BioBacta

Journal of Medical and Life Science
https://jmals.journals.ekb.eg/

# Anti-Obesity Activity of Carotenoid Extracted From Sea Urchin *Paracentrotus lividus*

**Noha M. Samak *, Gihan M. El-Khodary, Eman H. Radwan, Maha M. El kateb, and Amal Z. Ghoneim**

Zoology Department, Faculty of Science, Damanhour University, Damanhour, Egypt

*Corresponding Author: Noha M. Samak, Zoology Department, Faculty of Science, Damanhour University, Damanhour, Egypt. +201005301157: noha_samak@yahoo.com

**DOI: 10.21608/jmals.2023.318718**

**Abstract**:

The present study was carried out on marine edible sea urchin *Paracentrotus lividus* which are predominant in the local aquatic fauna in Egypt. The present research was conducted to extract carotenoids from soft parts of sea urchin and estimate their activity on obese mice that receive high-fat diet HFD as a protective and curative effect. The results showed the effect of carotenoid in a protective group, significantly reduced plasma triglyceride, HDL, and total cholesterol also lowered blood glucose levels. Oral administration of carotenoid extracts substantially decreased the rate of gaining weight and recovered lipid profile levels to normal as compared to the control group with improvement of liver function tests in serum and oxidative stress parameters in liver tissue. Histological differences in liver tissues revealed that the extracts protected the liver tissue from adiposeness by high-fat diet HFD fed.

In conclusion, this study demonstrates the potential anti-obesity effects of the carotenoid administration in mice supplied with a diet rich in fat and obese mice. The findings suggest that the bioactive compound carotenoid as astaxanthin supplementation extracted from *Paracentrotus lividus* exhibits anti-obesity activity that can improve lipid profiles, glucose metabolism, and antioxidant status.

**Keywords**: Anti-obesity activity; sea urchin; Carotenoids; Astaxanthin; High fat diet

---

## Introduction:

According to the recommendations of the World Healthcare Organization **(WHO)**, obesity and being overweight, both include an excess or abnormal accumulation of fat that is harmful to health. Many comorbidities are closely related to obesity, including metabolic inflammation, insulin resistance, hepatic steatosis, hypertension, dyslipidemia, depression, specific forms of cancer, type 2 diabetes, and cardiovascular disease that are highly correlated with obesity **(Larsson *et al.*, 1984)**. According to the **WHO**, approximately 39% of adults worldwide were overweight in 2016 and 13% of adults were obese **(WHO)**.

Animals nourished with a high-fat diet (HFD) display mild obesity, therefore being ideal for the development of preventive treatments for metabolic disorders such as NAFLD **(Park *et al.*, 2005; Tan *et al.*, 2017).** To identify the various pharmacological effects of the test substance, an HFD-fed mouse model was chosen.

There is a lot of interest in carotenoids as nutritional supplements, especially those derived from natural

Receive Date: 11 February 2023, Accept Date: 20 April 2023, Published: 26 September 2023

*Journal* of *Medical and* Life Science, *2023, Vol.5, No. 3, P.224-237* *pISSN: 2636-4093, eISSN: 2636-4107*

sources, because of their ability to promote health. All terrestrial and aquatic photoautotrophs, including plants, microalgae, macroalgae, and marine invertebrates, produce carotenoid pigments, which are fat-soluble pigments. Carotenoid pigments include red, yellow, and orange tetraterpenoid pigments **(Langi *et al.*, 2018)**. Red carotenoid astaxanthin is a powerful antioxidant found in many different kinds of plants and animals. these naturally occurring anti-oxidants, reduce oxidative damage by detoxification the free radicals molecules in the cells and inhibiting the diseases associated with them. Numerous extremely significant pharmacological actions, such as anti-oxidant, **(Fukuhara *et al.*, 1998, and Naguib, 2000)** anti-tumor and anti-cancer **(Chew *et al.*, 1999)** anti-diabetic (**Naito *et al.*, 2004**) and anti-inflammation activities. **(Guerin *et al.*, 2003, and Bennedsen *et al.*, 1999).**

Marine invertebrates belonging to the phylum Echinodermata include the sea urchin *Paracentrotus lividus*. The edible parts are coated with many sharp spines. The five gonads, which are situated below the interambulacral plates, are the edible tissues **(Gabin-Sánchez and Lorenzo, 1993).** The appearance, texture, flavor, and most crucially color of sea urchin gonads (roe), a highly prized meal have a significant impact on their marketability and price **(Unuma *et al.*, 2010)** and **(Symonds *et al.*, 2007)**. It is believed that the carotenoids and carotenoid precursors from the sea urchin's food, which are either changed or deposited in the gonads tissue, are what gives the gonads their distinctive color **(Symonds *et al.*, 2007),** and **(Symonds *et al.*, 2009**).

Hence, the objective of this study was to evaluate the potential anti-obesity effects of carotenoid extracts, specifically Astaxanthin derived from marine Echinodermata sea urchin *Paracentrotus lividus*, using mouse models with diet-induced obesity.

## Materials and methods:

**Sea urchin collection**. The identification of selected Sea urchin *Paracentrotus lividus* **(Lamarck, 1816)** samples was based on the morphological description provided by **(Guill, Micheal).** Samples were collected from Miami in the Mediterranean Sea, Alexandria, Egypt between July 2020 and February 2021. In obtaining bags filled with fresh seawater, the samples were carried to the lab.

**Dissection**. An incision was performed in the urchin's peristomal membrane, and the coelomic fluid was removed to extract the soft parts (muscles and gonads) as described by **Kelly *et al.* (2001)**.

**Carotenoid extraction from soft parts (muscles and gonads) of sea urchin *Paracentrotus lividus*:**

According to **Chen and Yang (1992)**, carotenoids were isolated from the soft sections (muscles and gonads) of Echinodermata sea urchin *Paracentrotus lividus*. A flask and shaken were used to combine precisely 15 g of soft sections of sea urchin with 150 ml of acetone for half an hour. The collection of the filtrate was accomplished by employing an aspiration pipe, and the leftover material was once more extracted using 150 ml of acetone. The extraction process was done twice with 150 ml of petroleum ether until the residue lost its color. In a flask, we mixed all of the extracts. Then, 600 ml of a ten percent solution of sodium sulfate was mixed. The solution was shaken for one minute before being placed in the dark, resulting in the formation of two distinct layers. The residue was extracted four more times with 150ml of petroleum ether after the supernatant was collected. The extracts were mixed and dehydrated by evaporation.

