decrease of the value of chromosomal aberrations.

**Keywords:** irradiation, food biosafety.

### Introduction

Food irradiation is a preservation process of exposing food to high energy rays to improve product safety and shelf life. It could be used to replace chemical preservatives as well as thermal treatment. It is considered as cold pasteurization of food and currently permitted in 35 countries worldwide for 40 different food products. Irradiation can be used to reduce the number of pathogens and so increase food safety, reduce the number of spoilage organisms, leading to an extension in shelf-life and a reduction in waste, due to spoilage. Irradiation can kill microorganisms, insects and parasites and this is a fundamental reason for applying the technology to improve the safety and quality of many foods and food products (Patterson 2005). The food industry is focused on manufacturing long-shelflife ready-to-eat (RTE) products in domestic portions from processed blocks (Cabeza *et al.*, 2009; Gil-Diaz *et al.*, 2009). Irradiation, as a method of meat products preservation, has excellent potential in the elimination of pathogenic and spoilage microorganisms from meat and meat products (Mayer- Miebach *et al.*, 2005; Badr, 2004; Satin, 2002). One of the major concerns in irradiation meat and meat products, however, is its effects on meat and meat products quality, mainly because of free radical reaction resulting in the possibility of color change, lipid oxidation and odor generation, and consumer response to these quality changes are quite negative (Du *et al.*, 2002). There is abundant literature on the effects of ionizing radiation on meat (Sweet *et al.*, 2006; 2005), meat products (Chouliara *et al.*, 2006), and prepared meals (Irawati *et al.*, 2007).

### Material and Methods

#### 1- Experimental model

Catfish, *Clarias garipinus* was used as the experimental animal model in this study. The fish were obtained from the Arab fisheries hatchery, Abu-Hammad, Sharkia Governorate, Egypt and transferred to the genetic and biotechnology lab, Faculty of Agriculture Benha University.

#### 2- Experimental Design and facilities

A one-way experiment was designed to study the main effect of different methods of preservation of meat product on growth performance, feed utilization and chromosome aberration of Catfish. The experimental design was outlined in **Table 1**. Prior to beginning of the experiment, fish were acclimatized to the experimental conditions and fed commercial diet (30% protein) twice daily to apparent satiation by hand for 15 days. After the acclimatization, the experimental fish were distributed randomly into the experimental plastic tanks. A set of 420 fish of *Clarias garipinus* average initial weight of ( $4.01 \pm 0.06$  g) were used in this trail. Thirty-five fish were randomly stocked into each tank with two replications for each treatment. De-chlorinated public utility water was supplied to each aquarium housed within an artificially illuminated room. About one-third of water volume in each tank was daily replaced by aerated fresh water after removing the accumulated excreta. During the 90-days experimental period, triplicate groups of catfish were hand-fed with twice daily at 09:00 am and 3:00 pm. The first time fed on artificial diet and the second-time fish fed on preservation of meat product.

**Table 1.** Experimental design of the present study

Treatment, T.	Artificial diet (4% biomass)	Meat product (0.5% biomass)
T <sub>1</sub> Control	Twice daily	Once daily
T <sub>2</sub> Frozen product	Twice daily	Once daily
T <sub>3</sub> Heat product	Twice daily	Once daily
T <sub>4</sub> Radiation 25 product	Twice daily	Once daily
T <sub>5</sub> Radiation 35 product	Twice daily	Once daily

### 3- Water quality for Aquaculture

Water temperature was recorded daily at 1.00 pm using a mercury thermometer. Dissolved oxygen (DO) was measured at 07.00 am using YSI model 56 oxygen meter (YSI Company, Yellow Springs Instrument, Yellow Springs, Ohio, USA). Total ammonia and nitrite were measured twice weekly using a DREL, 2000 spectrophotometer (Hash Company, Loveland, CO, USA). A pH was estimated on morning by using a pH meter (Model HANNA instruments HI 2210). All tested water quality criteria (temperature, pH value, Dissolved oxygen (DO) and total ammonia) were suitable and within the acceptable limits for rearing catfish. (Boyd, 1990).

