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Abstract

This study was conducted to investigate the effect of different levels of the red dragon (*Hylocereus polyrhizus*) fruit powder (2%, 4%, 6%) on hypercholesterolemic male albino rats. The study included 30 white male albino rats, weighing about (140 ± 10g), which were divided into (5) equal groups, each group (6) rats, one was kept as control (-ve) group, while the other (4) groups were induced hypercholesterolemic by the injection of Triton-x-100 (100 mg/kg body weight of the rat). Some biochemical analyses including, lipid profiles, atherogenic index (AI), glucose level, liver enzymes, kidney functions and hematological parameters have been determined. Also, the identification of phenolics compounds of the red dragon was done by using the HPLC method. The obtained results indicated that red dragon fruit powder contains different amounts of phenolics compounds, the highest levels recorded for catechin, chlorogenic acid, synergic acid, *p*-coumaric acid, ferulic acid. Data also showed a significant decrease in liver enzymes, kidney functions, serum glucose level and hematological parameters, lipid profiles (TC, TG, LDL-c, VLDL-c) in all treated groups, as compared to the positive control group, while HDL-c level increased. Therefore, red dragon fruit powder especially with a 6% level improved all tested biochemical analyses and could be considered therapeutic means for the treatment of hypercholesterolemia rats.

Keywords: un-common fruit, Hypercholesterolemia, Phenolic compounds, Biochemical analysis.

INTRODUCTION

Hyperlipidemia is a condition of increased blood lipid levels characterized by increased levels of total cholesterol, low-density lipoprotein (LDL), and triglycerides in the blood that exceed normal limits. The normal condition of total cholesterol is amounting to 10 - 54 mg/dl (**Agustina, 2014)**. Then in a study conducted by **Bashandy (2007),** it was reported that the normal condition of triglycerides in rats was 27.89-29.44 mg/dl, and a study conducted by **Gani *et al*., (2013)** reported that the normal threshold for LDL in mice was 7 - 27.2 mg/dl. Hyperlipidemia is considered one of the major risk factors causing cardiovascular diseases (CVDs). CVDs account for one-third of total deaths around the world, it is believed that CVDs will turn out to be the main cause of death and disability worldwide by the year 2020 **(Jorgensen *et al*., 2013)**.

Lipids are compounds that have an important role in cell structure and function. The main plasma lipids consist of cholesterol, triglycerides, phospholipids, free fatty acids. These hydrophobic lipids in the circulation are in the form of a lipid-protein or lipoprotein complex. Plasma lipoproteins consist of Kilomicrons, very low-density lipoprotein (VLDL), LDL, and high-density lipoprotein (HDL). The composition and function of each lipoprotein are different **(Guyton and Hall, 2007)**. Obesity contributed to double the burden of diseases particularly diabetes (44%), ischemic heart diseases (23%), and certain types of cancer (7-41%) **(BullA *et al*., 2007**).

Hyperlipidemia that occurs in the body is a free radical that will cause the formation of lipid peroxidation products such as malondialdehyde (MDA). MDA levels are widely used as biomarkers for assessing oxidative stress in the biomedical field. Lipid peroxidation is an indicator of tissue damage caused by free radical activity **(Zorawar Singh, 2014**). A high-fat diet in mice can lead to hypercholesterolemia, which plays an important role in increasing the production of free radicals and the mismatch of lipid peroxide development at the tissue level, which will cause changes in spermatozoa morphology **(Harini, 2009)**. The increase in cholesterol levels plays a role in producing free radicals which are accelerated by oxidative stress reactions. Reactive oxygen

species (ROS) can cause damage to biological macromolecules including oxidation of low-density lipoproteins (oxidized-LDL), triglycerides, endothelial dysfunction and an increase in inflammatory responses that originate from oxidation of unsaturated fatty acids in the lipid layer of cell membranes. This reaction initiates a chain of lipid oxidation which will cause damage to the cell membrane **(Agustina, 2014)**.

Antioxidants are compounds that can inhibit the fat oxidation process. Antioxidants stabilize free radicals by complementing the lack of electrons that free radicals have and will inhibit the chain reaction from forming free radicals. If the formation of free radicals is inhibited, the motility of spermatozoa in obese men will improve (**Hor *et al.*, 2012).** One study shows that the nutritional components of fruits and vegetables can lower cholesterol levels. Currently there are more and more studies on fruits that have a high antioxidant content, one of which is red dragon fruit (**Indriasari, 2012)**. Eating fruits and vegetables can ensure an adequate supply of micronutrients, dietary fibers and phytochemicals which in turn maintain the body in a healthy state **(World Health Organization, 2013)**.