**Calculation** UV-visible spectrophotometry is a widely used analytical technique that involves the measurement of the absorption or transmission of electromagnetic radiation throughout the spectral range of 380 to 750 nm, in triplicate, which was utilized to calculate the total amount of extracted carotenoids. The Lambert-Beer law was used to determine the carotenoid concentration, and the absorbance readings were used to calculate the results using the following equation:

$$\textbf{Total Carotenoids } \mu g/g = \frac{\left(\frac{Absorbance}{e} \times 1000 \times sample\ volume(ml)\right)}{Sample\ dry\ weigh(g)}$$

The exact coefficient of light absorption of Astaxanthin 124000 at 460 nm and the molecular mass of 596.84 (astaxanthin) were used. This was done by **Bunchwaldt and Jencks in 1968**. In the above solution, Acerafed was used **(Minguez and Mendez, 1993)**.

## Experimental design:

### Animal treatments and diets:

The studies were performed with 32 Swiss male albino adult mice obtained from a medical research institute (Alexandria, Egypt) with an average weight of 25 ± 5 g. Aged(5-6 weeks). Experimental animals were housed in plastic cages on wood shaving. Before the experimental work, all animals underwent a two-week acclimation period and were provided with a regular, high-fat diet (HFD). A constant 23° C was maintained in the animal room. The light was on its cycle of light and dark at 12:12 h. They were regularly monitored for any indications of disease, stress, or mortality and had free access to a routine lab. Feeding occurs by fat diet to obtain obese male mice which contain: Corn starch72.8 g, sucrose172.8 g, Lard177.5 g, mineral mix10 g, Calcium carbonate5.5 g, Soybean Oil 25 g, and beef tallow 60% calories of fat injection by gastric tube for 6 weeks the model determined by **Ikeuchi *et al.*, (2007)**.

### Animal grouping:

Four groups of animals were designed using random selection, each group composed of eight male mice.

**Group 1:** (control): Mice were fed a normal diet and administrated with only saline.

**Group 2:** (control obese mice) Mice were fed highly fatty meals (HFD) for 6 weeks.

**Group 3:** (protective group) Mice were injected with extracted carotenoids (30mg/ kg of body weight) **(Ikeuchi *et al.*, 2007)** by gastric tube and received a highly fatty meal (HFD) at the same time for 6 weeks.

**Group 4:** (curative group) A highly fatty meal (HFD) was administered to mice for 4 weeks till became obese mice, then mice were injected (30mg/ kg of body weight) **(Ikeuchi *et al.*, 2007)** of extracted carotenoids with a gastric tube for 2 weeks.

After a 6-week experimental study period, all mice were required to fast overnight before being put under anesthesia so that blood samples could be taken from their retro-orbital veins. The serum was then processed and frozen at -20$^{o}$ C in preparation for future biochemical examination.

## Experimental procedures

### Biochemical analysis of serum

Commercial kits were used for enzymatic colorimetric testing (triglyceride E-test, cholesterol E-test) to determine the concentrations of High-density lipoprotein (HDL), Triglyceride (TG), and Total cholesterol (TC) in the serum. whereas Total glucose was determined by a colorimetric method using Glucose Oxidase according to **Trinder (1969)**. In contrast, reagents from Biodiagnostic and Research Reagents Company kits were used to detect the enzymes alanine aminotransaminase (ALT) (EC 2.6.1.2), aspartate aminotransaminase (AST) (EC 2.6.1.1), and gamma-glutamyl transferase (GGT) (EC 2.3.2.2) via a colorimetric method that was in agreement with the indicated reference techniques **(Reitman and Frankel, 1957; IFCC, 1986)**. Whereas, Albumin, Total protein, and Total bilirubin were determined by the kit of Biodiagnostic and Research Reagents Company utilizing the colorimetric procedure following the respective suggested reference methods of **Walter and Gerade (1970)** and **Doumas (1971).**

### Biochemical markers in Liver tissue

To estimate liver enzymes, 10 percent of liver tissue homogenate was made and utilized to calculate **Glutathione (GSH)** which was determined by reducing 5,5-dithiobis (2-nitrobenzoic acid) (DTNB) with GSH to generate a yellow molecule, as described by **(Beutler *et al.*, 1963). Superoxide dismutase (SOD)**, **catalase, Total**

*Journal* of *Medical and* Life Science, *2023, Vol.5, No. 3, P.224-237* *pISSN: 2636-4093, eISSN: 2636-4107*

**malondialdehyde (MDA),** and **Nitric oxide** were measured by the kit of Biodiagnostic and Research Reagents Company by using colorimetric Method according to the recommended reference method of **(Nishikimi, 1972)**, **(Aebi, 1984), (Satoh, 1978 and Ohkawa *et al.*, 1979)** and **(Montgomery, 1961)** respectively.

### Histological examination of liver tissue

Tissues from the liver were preserved in 10% formalin and embedded in paraffin, serial sectioned (15 mm). Before being photographed at a final magnification of 200x, the sample was cleaned, mounted, and stained with hematoxylin and eosin.

### Statistical analysis

All experiments were conducted with triplicates of each sample, and the results were expressed as means ± standard deviations (SD). One-way analysis of variance (ANOVA) was used to determine whether there were statistically significant differences between the treatment groups, and a significance level of $p \leq 0.05$ was set. All statistical analyses were performed in SPSS version 20, a statistical package developed by IBM. (IBM-SPSS Version 20.0).

## Results:

### Biochemical analysis of soft parts (muscles and gonads) of sea urchin

### Total carotenoid contents in sea urchin soft parts

The carotenoid concentration, namely astaxanthin, was determined in the soft tissues (muscles and gonads) of sea urchin *Paracentrotus lividus*. The concentration was measured at 460 nm and found to be 5.60 ± 0.02 µg/g.