### 4- Diet preparation

The basal practical diet was formulated to contain approximately 28.99 % crude protein and 16.10 KJ/Kg diet<sup>1</sup> Metabolizable energy, which has been shown to be sufficient to support the optimal growth of *Clarias garipinus*. Soybean meal contributed the major portion of dietary protein, with the rest ingredients coming from local market (fish meal, corn gluten, yellow corn and wheat bran). All dry ingredients were thoroughly mixed with soybean oil, and vitamins and minerals mixture, and then passing the mixed feed through a laboratory pellet mill (2-mm die), and stored at -20 °C until used.

### 5- Preparation the samples

#### 5.1. Preparation of frozen Food

Samples were stored in deep freezer at -18 °C for two weeks, as in industrial methods, Samples were kept at -18 °C until used.

#### 5.2. Preparation of heat treatment Food

Samples were cooked by autoclave at 121 °C for 20 min, as in industrial methods, after autoclaving the samples were transported to the laboratory and were stored at room temperature until used.

#### 5.3. Preparation of Irradiated Food

Samples were subjected at ambient temperature to gamma irradiation from Co-60 source at National Center for Radiation Research and Technology at Nasr City, Cairo. The facility used was Gamma Chamber 400 a Co-60 facility of India. The applied dose was 25 and 35 kGy delivered at a dose rate of 5.212 kGy/h as calibrated using small pieces of the radio chromic film (McLaughlin et al., 1985), at the time of experimentation. After irradiation, the samples

were transported to the laboratory and were stored at room temperature until used.

### 6- Microbiological analysis

The samples were prepared for microbiological examination according to ICMSF (1996). All samples were examined for Total colony count (TCC); Total Mold and Yeast count; Coliform count and total *Staphylococcus* count/g, according to American public health Association (APHA, 1992 and 2001).

### 7- Growth performance and feed utilization parameters

Records of live body weight (g) was measured in all fish for each tank and registered every 14 days (two weeks) during the experimental period. Growth performance parameters were measured by using the following equations:

**7.1. Weight Gain:** Weight gain was determined between the final weight and initial weight of experimental fish. Weight gain = Final weight - Initial weight.

**7.2. Specific Growth Rate:** It is the percentage rate of change in the logarithmic body weight and was computed.

$$SGR = [(Ln \text{ final weight} - Ln \text{ initial weight}) / \text{Time (days)}] \times 100$$

**7.3. Survival rate:** at the end of the experiment, water in all ponds was drained and fish were counted. The number of fingerlings at the start and end of the experiment was used to calculate percentage survival rate (SR %).

### 8- Chromosomal preparation

Chromosomal preparation of kidney tissues were carried out according to the method described by (Al-Sabti, 1983) with some modification: In brief: the anterior kidney from each fish were excised and cut into fine particles in 5-7 ml of RBMI medium and 0.2 ml of 0.05 colchicine were added to each tube *in vitro*. Cultures were incubated at 37-38 °C for 1 h then the cells were centrifuged at 1000 rpm for 10 min and resuspended in prewarmed (37 °C) hypotonic solution (KCl 0.5 %) for 30 min at 37 °C. The sample were centrifuged and fixed in cold mixture of 1:3 glacial acetic acid and methyl alcohol. Two changes of the same fixative were applied with centrifugation and removal of the supernatant fluid each time then the sediment were suspended in a small amount of the

fixative. The slides were produced by the conventional method and stained with Giemsa stain. Chromosome analysis were carried out in one hundred metaphase spreads for each fish.

### Statistical analysis

All data were analyzed by using the software SAS, version 6.03 (**Statistical Analysis System 1993**). One-way analysis of variance (One-way ANOVA) was used to determine whether significant variation existed between the treatments. When overall differences were found, differences between means were tested by **Duncan (1955)** new multiple range test. One way ANOVA was used for analyzing the individual effects of these treatments. All differences were considered significant at  $P < 0.05$  and the results are presented as means with pooled standard error of the mean).