Dragon fruit or pitaya is a plant that belongs to the *Cactaceae* family. It grows widely in tropical areas such as Southeast Asia, Mexico, Cambodia, Indonesia, Australia and the United States. It has widely used in the fields of food, therapy and medicine **(Mihir, 2019)**. Red pitaya fruit is organically grown without the use of any pesticide and chemical fertilizers. Red pulp of pitaya fruit has generated a lot of interest as a source of natural red color for the food coloring, cosmetic industry and health potential for improving eyesight and preventing hypertension and combat anemia (**Stintzing *et al.*, 2002)**. The red dragon fruit, also known as the *Hylocereus polyrhizus*, has recently attracted attention stemming from farmers and consumers, and it is now found in exotic fruit markets around the world **Heryani, (2016)** not only for its famously attractive purple-red color or its economic value as products but also for its highly active biological compounds that include antioxidant, anti-inflammatory, anti-microbial **(Febrianti *et al.*, 2018)**. Red dragon fruit is a plant that is used as a source of antioxidants. It is believed to reduce

cholesterol levels, balance blood sugar levels, prevent colon cancer, and increase fertility. In addition, red dragon fruit seeds also contain unsaturated fats which are needed in the maturation process of spermatozoa **(Ortiz *et al*., 2012)**.

It also has the ability to inhibit the growth of cancer cells and has an anti-diabetic **Kim *et al*., (2011)** as well as an anti-atherosclerotic effect. Moreover, it has the ability to protect the liver and kidneys (reno and hepato-protection according to **(Hernawati *et al*., 2018)**. The importance of effective phytochemical bioactive compounds in the pulp of the red dragon fruit like polyphenols, flavonoids, and vitamins A, C & E. As they are antioxidants, they are able to assist with the balancing of oxidative stress, which the body is constantly exposed to the environmental and food system, especially foods with food additives, including preservatives **(Armutcu *et al*., 2018)**.

Red dragon fruit has various ingredients like saponins, and triterpenoids that work to inhibit HMG-CoA reductase. Phenol, betacyanins and ascorbic acid neutralize free radicals and peroxide radicals so that oxidative stress decreases. Flavonoids directly donate hydrogen ions to stabilize free radicals and indirectly stimulate antioxidant gene expression. In addition, flavonoids increase the secretion of the bile that can reduce cholesterol levels in the body **(Shi *et al*., 2014)**.

Lugo-Radillo *et al*., (2012) proved that dried dragon fruit with a dose of 9.6 mg/kg BW/day for 28 days in mice can improve lipid profile in dyslipidemia. All hypercholesterolemic groups that received red pitaya supplementation (0.5%, 0.38% and 1.17% daily diet) have high antioxidant properties and showed a good result in managing lipid profile. It was suggested that the consumption of red pitaya demonstrated the potential to reduce dyslipidemia and play a role in the prevention of cardiovascular disease **(Khalili *et al*., 2009)**.

Therefore, this study was conducted to investigate the potential effect of the red dragon fruit on hypercholesterolemic rats.

Material and Methods

Materials

Fresh samples of the red dragon (*Hylocereus polyrhizus*) were purchased in February 2021 from Agriculture Research Center, Giza, Egypt.

The chemical Triton X-100 was used to induce hypercholesterolemia obtained from SIGMA Chemicals Co., Cairo, Egypt.

Chemicals and kits

Pure white crystalline cholesterol powder and saline solutions casein, cellulose, choline chloride powder, and DL-methionine powder, were obtained from Morgan Co. Cairo, Egypt, oil and corn starch were obtained from a local market in Assiut, Egypt. Chemical kits used in this study (TC, TG, HDL-c, ALT, AST, ALP, urea, creatinine and uric acid) were obtained from Al-Gomhoria Company for Drugs, Chemicals and Medical Supplies, Cairo, Egypt.

Experimental animals

A total of 30 adult normal male albino rats Sprague Dawley strain weighing 140 ± 10g were obtained from the animal house of the Faculty of Medicine, Assiut University.