### Body weight :

The variations in body weight that occurred among the groups throughout the experiments are summarized in (Figure 1). Feeding (HFD) diet high in fat in the obese mice group (G2) caused a marked increase in body weight of 51±2.38 g as compared to the control group (G1) feeding a normal diet of 23.4 ±0.93g showed an extremely significant increase in body weight gain. However, feeding a high-fat diet plus carotenoid extract in the protective group (G3) 33.6 ±1.24 g shows low significant increase in body weight resulting from the consumption of a high-fat diet (HFD). while in the curative group (G4) 44.4±1.69 g that administrated carotenoid extract after receiving (HFD) showed a highly significant increase in body weight gain.

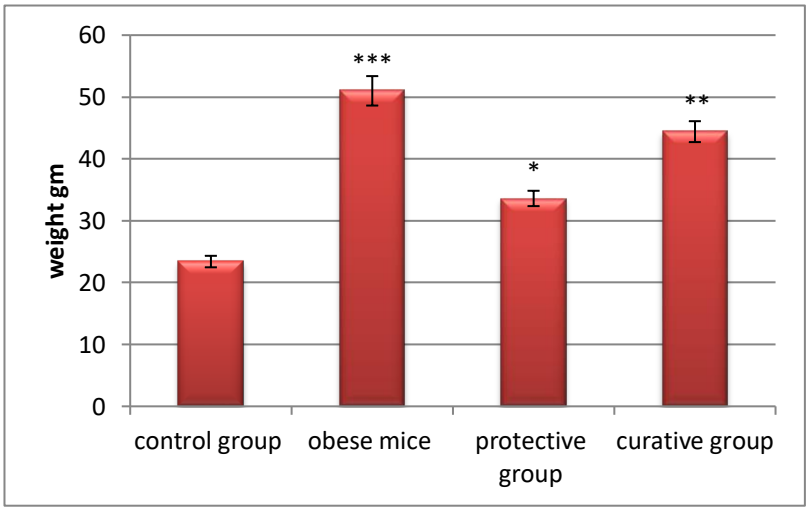


Fig. 1: The significance of carotenoid extracts on mice fed an HFD for 42 days (n = 8 per group; values are means ± SE) Significance was determined at a P-value threshold of ≤ 0.05 (*), ≤ 0.01 (**), and ≤ 0.001 (***), respectively, for the groups controlled by one-way analysis ANOVA.

### Biochemical markers in serum and Liver tissue liver function in serum

Aminotransferases (ALT and AST), and GGT enzyme showed a highly significant increase in the obese mice group (G2) if compared with the control group (G1). While, in protective group (G3) and curative group (G4) showed a significant increase in ALT, AST, and GGT enzyme levels as shown in (Table 1). The content of albumin and total protein has a significant decrease in the obese mice group (G2), protective group (G3), and curative group (G4) compared with the control mice group (G1). The content of bilirubin has a significant increase in the obese mice group (G2), and the curative group (G4) if compared with the control group (G1), while showing insignificant with the protective group (G3).

### Plasma lipids and glucose

The serum's lipid profile levels are displayed in (Table 2). In comparison to the control group (G1), the obese mice group's (G2) triglyceride level was higher. The values in the carotenoid-supplemented obese mice (protective group) (G3) were about 100.57±0.49 mg/dl and 199.63±0.56 mg/dl in the curative group(G4) that received an extract of carotenoid after receiving the high-fat diet (HFD). The obese mice (G2), protective (G3), and curative (G4) groups show a highly significant increase in TG amount if compared with the control mice. In contrast to the control group (G1), the obese mice group's (G2) total blood cholesterol content was greater. The protective group's (G3) results, which were around 98.57±0.49 mg/dl, were lower than those of the obese mice group (G2) 199.90±0.11 mg/dL receiving a diet rich in fat only. The HDL concentration in the serum of the protective group (G3) was 16.20±0.11 mg/dl. The obese mice (G2), protective(G3), and curative (G4) groups show highly significant increases in HDL levels if compared with the normal group (G1). On the other hand, the concentration of glucose in the serum of the obese mice group (G2) was about (129.11±0.12 mg/dl) which is a highly significant increase in the control groups (G1) (88.90 mg/dl).

Table 1: Mean and standard deviation (SD) of Liver functions in serum of mice in experimental groups.

| *Groups* | ALT U/L | AST U/L | GGT U/L | Albumin g/dL | T.protein g/dL | T.Bil mg/dL |
|---|---|---|---|---|---|---|
| *G1 Control* | 65.87±0.15 | 96.53±0.50 | 32.67±0.49 | 4.67±0.15 | 6.17±0.15 | 0.89±0.01 |
| *G2 Obese mice* | 99.60±0.53** | 230.33±0.49** | 53.00±0.10* | 3.70±0.13* | 4.01±0.11* | 1.90±0.11* |
| *G3 Protective Group* | 73.57±0.49* | 127.67 ±0.94* | 50.00±0.08* | 3.90±0.12* | 5.22±0.12* | 1.01±0.12 |
| *G4 Curative group* | 75.57±0.49* | 185.23±1.07** | 55.00±0.12* | 3.00±0.11* | 4.91±0.12* | 1.20±0.10* |

Note: Values are reported as averages of three replicates ± standard deviations (SD), and Significance was determined at a P-value threshold of ≤ 0.05 (*), ≤ 0.01 (**), and ≤ 0.001 (***), respectively, for the groups controlled by one-way analysis ANOVA.