### Results and Discussion

#### Microbiological analysis.

The results presented in table (2) showed microbiological load for untreated and treated samples which exposed to different food preservation methods such as freezing, thermal treatment and irradiation treatment ( using doses 25 kGy and 35 kGy) before treatment, after treatment directly and after two weeks from the treatment time. The statistical analysis of the data demonstrated that there were no significantly different in bacterial growth count between all the samples before treatment. However, the samples in the treatment T<sub>3</sub>, T<sub>4</sub> and T<sub>5</sub> showed that no growth (not

detected) after treatment directly and after two weeks from treatment. However, there were significantly ( $P < 0.05$ ) decreased the total (mesophilic aerobic) plate microorganism counts (TPCs), Total mould count (TMC), *Staphylococci* and *E. coli* with the increase of storage period in the treatment T<sub>2</sub>. **Al-Bachir, 2013** indicated that irradiation with 4 or 6 kGy and storage under refrigeration (5 °C) reduced the total microorganisms count and increased the shelf-life of ground beef (**Mohamed et al., 2011**), *Sheesh Tawoq* (**Al-Bachir, 2010**), Chicken *Kabab* (**Al-Bachir et al., 2010**), corned beef (**Sallam et al., 2000**), chicken vegetable and chicken sweet corn soup (**Irawati et al., 2007**), mutton shammi *Kababs* and pork salami (**Sweet et al., 2005**), *Sausage* production (**Chouliara et al., 2006**), *Borak* (**Al-Bachir, 2007**), luncheon meat (**Al-Bachir, 2005**), and Camel meat (**Al-Bachir and Zeino, 2009**). **Shewan, 1975** recommended that the microbial limit as 1-10<sup>6</sup> CFU/g of fish flesh for tropical fishes. Hence, TBC values in the present investigation suggest that the irradiated samples remain acceptable after 90 days at -20°C. In case of total mould count (TMC) it was found that the population increased with the increase of storage period. **Kawser, et al., (2009)** showed that at the beginning of the storage period bacterial growths were affected by the radiation. The initial bacterial load of control was maximum (1.3×10<sup>4</sup> cfu/g) followed by 3 kGy irradiated fishes (2×10<sup>2</sup> cfu/g). At 5 and 8 kGy radiation the samples were completely sterilized resulting no bacterial growth. At days 90, this value increased as 2.1×10<sup>5</sup>cfu/g in control sample stored at -20°C, 2.3×10<sup>4</sup>cfu/g in 3KGy, 6.7×10<sup>3</sup>cfu/g in 5 kGy and 3.5×10<sup>3</sup>cfu/g in 8 kGy sample stored at -20°C.

**Table 2.** Microbiological analysis for product of meat exposed to different food preservation methods.

	Treatments	Yeast	Total count	Staph.	E. coil
Before treatment	T <sub>1</sub>	4.883 cd	6.767 d	4.390 bc	2.420 d
	T <sub>2</sub>	4.847 d	6.807 cd	4.397 bc	2.330 f
	T <sub>3</sub>	4.907 bc	6.897 b	4.410 bc	2.393 e
	T <sub>4</sub>	4.890 bcd	6.830 c	4.447 b	2.437 c
	T <sub>5</sub>	4.940 b	6.917 b	4.297 c	2.507 b
After treatment, directly	T <sub>1</sub>	5.883 a	7.767 a	4.690 a	3.420 a
	T <sub>2</sub>	3.810 f	6.347 e	3.040 e	2.227 g
	T <sub>3</sub>	0.0000 h	0.0000 h	0.0000 g	0.0000 j
	T <sub>4</sub>	0.0000 h	0.0000 h	0.0000 g	0.0000 j
	T <sub>5</sub>	0.0000 h	0.0000 h	0.0000 g	0.0000 J
Two weeks after treatment	T <sub>1</sub>	3.883 e	5.767 f	4.123 d	2.220 h
	T <sub>2</sub>	2.677 g	4.743 g	2.860 f	1.940 i
	T <sub>3</sub>	0.0000 h	0.0000 h	0.0000 g	0.0000 j
	T <sub>4</sub>	0.0000 h	0.0000 h	0.0000 g	0.0000 j
	T <sub>5</sub>	0.0000 h	0.0000 h	0.0000 g	0.0000 j

Means followed by different letters in each column are significantly ( $p < 0.05$ ) different.