Methods

Preparations of the red dragon

Fresh samples of the red dragon were washed thoroughly under running tap water, cut into small slices. Slices were summarizing dried in an oven at 55^{o}C for 8 hours to save the phenol compounds as they are until constant moisture level and ground to a fine homogenous powder using an air mill, the high-speed mixture (Molunix, Al-Araby Company, Benha, Egypt), then serving as powder seize, and kept in polyethylene bags at freezing temperature until using.

Experimental design

Thirty male albino rats, weighing 140 ± 10g, were used in this experiment. All rats were fed on the basal diet (casein diet) prepared according to **AIN, (1993)** for 7 consecutive days. After this adaptation period, rats are divided into 5 groups, each group which consists of six rats as follows: group (1): rats fed on basal

diet as a negative control. Group (2): A group injected by a single dose of freshly prepared solution of Triton-X-100 (100 mg/kg) and used as a positive control group. Group (3): A group infected by hypercholesterolemia fed on a basal diet containing 2% of the red dragon as powder. Group (4): A group infected by hypercholesterolemia fed on a basal diet containing 4% of the red dragon as powder. Group (5): A group infected by hypercholesterolemia fed on a basal diet containing 6% of the red dragon as powder. The experiment period was taking 28 days, at the end of the experimental period each rat weighed separately then, rats are slaughtered.

Blood sampling

Blood samples were collected after 12 hours of fasting at the end of the experiment. Using the retro-orbital method by means of microcapillary glass tubes, blood was collected into a dry clean centrifugal tube and left to clot in a water bath (37°C) at room temperature for half an hour. The blood was centrifuged for 10 minutes at 3000 rpm to separate the serum in a clean glass well stoppered and stored at and kept (-20°C) until analysis **(Schermer, 1967)**.

Identification of phenolic compounds using HPLC.

Extraction, separation and quantification of phenolic compounds were determined according to the method described by **(Goupy *et al.*, 1999)**. The HPLC system Perkin Elmer PE200 was composed of a binary pump, a column thermostat, and an autosampler. A bin pump model G1312A, an auto-sampler model G1313A and a RR Zorbax Eclipse Plus C18 column (1.8 µm, 150 mm ×4.6 mm). Mobile phase A was 0.2 % formic acid in water and mobile phase B was acetonitrile. Quantification of the compounds was realized using calibration curves obtained by HPLC of pure standards: gallic acid, caffeic acid, (+)-catechin, (-)-epicatechin, and ellagic acid. Rutin was used as an internal standard. Some compounds were quantified as equivalents of the most similar chemical structures: gallic acid for gallic acid glucoside, gentisic acid glucoside, protocatechuic acid, p-hydroxybenzoic acid and methyl gallate; caftaric acid as caffeic acid; (+)- -catechin for proanthocyanidin dimers and trimers and

their monogallates; (–)-epicatechin for epicatechin gallate; ellagic acid for ellagic acid pentoxide.

Biochemical analysis

Serum lipids profile

Serum total cholesterol was determined according to the colorimetric method described by **(Thomas, 1992)**. Serum triglycerides were determined by the enzymatic method using kits according to the **(Young and Pestaner, 1975)** and **(Fossati & Principle, 1982)**. HDL-c was determined according to the method described by **(Grodon and Amer, 1977)**.

VLDL-c was calculated in mg/dl according to **Lee and Nieman (1996)** was using the following formula: **VLDL-c (mg/dl) = Triglycerides / 5**

LDL-c was calculated in mg/dl according to **Lee and Nieman (1996)** as follows:

LDL-c (mg/dl) = Total cholesterol – HDL-c – VLDL-c.

Calculation of atherogenic index (AI)

This index was calculated as the (VLDL-c+ LDL-c/HDL-c ratio according to the formula of **(Kikuchi-Hayakawa *et al.*, 1998)**.

Liver enzymes

Determination of serum alanine aminotransferase (ALT), serum aspartate aminotransferase (AST) and serum alkaline phosphatase (ALP) were carried out according to the method of **(Srivastava *et al.*, 2002); (Chawla, 2003) and (Huang *et al.*, 2006)**; respectively.