*Journal* of *Medical and* Life Science, *2023, Vol.5, No. 3, P.224-237* *pISSN: 2636-4093, eISSN: 2636-4107*

**Table 2: Mean and standard deviation (SD) of Lipid profile and glucose in serum of mice in experimental groups**

| *Groups* | HDL mg/dL | TG mg/dL | Cholesterol mg/dL | Glocose mg/dL |
|---|---|---|---|---|
| *G1 Control* | 17.10±0.10 | 109.63±0.55 | 88.27±0.56 | 88.90±0.10 |
| *G2 Obese mice* | 8.83±0.16* | 169.23±0.66* | 199.90±0.11* | 129.11±0.12* |
| *G3 Protective Group* | 16.20±0.11* | 100.57±0.49* | 98.57±0.49* | 100.01±0.11* |
| *G4 Curative group* | 12.00±0.10* | 199.63±0.56* | 104.83±0.08* | 108.33±0.49* |

Note: Values are reported as averages of three replicates ± standard deviations (SD), and Significance was determined at a P-value threshold of ≤ 0.05 (*), ≤ 0.01 (**), and ≤ 0.001 (***), respectively, for the groups controlled by one-way analysis ANOVA.

## Oxidative stress in Liver tissue

Oxidative damage was commonly observed in the obese mice group (G2), protective group (G3), and curative group (G4) as indicated by the depletion of GSH, SOD, and CAT and elevated MDA, and nitric oxides as shown in (Table 3). In comparison to the control group (G1) GSH, SOD, and CAT show a significant decrease in obese mice group (G2), protective group (G3), and curative group (G4). On the other hand, MDA and nitric oxide levels increased significantly in the obese mice group (G2), protective group (G3), and curative group (G4) when compared to the (G1) control mice group.

**Table 3: Mean and standard deviation (SD) of Anti-oxidant in liver tissue of mice in experimental groups**

| *Groups* | MDA Nmol/ g. protein | Nitric oxide µmol / g. protein | GSH mg/g. protein | SOD U/g. protein | CAT U/g. protein |
|---|---|---|---|---|---|
| *G1 Control* | 1.99±0.01 | 9.40±0.10 | 129.90±0.10 | 11.00±0.11 | 60.23±1.07 |
| *G2 Obese mice* | 16.90±0.10* | 65.83±0.15* | 91.23±0.59* | 6.13±0.15* | 8.00±0.11* |
| *G3 Protective Group* | 3.00±0.11* | 21.90±0.11* | 106.90±0.11* | 9.00±0.10* | 24.01±0.12* |
| *G4 Curative group* | 16.07±0.15* | 65.33±0.49* | 91.23±0.49* | 8.90±0.12* | 16.57±0.49* |

Note: Values are reported as averages of three replicates ± standard deviations (SD), and Significance was determined at a P-value threshold of ≤ 0.05 (*), ≤ 0.01 (**), and ≤ 0.001 (***), respectively, for the groups controlled by one-way analysis ANOVA.

### Histological examination of liver tissues

Histologically, hematoxylin and eosin staining were used to look at the hepatic fat accumulation. In comparison to the curative and protective group, the hepatic adipocytes in the group of mice with obesity induced by a high-fat diet exhibited a notable increase in both their size and number. The carotenoid extracts treated in the protective group (G3) had a morphology that was almost identical to that of the control group (G1). This suggests that these extracts when taken orally, help keep the liver's adipocyte count and size from altering. (Figure 2)