T<sub>1</sub> control, T<sub>2</sub> samples exposed to freezing as a food preservation methods, T<sub>3</sub> samples exposed to thermal treatment as a food preservation methods, T<sub>4</sub> samples exposed to irradiation using 25 kGy as a food preservation methods and T<sub>5</sub> samples exposed to irradiation using 35 kGy as a food preservation methods.

### Growth Parameters

#### Live Body weight

The results presented in table (3) showed average live Body weight (g) from 1<sup>st</sup> to 8<sup>th</sup> weeks for catfish fed on Diet (control) and product of meat exposed to different food preservation methods. The data indicated that catfish fed on diet, product of meat stored under freezing and other which fed on product of meat exposed to heat treatment had no significantly ( $p < 0.05$ ) different, during the experimental period 8 weeks. The groups of catfish which fed on product of meat exposed to irradiation (25kGy and 35kGy) had lower body weight than those fed diet (control), during

the experimental period 8 weeks. **Chaubey, et al., (1999)** found that the feeding studies with diets containing irradiated (at a dose of 30 kGy) paprika have shown no deleterious effects in rats with regard to growth. **Irawati and Sani (2012)** reported that average changing in Body Weight of rats showed an increase after feeding of the irradiated ethnic foods. After feeding, the animals seemed to be healthy and have better movement and no leftover of food found in the cage. **Bayoumi (2005)** noticed that the irradiation treatment up to 20 kGy of raw Lentil and Cow Pea produced increasing in live body weight of rats as a compared with those receiving raw samples.

**Table 3.** Body weight (g) of Catfish fed on product of meat exposed to different food preservation methods

Treatments	Initial	2 weeks	4 weeks	6 weeks	8 weeks
T <sub>1</sub> (Control)	4.00	6.50	12.40a	15.47a	18.14a
T <sub>2</sub> Frozen product	4.10	6.49	12.49a	15.64a	17.58a
T <sub>3</sub> Thermal treatment	3.98	6.45	12.45a	14.87a	17.62a
T <sub>4</sub> Radiation 25 product	4.10	5.52	9.51b	10.05a	12.42a
T <sub>5</sub> Radiation 35 product	4.10	5.55	9.56b	10.20b	11.85b
Standard error (SE)	0.01	0.06	0.07	0.04	0.14

Means followed by different letters in each column are significantly ( $p < 0.05$ ) different.

#### Weight gain.

Table (4) presented that change in body weight gain of catfish fed artificial diet and product of meat exposed to different food preservation methods. Data of this table demonstrate that during the experimental period (0-4week) the groups of catfish fed on product of meat exposed to irradiation (25kGy and 35kGy) have significantly lower in WG of compared with other treatment group. Also, there was no significant different in body weight gain between control group and groups of catfish fed on product of meat exposed to freezing and heat treatment. During the experimental period (4-6week) the body WG of catfish groups fed on product of meat exposed to irradiation (25kGy and 35kGy) of gamma irradiation and heat treatment were significantly ( $p < 0.05$ ) lower

than those of the control. During the experimental period (6-8week). There was no significantly different in body weight gain between control group and groups of catfish fed on product of meat exposed to heat treatment and irradiation treatment at 25kGy. The groups of catfish fed on product of meat exposed to irradiation treatment at 35kGy and freezing treatment had significantly (lower body weight gain) compared with control group. **Bayoumi (2005)** found that the irradiation treatment up to 20 kGy of raw lentil and cow pea improved the total body weight gain compared with raw sample. Also, **El-Niely, (2001)** obtained that the radiation processing improved the total body weight gain when rats fed rats irradiated peanut kernels diets at dose levels 5, 7.5 and 10 kGy.

**Table 4.** Weight gain of Catfish fed on product of meat exposed to different food preservation methods

Treatments	0-2 week	2-4 week	4 – 6 week	6 - 8 weeks
T <sub>1</sub> (Control)	2.50a	5.90a	3.07a	2.67a
T <sub>2</sub> Frozen product	2.39a	6.00a	3.15a	1.94b
T <sub>3</sub> Thermal product	2.47a	6.00a	2.42b	2.75a
T <sub>4</sub> Radiation 25 product	1.42b	3.99b	0.54c	2.37
T <sub>5</sub> Radiation 35 product	1.45b	4.01b	0.64c	1.65
Standard error (SE)	0.01	0.06	0.07	0.04

Means followed by different letters in each column are significantly ( $p < 0.05$ ) different.