Kidney functions

Serum uric acid was determined calorimetrically according to the method of **(Barham** and **Trinder, 1972)**. Serum urea was determined according to the enzymatic method of (**Patton** and **Crouch, 1977)**. Creatinine was determined according to the kinetic method of **(Henry, 1974)**.

Serum glucose

Enzymatic determination of plasma glucose was carried out calorimetrically according to the method of **(Wang *et al.*, 2010)**.

Determination of RBC, WBC, Hb and PLT

The concentration of hemoglobin (Hb), red blood cell count (RBC), white blood cell (WBC) and platelets (PLT) were estimated according to the method described by **(Dacie and Lewis, 1998)**.

Statistical analysis

The data were analyzed using a completely randomized factorial design **SAS, (1988)** when a significant main effect was detected; the means were separated with the Student-Newman-Keuls test. Differences between treatments at $P \leq 0.05$ were considered significant using Costas Program. Biological results were analyzed by One Way ANOVA.

Results and Discussion

Data presented in Table (1) showed the identification of phenolic compounds of red dragon fruit. It is clear to notice that the highest phenolic compounds in red dragon fruit recorded for catechin, chlorogenic acid, synergic acid, *p*-coumaric acid and ferulic acid. The values were 7.42, 4.15, 3.0, 2.78 and 2.60 mg/g DW; respectively. On the other hand, the lowest phenolic compound was recorded for *p*-hydroxybenzoic acid and gallic acid. The values were 1.25 and 1.75 mg/g DW; respectively. While caffeic acid, sinapic acid and protocatechuic acid were not detected at these conditions. These findings support by **Wu *et al.*, (2006)**, who found that phenolic acids are the most abundant bioactive compounds in various fruits. Both the flesh and the peel of the red dragon were high in phenolics and antioxidants. The antioxidant ability of red dragon fruit was discovered to be primarily dependent on betalains, followed by their biosynthetic precursors, and then non- betalain phenolics including gallic acid and acetylcoumarin **(Esquivel *et al*., 2007).**

Data tabulated in Table (2) show the effect of red dragon fruit as powder on the serum total cholesterol (TC) and triglycerides (TG) of hypercholesterolemic rats. The obtained results indicated that the total cholesterol levels of the positive control group recorded a higher value when compared with the negative control group with a significant difference ($P \leq 0.05$). The mean values were 240 and 95 mg/dl; respectively. On the other

hand, the lowest total cholesterol levels of treated groups (hypercholesterolemic rats) were recorded for the group fed on 6 % red dragon fruit powder, while the highest value was recorded for 2% red dragon fruit powder with a significant difference (P≤0.05). The mean values were 112 and 167 mg/dl; respectively.

On the other hand, the obtained results indicated that triglycerides of the positive control group recorded higher values when compared with the negative control group with a significant difference (P≤0.05). The mean values were 190 and 105 mg/dl; respectively. All treated groups showed significant decrease in the mean values of serum triglycerides, as compared to the positive control group. The lowest triglycerides level of treated groups (hypercholesterolemic rats) was recorded for the group fed on 6 % red dragon fruit powder, while the highest value was recorded for 2% red dragon fruit powder with a significant difference (P≤0.05). The mean values were 115 and 152 mg/dl; respectively. Hyperlipidemia, including hypercholesterolemia and hypertriglyceridemia, is a significant risk factor for the development of cardiovascular disease, according to **(Robert and Nelson, 2013)**. In the early stages of atherosclerotic fatty streak lesions, macrophages pick up oxidatively damaged LDLs, which accumulate in the endothelial wall as lipid-laden foam cells. As a result, lowering circulating TGs, total cholesterol, and LDLs is critical for preventing vascular disease.

Fiber, vitamin C, and anthocyanins contained in the fruit and peel of the red dragon fruit are likely to induce a reduction in total cholesterol levels **(Ide, 2009)**. The fibers bind bile in the intestine, causing bile salts to be excreted from the enterohepatic cycle and wasted with the feces, resulting in a reduction in bile salts and exogenous cholesterol levels. As a result, the liver can use endogenous cholesterol as a source of bile salts (**Murray *et al.*, 2012)**.