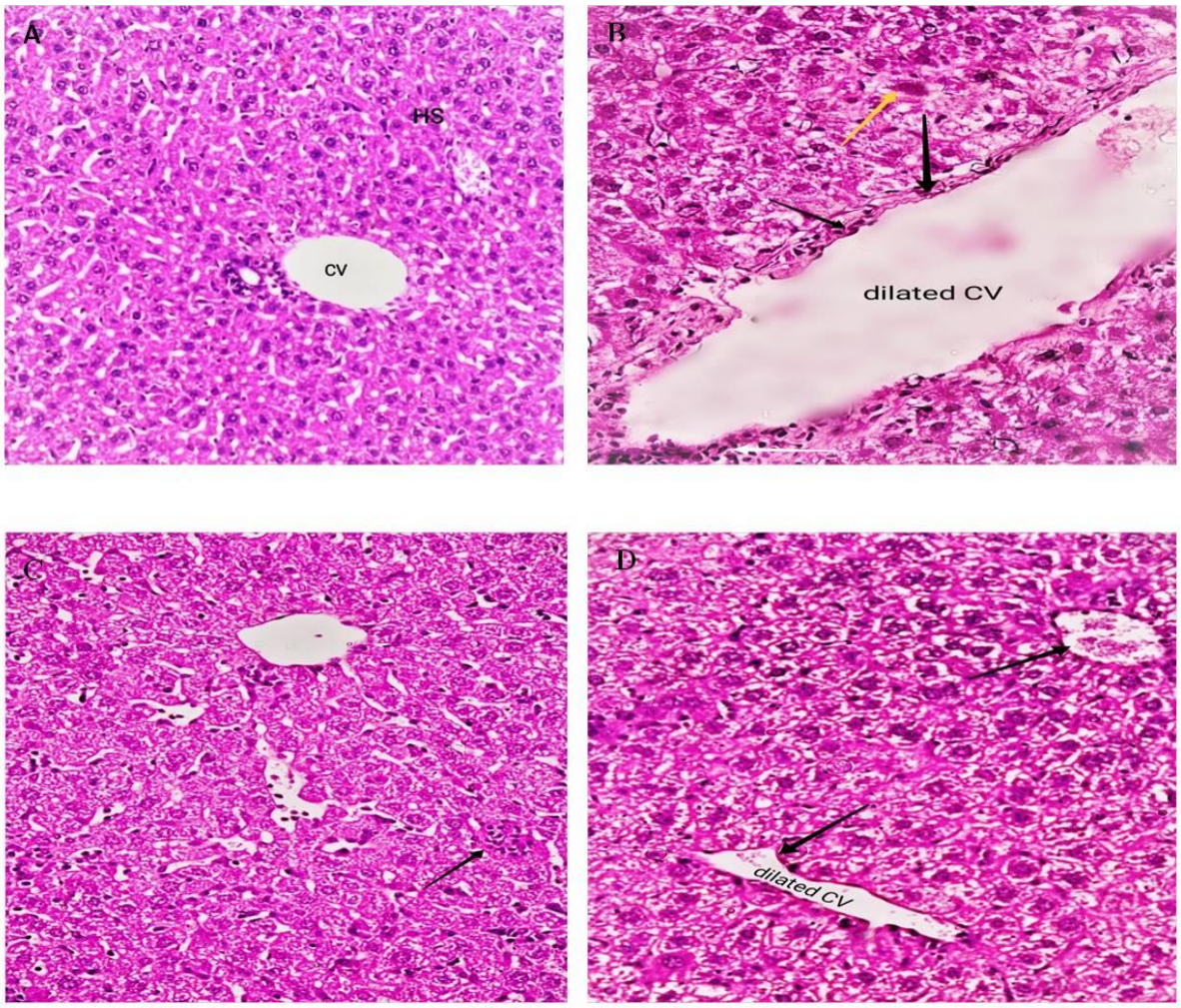


Fig.2. Representative histological images of the liver were stained with H&E. The original magnification was 200x .(A) The control group shows normal hepatic structure. (B) obese mice group shows distorted architecture with severe fatty liver change, the black arrow shows moderate fibrosis around the highly dilated central vein, the white arrow shows infiltration, and the yellow arrow shows Mallery denk bodies (MDB). (C) protective group shows restored normal hepatic structure. (D) curative group shows a moderately dilated central vein with slight hemorrhage.

## Discussion:

Obesity is a metabolic condition that results from significantly reduced consumption of calories in comparison to calorie intake. Instead of a simple increase in body weight, increased levels of adipose tissue are an indicator of obesity. Although the cause of obesity may appear to be straightforward, it involves several environmental and genetic factors, in addition to bad eating and exercise habits **(Lobstein, 2019)**. Healthy food consumption is regarded as crucial to protecting our bodies from the ailments that obesity can cause.

According to **Astrup (1993)** and **Bell *et al.* (1995)**, HFD promotes obesity in both humans and animal models, respectively. It is commonly acknowledged that a high dietary fat intake enhances the risk of obesity and chronic illnesses such as diabetes, hyperlipidemia, hypertension, cardiovascular disease, and several types of cancer **(Sanders, 2003)**.

In one instance, **Reaven (2000)** found a link between high fat consumption and the frequency of metabolic disorders in developed nations. In the current research, mice fed an HFD plus a carotenoid called astaxanthin were able to maintain their adipose tissue and body weight without modifying their energy consumption compared to the mice given a typical diet. After the studies, carotenoids such as astaxanthin helped to lower the rise in body weight Additionally, astaxanthin prevented the rise in lipid profile and glucose levels. Significant improvement in liver tissue antioxidant properties and function. According to these findings, Astaxanthin has shown an ameliorating impact on obesity and fatty liver resulting from a high-fat diet.

Astaxanthin is a naturally occurring carotenoid antioxidant that can be found in several kinds of different organisms. In obese mice consuming a high-fat diet, we noticed the effects of dietary astaxanthin supplementation. Astaxanthin inhibited the body weight and the weight of adipose tissue from rising. Furthermore, astaxanthin reduced plasma triglycerides and total cholesterol. According to these results, astaxanthin may be useful in lowering the risk of obesity and metabolic syndrome **(Ikeuchi *et al.*, 2007)**.

Evaluation of the carotenoid's anti-obesity effects in male Swiss albino mice, 5–6 weeks old, and 25–5 g in weight. Our research revealed that obese mice receiving HFD gained weight. However, A highly significant rise in body weight gain was seen in the curative group, whereas the protective group, which received carotenoid extract at the same time as eating HFD, revealed a slightly significant rise in body weight growth in contrast with the control group. This was because the carotenoid extract significantly lowered adipocyte fat formation and adipogenesis. According to **Lee *et al.* (2010),** increasing weight is the primary manifestation of obesity.

Consequently, the liver was chosen as the study's focus organ to determine how carotenoid extract affects the buildup of hepatic fat, which is accompanied by an increase in the size and quantity of adipocytes. Due to pathological alterations in liver steatosis, ROS production, and changes in liver enzyme activity, the liver is one of the key organs for fat storage **(Tiwari *et al.*, 2011)**. Transaminases (AST and ALT), particularly, are good biomarkers of liver damage with some degree of preserved liver function **(Johnston, 1999).** According to the data gathered from the results, G2 (obese mice) revealed a highly significant rise in ALT and AST enzymes in contrast to various groups. Compared to the control group, the protective group exhibits minimally significant increases. GGT is an enzyme that aids in the transport of peptides having the g-glutamyl group. GGT is present in the cell membranes of several organs, including the proximal renal tubule, liver, pancreas, intestine, and spleen (**Mukherjee and Gollan, 2011)**. Due to the fact, that GGT is a sensitive diagnostic of the existence of liver or bile duct injury, the GGT activities reported in this investigation demonstrated an extraordinarily high level of G2. However, because several non-hepatic

*Journal* **of** *Medical and* **Life Science**, *2023, Vol.5, No. 3, P.224-237* *pISSN: 2636-4093, eISSN: 2636-4107*

illnesses can induce elevation, its value is restricted by its lack of specificity **(Tinsay *et al.*, 2014)**. Similar observations have been reported by **Kim *et al.* (2018),** who observed that obese mice receiving HFD and BHe treatment had higher levels of ALT, AST, ALP, and GGT. Low serum albumin may be an indication of liver failure or conditions like cirrhosis or chronic hepatitis because albumin is generated in the liver **(Anderson and Douglas, 2000).** This can be considered as a good improvement for the liver and its synthetic functions capacity recovery. In this study, the level of albumin and total protein was significantly reduced in the protective and curative groups, respectively compared to the control mice group. As an indicator of hepatic disease, bilirubin analysis is one of the most important liver function tests **(Raymond and Galambos, 1971)**. A naturally occurring pigment called bilirubin is produced when heme-containing proteins break down **(Lester and Schmid 1964).** The liver and spleen's reticuloendothelial cells are in which bilirubin is primarily produced **(Tinsay *et al.*, 2014)**. Our results demonstrated that the content of bilirubin in serum had an extremely significant increase in the obese mice group compared to the control group, but the content of bilirubin level in the protective group showed no significant increase in contrast with control mice.