#### Specific growth rate

As described in Table (5), average values of SGR ranged between 5.52 to 6.50 for the all experimental

period (0-8 weeks) and the differences between fish groups attributed to the feeding of product of meat exposed to heat treatment were not significant.

**Table 5.** Specific growth rate of Catfish fed on product of meat exposed to different food preservation methods

Treatments	0-2 week	2-4 week	4 – 6 week	6 - 8 weeks
T1 (Control)	0.81	1.08	0.37	0.27
T2 Freezing treatment	0.77	1.09	0.37	0.19
T3 Thermal treatment	0.80	1.10	0.30	0.28
T 4 Radiation treatment 25 kGy	0.50	0.91	0.90	0.35
T5 Radiation treatment 35 kGy	0.50	0.91	0.11	0.25
Standard error (SE)	0.01	0.06	0.07	0.04

Means followed by different letters in each column are significantly ( $p < 0.05$ ) different.

### Survival rate of Catfish

The results presented in table (6) showed average survival rate of catfish fed on artificial diet and product of meat exposed to different food preservation methods, during the experimental period (0 - 8 week).

There was no significantly different ( $p < 0.05$ ) during the experimental period 8 weeks. **Irawati and Sani (2012)** reported that no death animals were found after feeding both on unirradiated and irradiated conventional feeds.

**Table 6.** Survival rate of Catfish fed on product of meat exposed to different food preservation methods

Treatments	2 weeks	4 weeks	6 weeks	8 weeks
T1 (Control)	100.00a	97.50a	95.50a	92.00a
T2 Freezing treatment	97.50b	95.00b	95.00a	92.50a
T3 Thermal treatment	100.00a	97.50a	92.50b	92.50a
T 4 Radiation treatment 25 kGy	97.50b	95.00b	92.50b	92.50a
T5 Radiation treatment 35 kGy	95.00b	92.50c	90.00c	90.00b
Standard error (SE)	1.02	1.12	1.01	0.92

Means followed by different letters in each column are significantly ( $p < 0.05$ ) different.

### Chromosomal aberration.

The presented results in table (7) showed that the average values of different types of chromosomal aberrations in head kidney of *Clarias gariepinus*. high percentage of chromosomal abnormalities in head kidney cells of *Clarias gariepinus* were observed clearly in the form of centromeric attenuation, chromatid breaks, chromatid gaps, chromatid deletions, centric fusion, End to end associations and fragmentation. The current results revealed that the values of different types of chromosomal aberration in head kidney cells of *Clarias gariepinus* in treatment T<sub>2</sub>

and T<sub>3</sub> were no significant ( $P < 0.05$ ) compared with control. Also, data showed a significant ( $P < 0.05$ ) decrease of the value of aberrations in treatment T<sub>4</sub> and T<sub>5</sub>. In spite of, chromosomal abnormalities which formed in chromatid deletions and End to end associations showed no significantly ( $P < 0.05$ ) different. **Chaubey, et al., (1999)** studied genetic toxicological for somatic and germinal effects in salted, dried and irradiated (2.0 kGy) tackerel and showed no evidence of any induced chromosomal aberrations in mice.

**Table 7.** Structural aberration (deletion, fragments, ring, centromeric attenuation, end, brake and gab) of Catfish fed on product of meat exposed to different food preservation methods

Treatments	Structural aberrations						
	Deletion	Fragments	Ring	Centromeric attenuation	End	Brake	Gab
T <sub>1</sub> (Control)	4.00	6.50	12.40	15.47a	4.00	6.50	12.40
T2 Thermal treatment	4.10	6.49	12.49	15.64a	4.10	6.49	12.49
T3 Heat treatment	3.98	6.45	12.45	14.87a	3.98	6.45	12.45
T 4 Radiation treatment 25 kGy	4.10	5.52	9.51	10.05b	4.10	5.52	9.51
T5 Radiation treatment 35 kGy	4.10	5.55	9.56	10.20b	4.10	5.55	9.56
Standard error (SE)	0.01	0.06	0.07	0.04	0.01	0.06	0.07

Means followed by different letters in each column are significantly ( $p < 0.05$ ) different