Data in Table (3) show the effect of red dragon fruit as powder on serum lipoproteins and atherogenic index of hypercholesterolemic rats. The obtained results indicated that the HDL-c of the negative control rats group recorded a higher value when compared with the positive control group with significant difference (P≤0.05). The mean values were 60 and 32 mg/dl;

respectively. While the highest mean value of serum HDL-c in all treated groups was recorded for the group fed on 6% red dragon fruit powder but, the lowest value recorded for the group fed on 2% red dragon fruit powder with a significant difference (P≤0.05). The mean values were 54 and 38 mg/dl; respectively. On the other hand, the LDL-c of the positive control rats group recorded a higher value when compared with the negative control group with a significant difference (P≤0.05). The mean values were 170 and 14 mg/dl; respectively. While the highest LDL-c of the treated group was recorded for the group fed on 2 % red dragon fruit powder but, the lowest value was recorded for the group fed on 6% red dragon fruit powder with a significant difference (P≤0.05). The mean values were 98.60 and 35 mg/dl; respectively. In the case of VLDL-c, the positive control rats group recorded a higher value when compared with the negative control group with a significant difference (P≤0.05). The mean values were 38 and 21 mg/dl; respectively. While the highest VLDL-c of the treated group was recorded for the group fed on 2 % red dragon fruit powder but, the lowest value recorded for the group fed on 6% red dragon fruit powder with a significant difference (P≤0.05). The mean values were 30.40 and 23 mg/dl; respectively.

The obtained results also indicated that the atherogenic index (AI) of the positive control rats group recorded a higher value when compared with the negative control group with significant difference (P≤0.05). The mean values were 6.50 and 0.58 %; respectively. While the highest atherogenic index of the treated group was recorded for the group fed on 2% red dragon fruit powder but, the lowest value recorded for the group fed on 6% red dragon fruit powder with a significant difference (P≤0.05). The mean values were 3.39 and 1.07 %; respectively.

Since anthocyanin in dragon fruit inhibits cholesteryl ester transfer protein activity (CETP), there is no exchange between HDL cholesterol ester in triglyceride in LDL, a decrease in LDL cholesterol is suspected. As a result of HDL3 not converting to HDL2, HDL cholesterol rises when LDL cholesterol falls. This will increase cholesterol clearance in the periphery, allowing it to be transported to the liver and then removed by bile acid secretion,

preventing lipoprotein oxidation and thus reducing LDL cholesterol oxidation **(Murray *et al.*, 2012)**.

Ferulic acid, which can be contained in red dragon fruit, can help rats have lower levels of low-density lipoproteins. They also said that ferulic acid inhibited hydroxy methyl glutaryl coenzyme A reductase (HNG-CoA reductase) and thus reduced cholesterol synthesis. This enzyme is the most powerful regulator in cholesterol biosynthesis **(Kim *et al.*, 2003)**.

Soluble fiber, unsaturated fatty acids, and minerals, especially potassium, sodium, magnesium, phosphorus, and zinc have been shown to have a hypocholesterolemic effect via increased bile acid excretion in red dragon **(Khalili *et al.*,2006)**.

The red pitaya fruit, which is high in phenolics and antioxidants, has an important impact on the lipid metabolism of rats. The red pitaya supplement diet has the ability to lower TC, TG, and LDL-C levels while increasing HDL-C. Because of its strong antioxidant activity and phenolic content, red pitaya supplementation in the diet may be helpful in the prevention of dyslipidemia **(Khalili *et al.*, 2009)**.

The effect of red dragon fruit as powder on the serum glucose level of hypercholesterolemic rats is shown in Table (4). It is clear to mention that the positive control rats group recorded a higher value when compared with the negative control group with a significant difference ($P \leq 0.05$). The mean values were 180 and 96 mg/dl; respectively. While the highest glucose level of the treated group was recorded for the group fed on 2 % red dragon fruit powder but, the lowest value recorded for the group fed on 6% red dragon fruit powder with a significant difference ($P \leq 0.05$). The mean values were 140 and 119 mg/dl; respectively. These findings support **Choo and Yong**'s**, (2011)** findings that dragon fruit is high in natural antioxidants such as betacyanin, flavonoids, phenolic acid, ascorbic acid, and fiber. It has a preventive impact on the histopathological picture of pancreatic cells in alloxan-induced diabetes rats by reducing reactive oxidative species, thanks to its high antioxidant and free radical scavenging function **(Ismaviani, 2014)**.