Obesity is characterized by the presence of chronic moderate inflammation, referred to as metaflammation, and is defined by an abnormal buildup of adipose tissue resulting in a body mass index (BMI) above 30 kg/m2 (**Khanna *et al.*, 2022**, and **Denizli *et al.*, 2022**). The main form of energy storage in adipose tissue is triglycerides (TG) which will then be stored in fatty tissue. Adipose tissue can grow (hypertrophy) over time, leading to larger fat cells and eventually, weight gain **(Chooi *et al.*, 2018)**. The presence of an excessive amount of triglycerides (TG) within adipocytes has the potential to result in pathological dysfunctions, including hepatic steatosis **(Frühbeck *et al.*, 2001)**. In the current investigation, the administration of sea urchin carotenoid was found to successfully reduce the elevated triglyceride (TG) levels observed during the adipogenesis of liver tissue induced by high-fat diet (HFD) feeding. The findings of this study indicate that the extracts have the potential to keep the liver against dysfunctions associated with hepatic steatosis. In addition, The administration of the extracts resulted in significant improvements in the levels of total cholesterol, high-density lipoprotein (HDL), and triglycerides (TG), as well as liver histology, when compared to the high-fat diet (HFD) group. The study found that the administration of carotenoid extract was more efficacious in modulating triglyceride levels in the bloodstream, as well as enhancing lipid profile and glucose metabolism. These results agreed with **Ikeuchi *et al.*, 2007** who stated that the observed effects of astaxanthin on body fatty tissue appear to be depending on the dosage administered. Additionally, astaxanthin decreased total cholesterol, liver, and plasma triglycerides. These results demonstrate that astaxanthin protects against the fatty liver and obesity that a high-fat diet can cause. Furthermore, this has the same outcome as **Kim *et al.*, 2020**, who reported elevated levels of total cholesterol. TG, and HDL in obese mice as a result of receiving HFD and an improvement in lipid profile when treated with color-fleshed sweet potato extracts. Hypolipidemia and hepatic steatosis are features of the obesity mouse model, which was produced by feeding the animals an HFD. HFD-fed animals are useful for research into metabolic syndrome prevention because they develop modest obesity and hyperglycemia. The present investigation found that carotenoid extract improved the level of glucose in the blood in both the protective group (G3) and the curative group (G4) of obese mice.

The excessive consumption of a high-fat diet (HFD) may be identified as a prevalent contributing factor to the development of obesity. As a result, lipid peroxidation and fatty acid oxidation will be dysregulated in obese mice (**Westerterp-Plantenga, 2006).** High levels of oxidative stress and inflammation, combined with a decrease in

*Journal* of *Medical and* Life Science, *2023, Vol.5, No. 3, P.224-237* *pISSN: 2636-4093, eISSN: 2636-4107*

antioxidant defense. All cases of obesity are associated with oxidative stress and inflammation and with increased levels of reactive oxygen species (ROS) and oxidative stress, which are considered harmful alterations and are common results **(Jiang *et al.*, 2021).** Mitochondria in adipocytes are an important source of reactive oxygen species (ROS) in obesity **(Tirichen *et al.*, 2021)**. Current research examined the effects of carotenoids on the liver of obese mice, looking for any evidence of their hepatoprotective, hypolipidemic, or anti-obesity properties. Anti-obesity activities of carotenoid extracts from marine Echinodermata sea urchin *Paracentrotus lividus* were evaluated in vivo using obesity models and antioxidant defense systems (lipid peroxidation and MDA content, GSH content, CAT, and SOD activity) were also assessed.

There are numerous enzymatic and non-enzymatic methods for reducing ROS and attempting to remove their effects. Glutathione (GSH), catalase, and superoxide dismutase (SOD) are the most important enzymes. Due to its ability to bind and eliminate ROS, GSH serves a crucial role in protecting cells from oxidative stress **(Kim and Ryu, 2013).** In our study, ten percent liver homogenate was prepared and used to measure these enzymes. Comparing G2, G3, and G4 to G1 revealed that oxidative stress developed frequently, as evidenced by the depletion of hepatic GSH, SOD, and catalase and the elevation of MDA, and nitric oxides. Increased hepatic lipid peroxidation and impairment of the endogenous antioxidant defense mechanisms were observed histologically in the HFD obese mice. This was accompanied by declines in liver CAT, SOD activity, and GSH levels. These findings are consistent with those from previous studies showing that HFD control mice had lower cytoplasmic content, increased hepatic lipid peroxidation, and deteriorated endogenous antioxidant defense systems, including lower liver CAT, SOD activities, and GSH content **(Kim *et al.*, 2018).** Similarly, these results agreed with **Naomi *et al.*, 2023** who noticed that obesity may cause the generation of ROS and inflammation due to the increased quantity of fat deposited in individual adipocytes. Oxidative stress will be generated as a result, and this could alter the tissue's morphology.

## Conclusion

In conclusion, Carotenoids extracted from the marine Echinodermata sea urchin *Paracentrotus lividus* were found to have anti-obesity effects. Mice administered a high-fat diet, and astaxanthin extract decreased the formation of adipose tissue and raised the availability of free fatty acids in the blood during the early phases of physical activity. During exercise, astaxanthin supplementation decreases the consumption of glucose and increases fatty acid utilization as an energy source also, preserving glycogen. The extracts showed potential for preventing the accumulation of fat in adipocytes, reducing weight gain, improving TG and total cholesterol levels, controlling blood sugar levels, and liver function tests as well so that astaxanthin can promote an increase in fatty acid usage.

## Conflict of interest:

There are no conflicts of interest

## Ethical approval:

Ethical approval was obtained from the Institutional Animal Care and Use Committees Of Damanhour University, Egypt With Code Number DMU-SCI-CSRE- 23 02 03.

## References :

**Aebi, H. (1984).** Colorimetric analysis of catalase assay methods enzymol. 105, 121- 126.

**Anderson, D., and Douglas, M. (2000).** Dorland's illustrated medical dictionary (29ed). Philos. Saunders., 860.

**Astrup, A. (1993).** Dietary composition, substrate balances and body fat in subjects with a predisposition to obesity. Int. J. Obes., 17, s32–s36.