## Conclusion

Preservation using thermal treatment and preservation by gamma Irradiation at doses 25 and 35 kGy can be effective to control microorganisms in soy sauces beef, compared with preservation by freezing which decreased the total (mesophilic aerobic) plate microorganism counts (TPCs), total Mould count (TMC), *Staphylococci* and *E.coli* with the increase of storage period and extending their refrigerated shelf-life for more than 8 weeks. The obtained results that feeding studies on soy sauces beef (as a meat product) exposed to gamma irradiated at 25 kGy and 35 kGy as a method of food preservation on *Clariasgaripinus*, respectively did not show any abnormalities in growth performance, feed utilization and chromosomal aberration of the observed *Clariasgaripinus*, in comparison to the other used food preservation methods such as preservation using thermal treatment and preservation by freezing.

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### تأثير معاملات الإشعاع على الأمان الحيوي للغذاء

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قسم الوراثة والهندسة الوراثية، كلية الزراعة بمشتر، جامعة بنها

حفظ الغذاء بالتشعيع يتم من خلال تعريض الأغذية لجرعات كبيرة من الأشعة المؤينة الهدف منها قتل أو تثبيط نشاط البكتيريا والفيروسات والحشرات وأى كائنات حية أخرى تسبب تلف الأغذية. الإشعاع يحفظ جودة الأغذية ويطيل فترة صلاحيتها عن طريق قتل الكائنات الحية المسؤولة عن الفساد وتدمير الكائنات الحية الدقيقة الضارة في الغذاء، وبالتالي زيادة فترة صلاحيته مما يسهل من عملية التبادل التجاري عن طريق التصدير.

أجريت هذه الدراسة بقسم الوراثة والهندسة الوراثية بكلية الزراعة بمشتر، جامعة بنها. بهدف دراسة تأثير معاملات الإشعاع على الأمان الحيوي للغذاء وجودته ومقارنة ذلك بتأثير طرق الحفظ الأخرى وهي التجميد والمعاملة الحرارية. حيث شملت الدراسة أربع مجاميع من العينات كالتالي:

- 1- المجموعة الأولى: عينات البيف الغير معاملة (المجموعة الضابطة).
- 2- المجموعة الثانية: عينات البيف المعاملة أو المحفوظة بالتجميد:-

هذه العينات تم حفظها في الديب فريزر على حرارة -18°م لمدة إسبوعين تماماً كما هو الحال في طرق التصنيع التجاري.

- 3- المجموعة الثالثة: عينات البيف المعاملة بالحرارة.

هذه العينات تم طبخها في الأوتوكلاف الخاص بالمعمل على حرارة 121°م لمدة 20 دقيقة. بعد عملية الطبخ هذه تم نقل العينات إلى المعمل وتم الحفظ في درجة حرارة الغرفة لحين الاستخدام.

- 4- المجموعة الرابعة: وهذه المجموعة تم تقسيمها إلى مجموعتين وهما:

أ: العينات المعاملة بالإشعاع على جرعة 25 كيلو جرای.

ب: العينات المعاملة بالإشعاع على جرعة 35 كيلو جرای.

هذه العينات تم تعريضها لأشعة جاما من مصدر كوبلت 60 (Co-60) في المعمل القومي لأبحاث الإشعاع والتكنولوجيا بمدينة نصر، محافظة القاهرة. تم نقل العينات المعاملة بالإشعاع إلى المعمل والحفظ على حرارة الغرفة لحين الاستخدام.

تم دراسة تأثير هذه المعاملات على بيف الصويا كأحد منتجات اللحوم من حيث التأثير على الحمل الميكروبي وتم إعداد العينات للفحص البكتريولوجي طبقاً لـ ICMSF 1996. تم تقدير التالي ذكره في كل العينات طبقاً لتوصيات الجمعية الأمريكية العامة للصحة 1992, 2001, APHA.

1- العد الكلي البكتيري ( العدد الأكثر احتمالاً).

2- الكشف عن الفطر والخميرة.

3- الكشف والتعرف على الكوليفورم.

4- الكشف والتعرف على *Staphylococcus*.