Since dragon fruit's glucose-lowering effect is thought to be due to betacyanin and antioxidant activity, the efficacy of red and white flesh dragon fruit may differ **(Suh *et al.*, 2014)**.

Data given in Table (5) show the effect of red dragon fruit powder on liver enzymes (ALT, AST and ALP) of hypercholesterolemic rats. The obtained results indicated that the ALT liver enzyme of the positive control rats group recorded a higher value when compared with the negative control group with a significant difference ($P \leq 0.05$) which were 23 and 12 U/L; respectively. While the highest ALT liver enzyme of all treated groups was recorded for the group fed on 2 % red dragon fruit powder but, the lowest value was recorded for the group fed on 6% red dragon fruit powder with a significant difference ($P \leq 0.05$). The mean values were 19 and 15 U/L; respectively.

On the other hand, the AST liver enzyme of the positive control rats group recorded a higher value when compared with the negative control group with a significant difference ($P \leq 0.05$). The mean values were 95 and 42 U/L; respectively. While the highest AST liver enzyme of the treated group was recorded for the group fed on 2 % red dragon fruit powder but, the lowest value was recorded for the group fed on 6% red dragon fruit powder with a significant difference ($P \leq 0.05$). The mean values were 61 and 41 U/L; respectively.

As for the ALP liver enzyme, data indicated that the ALP liver enzyme of the positive control rats group recorded a higher value when compared with the negative control group with a significant difference ($P \leq 0.05$). The mean values were 135 and 90 U/L; respectively. While the highest ALP liver enzyme of the treated group was recorded for the group fed on 2 % red dragon fruit powder but, the lowest value was recorded for the group fed on 6% red dragon fruit powder with a significant difference ($P \leq 0.05$). The mean values were 119 and 104 U/L; respectively.

Both the crude and ethanolic extracts of the red dragon have been found to protect the liver from carbon tetrachloride (CCl_4)-induced hepatic damage. However, as compared to ethanolic extract, the crude extract has a stronger hepatoprotective effect against Carbon Tetrachloride (CCL_4)-induced hepatic damages **(Cauilan, 2019)**.

In alcoholic Liver disease (ALD) caused by chronic ethanol exposure, red pitaya supplementation can reduce hepatic steatosis and inflammation by controlling lipid metabolism and modulating oxidative stress and the hepatic TLR4–MyD88 pathway. The peel of the red pitaya contains more bioactive compounds than the meat, making it a good source of dietary betacyanins **(Yeh *et al.*, 2020)**.

The effect of red dragon fruit as powder on kidney functions (uric acid, urea and creatinine) of hypercholesterolemic rats are shown in Table (6). It is clear to notice that the uric acid of the positive control rats group recorded a higher value when compared with the negative control group with a significant difference (P≤0.05). The mean values were 6.90 and 2.50 mg/dl; respectively. While the highest uric acid level of all treated groups was recorded for the group fed on 2 % red dragon fruit powder but, the lowest value was recorded for the group fed on 6% red dragon fruit powder with a significant difference (P≤0.05). The mean values were 4.60 and 2.70 mg/dl; respectively.

As for urea level, data indicated that the positive control rats group recorded a higher value when compared with the negative control group with significant difference (P≤0.05). The mean values were 60 and 24 mg/dl; respectively. The highest mean value of serum urea of all treated groups was recorded for the group fed on 2 % red dragon fruit powder but, the lowest value was recorded for the group fed on 6% red dragon fruit powder with a significant difference (P≤0.05). The mean values were 41 and 30 mg/dl; respectively.

On the other hand, the creatinine level of the positive control rats group recorded a higher value when compared with the negative control group with a significant difference (P≤0.05). The mean values were 1.73 and 0.71 mg/dl; respectively. While the highest creatinine level of the treated group was recorded for the group fed on 2 % red dragon fruit powder but, the lowest value was recorded for the group fed on 6% red dragon fruit powder with a significant difference (P≤0.05). The mean values were 1.58 and 0.82 mg/dl; respectively.

Red dragon fruit juice administration can prevent or reduce the effects of doxorubicin (DOX)-induced nephrotoxicity in rats,

as evidenced by a decrease in the number of glomerular with protein sediment and necrotic tubular epithelium cells, as well as improved kidney functions (**Prasetyo *et al.*, 2018)**.