**Bell, R. R., Spencer, M. J., and Sherriff, J. L. (1995).** Diet induced obesity in mice can be treated without energy restriction using

exercise and/or a low fat diet. J. Nutr., 125, 2356–2363.

**Bennedsen, M., Wang, X., Willen, R., Wadstroem, T., and Andersen, L. P. (1999).** Treatment of H. pylori infected mice with antioxidant astaxanthin reduces gastric inflammation, bacterial load and modulated cytokine release by splenocytes. Immunol. Lett., 70, 185–189.

**Beutler, E. , Duron, O. , and Kelly, M.B. ( 1963 ).** colorimetric analysis of glutathione reduced, J. Lab. Clin. Med. 61, 882.

**Buchwaldt, M., and Jencks, W.P. (1968).** Optical properties of astaxanthin solutions and aggregates. Biochemistry-US., 2: 834-843.

**Chen, B.H., and Yang, S.H. (1992).** An improved analytical method for determination of carotenes and xanthophylls in dried plant materials and mixed feeds. Food Chem., 44: 61-66.

**Chew, B. P., Park, J. S., Wong, M. W., and Wong, T. S. (1999).** A comparison of the anticancer activities of dietary beta-carotene, canthaxanthin and astaxanthin in mice in vivo. Anticancer Res., 19, 1849–1853.

**Chooi, Y.C., Ding, C., and Magkos, F. (2018).** The epidemiology of obesity. Metabolism, 92, 6 10.

**Denizli, M., Capitano, M.L., and Kua, K.L.(2022).** Maternal obesity and the impact of associated early-life inflammation on long-term health of offspring. Front. Cell. Infect. Microbiol. 12, 1348.

**Doumas, B.T., Biggs, H. G., and Watson, W. A. (1971).** Albumin Standards and the Measurement of Serum Albumin with Bromcrescol Green. Clin. Chim. Acta., 31-87.

**Frühbeck, G., Gómez-Ambrosi, J., Muruzábal, F. J., and Burrell, M. A. (2001).** The adipocyte: A model for integration of endocrine and metabolic signaling in energy metabolism regulation. *American Journal of Physiology*, *280*(6), E827–E847. https://doi.org/10.1152/ajpen do.2001.280.6.E827.

**Fukuhara, K., Inokami, Y., Tokumura, A., Terao, J., and Suzuki, A., (1998).** Rate constants for quenching singlet oxygen and activities for inhibiting lipid peroxidation of carotenoids and alpha-tocopherol in liposomes. Lipids, 33, 751–756.

**Gabin-Sánchez, C., Lorenzo, D., D. F. (1993).** El erizo de mar un recurso con futuro. Aula del Mar Fundación Caixa Galicia, Spain, pp 8–9

**Guerin, M., Huntley, M. E., and Olaizola, M. (2003).** Hematococcus astaxanthin: applications in human health and nutrition. Trends Biotechnol., 21, 210–216.

**Guill, Michael. "** Taxonomic Etymologies EEOB 111" Retrieved 13 March 2018.

**IFCC (1986).** IFCC methods for measurement of enzymes, IFCC methods for gamma-glutamyltransferase [(gamma glutamyl)-peptide: amino acid gamma glutamyl transferase, EC 2.3.2.2. Clin. Chim. Acta., 135: 315-338.

**Ikeuchi, M., Koyama, T., Takahashi, J., and Yazawa, K. (2007).** Effects of Astaxanthin in Obese Mice Fed a High-Fat Diet. Biosci. Biotechnol. Biochem., 71 (4), 893–899.

**Jiang, S., Liu, H., and Li, C. (2021).**Dietary Regulation of Oxidative Stress in Chronic Metabolic Diseases. Foods 10, 1854.

**Johnston, D.E. (1999).** Special considerations in interpreting liver function tests. Am Fam Physician;59:2223–2230.

**Kelly, M.S. ,Owen, P. V., and Pantazis, P. (2001).** The commercial potential of the sea urchin *Echinus esculentus* from the west coast of Scotland Hydrobiologia 465(1);85-94.

**Khanna, D. , Khanna, S., Khanna, P., Kahar, P., and Patel, B.M. (2022).** Obesity A Chronic Low-Grade Inflammation and Its Markers. Cureus 14, e22711.

**Kim , H. J., Koo1,K.A. , Park,W.S., Kang,D.M., Kim,H.S., , Bo Young Lee,B.Y.| Goo,Y.M., Kim,J.H., Lee,M.K., Dong Kyun Woo,D.K., Kwak,S.S., Ahn,M.J. (2020).** Anti-obesity activity of anthocyanin and carotenoid extracts from color-fleshed sweet potatoes J Food Biochem. 2020;00:e13438.

**Kim, S., and Ryu, D.Y. (2013).** Silver nanoparticle-induced oxidative stress, genotoxicity and apoptosis in cultured cells and animal tissues. J. Appl. Toxicol., 33: 78–89.

**Kim, S.G., Woo, S.O., Bang, K.W., Jang, H.R., Han, S.M.(2018).** Chemical composition of drone pupa of Apis mellifera and its nutritional evaluation. J Apiculture ;33(1): 17–23. https://doi.org/10.17519/apiculture.2018.04.33.1.17.

**Langi, P., Kiokias, S., Varzakas, T.,and C. Proestos, C. (2018).** Carotenoids: From Plants to Food and Feed Industries, Methods Mol. Biol., 1852, 57–71.

**Larsson, B., Svärdsudd, K., Welin, L., Wilhelmsen, L., Björntorp, P., & Tibblin, G. (1984).** Abdominal adipose tissue distribution, obesity, and risk of cardiovascular disease and death: 13 year follow up of participants in the study of men born in 1913. *British Medical Journal*, *288*(6428), 1401–1404. https://doi.org/10.1136/bmj.288.6428.1401

**Lee, J. M. , Pilli, S., Gebremariam, A., Keirns, C. C., Davis, M. M., Vijan, S., and Gurney, J. G., (2010).** Getting heavier, younger: Trajectories of obesity over the life course. International Journal of Obesity, 34(4), 614–623.