تم إجراء هذه الإختبارات قبل المعاملات مباشرة ، بعد المعاملات مباشرة وبعد مرور إسبوعين من وقت المعاملة. ومن خلال النتائج المتحصل عليها وجد الآتي.

1- لا يوجد أى فروق معنوية في عدد النيمات البكتيرية بين كل العينات قبل المعاملة.

2- لا يوجد أى نيمات بكتيرية في كلاً من العينات المأخوذة من المجموعة الثالثة والمجموعة الرابعة بقسميها (25 ، 35 كيلو جرای) بعد المعاملات مباشرة كذلك بعد مرور إسبوعين على وقت المعاملات.

وأستهدف البحث دراسة تأثير مدى الأمان الحيوي لمعاملات الحفظ المستخدمة على منتج البيف كمنتج إستهلاكي للإنسان بدراسة تأثير هذه المعاملات على:

أداء النمو ومدى الاستفادة من الغذاء وكذلك التشوهات الكروموسومية في أسماك القرموط الإفريقي *Clarias garipinus* وذلك بعمل تجربة تغذية على القرموط الإفريقي *Clariasgaripinus* كموديل.

شملت الدراسة على مجموعتين من الأسماك الأولى عبارة عن كنترول والأخرى تم تقسيمها إلى أربع مجاميع مختلفة كلاً منها يحتوى على 35 سمكة كالتالي:

1- تم تغذية المجموعة الأولى على عليقة تحتوى ضمن مكوناتها على البيف المحفوظ بالتجميد.

2- تم تغذية المجموعة الثانية على عليقة تحتوى ضمن مكوناتها على البيف المحفوظ بالمعاملة الحرارية.

3- تم تغذية المجموعة الثالثة على عليقة تحتوى ضمن مكوناتها على البيف المحفوظ بالإشعاع على جرعة 25 كيلو جرای.

4- تم تغذية المجموعة الرابعة على عليقة تحتوى ضمن مكوناتها على البيف المحفوظ بالإشعاع على جرعة 35 كيلو جرای.

تم قياس وتسجيل الوزن الحى لكل الأسماك في كل مجموعة كل 14 يوم (إسبوعين) من فترة التجربة. وتبين الآتى:-

- لا توجد أى فروق معنوية في وزن الجسم النهائى على مدار التجربة لأسماك كلاً من المجموعة الأولى والثانية ووجود نقص معنوى في وزن الجسم النهائى لأسماك المجموعة الثالثة والرابعة عند مقارنة ذلك بالمجموعة الكنترول.
- لا يوجد فرق معنوى في وزن الجسم المكتسب في كلاً من المجموعة الأولى والثانية من بداية التجربة حتى الإسبوع الرابع. وبدأ ظهور الفرق المعنوى في الفترة من الإسبوع الرابع حتى السادس في المجموعة الثانية حيث يوجد فرق معنوى بسيط ، وفي الفترة من الإسبوع السادس إلى الثامن في المجموعة الأولى حيث يوجد فرق معنوى بسيط مقارنة بالمجموعة الكنترول. إضافة إلى عدم وجود فرق معنوى في كلاً من المجموعة الثالثة والرابعة مقارنة بالمجموعة الكنترول والمجموعات الأخرى على مدار التجربة.
- لا يوجد أى فروق معنوية في معدل الاعاشة على مدار فترة التجربة (0-8) أسابيع.

وقد تم تشريح الأسماك وتجميع العينات من الكلية لدراسة تأثير التغذية على البيف المعامل بطرق الحفظ المختلفة على بعض العمليات الوراثية في القرموط الإفريقي *Clarias garipinus* مثل التشوهات الكروموسومية. وأوضحت الدراسة أن التغذية على المنتج المعامل بالإشعاع بجرعته 25، 35 كيلو جرای لها تأثير مشوه على أشكال الكروموسومات

1- بالنسبة للتشوهات الكروموسومية التركيبية فقد إشتملت على

Centromeric attenuation, chromatid breaks, chromatid gaps, chromatid deletions, centric fusion, End to end associations and fragmentation.

2- وقد أدت التغذية على المنتج المعامل بالإشعاع بجرعته 25، 35 كيلو جرای إلى انخفاض معنوي في جميع أنواع التغيرات التركيبية.