**Lester, R., and Schmid, R. (1964).** Bilirubin metabolism. N. Engl. J. Med., 270:779– 86.

**Lobstein, T. (2019).** Obesity prevention and the global syndemic: Challenges and opportunities for the World Obesity Federation. Obesity Reviews, 20(S2), 6–9. https://doi.org/10.1111/obr.12888

**Minguez-Mosquera, M.I .,and Hornero-Mendez, D . (1993).** Separation and quantification of the carotenoid pigments in red peppers (Capsicum annuum L paprika, and oleoresin by reverse-phase HPLC. J. Agr. Food Chem., 41(10): 1616–1620.

**Montgomery, H. A . C. and Dymock, D. ( 1961 ).** The determination of nitrite in water. Analyst, J . F ., 86: 414-416.

**Mukherjee, S. and Gollan, J.L. (2011).** Assessment of liver Function. In: Lok AS, Dooley JS, Burroughs AK (editors). Sherlock's diseases of the liver and biliary System. Wiley-Blackwell, pp.20-35.

**Naguib, Y. M. (2000).** Antioxidant activities of astaxanthin and related carotenoids. J. Agric. Food Chem., 48, 1150– 1154.

**Naito, Y., Uchiyama, K., Aoi, W., Hasegawa, G., Nakamura, N., Yoshida, N., Maoka, T., Takahashi, J., and Yoshikawa, T. (2004).** Prevention of diabetic nephropathy by

treatment with astaxanthin in diabetic db/db mice. BioFactors, 20, 49–59.

**Naomi, R., Teoh, S.H. , Embong, H. , Balan, S. S. , Othman, F. , Bahari, H. , and Muhammad Dain Yazid, M. D. (2023).** The Role of Oxidative Stress and Inflammation in Obesity and Its Impact on Cognitive Impairments—A Narrative Review. Antioxidants 2023, 12, 1071. https://doi.org/10.3390/antiox12051071.

**Nishikimi, M., Roa, N.A., and Yogi, K. ( 1972 ).** The occurrence of superoxide anion in the reaction of reduced phenazine methosulfate and molecular oxygen. Biochem. Bioph. Res. Common., 46(2):849-54. 46, 849 – 854.

**Ohkawa, H. , Ohishi W, and Yagi K. ( 1979 ).** Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. Anal. Biochem., 95(2):351-8.

**Park, S.H., Ko, S.K., and Chung, S.H.(2005).** Euonymus alatus prevents the hyperglycemia and hyperlipidemia induced by high-fat diet in ICR mice. J Ethnopharmacol 102: 326-335.

**Raymond, G.D. and Galambos, J.T. (1971).** Hepatic storage and excretion of bilirubin in man. Am. J. Gastroenterol., 55(2):135-144.

**Reaven, G. M. (2000).** Diet and syndrome X. Curr. Atheroscler. Rep., 2, 503–507.

**Reitman, A., and Frankel, S. ( 1957).** A colorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. Amer. J. Clin. Path., 28(1):56-63.

**Sanders, T. A. (2003).** High versus low fat diets in human disease. Curr. Opin. Clin. Nutr. Metab. Care, 6, 151–155.

**Satoh, K. ( 1978 ).** Colorimetric method test for malondialdehyde (MDA). Clinica. Chimica. Acta., 90, 37.

**Symonds, R., Kelly, M., Caris-Veyrat, C. Young, A. (2007).** Carotenoids in the sea urchin *Paracentrotus lividus*: Occurrence of 9'-cis-echinenone as the dominant carotenoid in gonad colour determination. *Comp Biochem Physiol Part B: Biochem Mol Biol* 148: 432–444.

**Symonds, R., Kelly, M., Suckling, C., Young, A. (2009).** Carotenoids in the gonad and gut of the edible sea urchin *Psammechinus miliaris*. *Aquaculture* 288: 120–125.

**Tan, Y., Kim, J., Cheng, J., Ong, M., Lao, W.G., Jin, X.L., Lin, Y.G., Xiao, L., Zhu, X.Q., and Qu, X.Q. (2017).** Green tea polyphenols ameliorate non-alcoholic fatty liver disease through upregulating AMPK activation in high fat fed Zucker fatty rats. World J Gastroenterol 23: 3805-3814.

**The World Health Organization (WHO).** Obesity and Overweight; WHO Media Center: Geneva, Switzerland, 2012.

**Tinsay, A., Woreta, M.D., Saleh, A., and Alqahtani, M.D. (2014).** Evaluation of Abnormal Liver Tests. Med. Clin. N. Am. 98: 1–16.

**Tirichen, H.; Yaigoub, H.; Xu, W.; Wu, C.; Li, R.; Li, Y.(2021).** Mitochondrial Reactive Oxygen Species and Their Contribution in Chronic Kidney Disease Progression Through Oxidative Stress. Front. Physiol. 12, 627837.

**Tiwari, D. K., Jin, T., and Behari, J. (2011).** Dose-dependent in-vivo toxicity assessment of silver nanoparticle in Wistar rats. Toxicol. Mech. Methods, 21:13–24.

*Journal* **of** *Medical and* **Life Science***, 2023, Vol.5, No. 3, P.224-237* *pISSN: 2636-4093, eISSN: 2636-4107*

**Trinder, P. (1969).** Enzymatic determination of glucose in blood serum. Annals of Clinical Biochemistry, 6, 24.

**Unuma, T., Nakamura, A., Yamano, K., Yokota, Y. (2010).** The sea urchin major yolk protein is synthesized mainly in the gut inner epithelium and the gonadal nutritive phagocytes before and during gametogenesis. *Mol Rep Dev* 77: 59–68.

**Walter, M. , and Gerade, H. (1970).** colorimetric analysis of total bilirubin, Microchem. J., 15. 231.

**Westerterp-Plantenga, M.S.(2006).** Specific fatty acids and structured lipids for weight control. In Improving the Fat Content of Foods; Williams, C., Buttriss, J.,vEds.; Woodhead Publishing: Berkshire, UK; pp. 162–181, ISBN 9781855739659.