Data tabulated in Table (7) show the effect of red dragon fruit as powder on blood parameters (red blood cell, white blood cell, hemoglobin and platelet) of hypercholesterolemic rats. It is obvious that the red blood cell (RBC) level of the negative control rats group recorded a higher value when compared with the positive control group with a significant difference (P≤0.05). The mean values were (6.50 and 3.60) 10^6/mm^3; respectively. While the highest RBC level of the treated group was recorded for the group fed on 6 % red dragon fruit powder but, the lowest value was recorded for the group fed on 2% red dragon fruit powder with a significant difference (P≤0.05). The mean values were (5.60 and 4.70) 10^6/mm^3; respectively.

In the case of white blood cell (WBC), data showed that the level of the negative control rats group recorded a higher value when compared with the positive control group with a significant difference (P≤0.05). The mean values were (10.50 and 7) 10^3/mm^3; respectively. While the highest WBC level of the treated group was recorded for the group fed on 2 % red dragon fruit powder but, the lowest value was recorded for the group fed on 6% red dragon fruit powder with a significant difference (P≤0.05). The mean values were (11.10 and 8.80) 10^3/mm^3; respectively.

As for hemoglobin level, the negative control rats group recorded a higher value when compared with the positive control group with a significant difference (P≤0.05). The mean values were 16.80 and 11.40 g/dl; respectively. While the highest hemoglobin level of treated group was recorded for the group fed on 6 % red dragon fruit powder but, the lowest value was recorded for the group fed on 2% red dragon fruit powder with a significant difference (P≤0.05). The mean values were 16.60 and 14.90 g/dl; respectively.

On the other hand, the platelet level of the positive control rats group recorded a higher value when compared with the negative control group with a significant difference (P≤0.05). The mean values were (760 and 215) 10^6/mm^3; respectively. While the highest platelet level of the treated group was recorded for the

group fed on 6 % red dragon fruit powder but, the lowest value was recorded for the group fed on 2% red dragon fruit powder with a significant difference ($P \leq 0.05$). The mean values were (485.00 and 265.00) $10^6/mm^3$; respectively. These findings are consistent with those of **Arifin *et al.*, (2012)**, who found that giving dragon fruit juice (*H. polyrhizus*) to female white mice increased hemoglobin levels, erythrocyte amount, and hematocrit percentage.

Flavonoids and other phytochemicals found in red dragon fruit are beneficial to one's wellbeing **(Rebecca *et al.*, 2010)**. The flavonoids in red dragon fruit have the ability to capture superoxide and peroxynitrite directly. Flavonoids increase the bioavailability of nitric oxide and prevent the formation of peroxynitrite by trapping superoxides. Local vasodilation is caused by nitric oxide, a major molecule. Flavonoids may also bind to peroxynitrite, which damages the endothelium, improving blood flow in the coronary arteries **(Akhlaghi and Brian, 2009)**.

In rats with HIT, red dragon fruit peel yogurt improves hemoglobin, hematocrit, and platelet levels. In thrombocytopenic rats, yogurt containing 25% red dragon fruit peel offers an important dose for improving hemoglobin, hematocrit, and platelet levels **(Cahyati *et al.*, 2021).**

Table (1): Identified phenolic compounds in the red dragon fruit by HPLC

Phenolic compounds	Concentrations (mg/g)
Gallic acid	1.75
Caffeic acid	ND
Chlorogenic acid	4.15
Ferulic acid	2.60
Sinapic acid	ND
***p*-Coumaric acid**	2.78
***p*-hydroxybenzoic acid**	1.25
Protocatechuic acid	ND
Synergic acid	3.00
Catechin	7.42

ND= Not detectable

Table (2): Effect of the red dragon fruit as powder on the serum total cholesterol and triglycerides of hypercholesterolemic rats

Parameters / Treatments	Total cholesterol (mg/dl)	Triglycerides (mg/dl)
	Mean ±SD	Mean ±SD
Control group (-)	95.0±0.11^{e}	105±0.10^{e}
Control group (+)	240±0.10^{a}	190±0.16^{a}
Hypercholesterolemic rats + 2% red dragon fruit	167±0.12^{b}	152±0.11^{b}
Hypercholesterolemic rats + 4% red dragon fruit	125±0.14^{c}	131±0.15^{c}
Hypercholesterolemic rats + 6% red dragon fruit	112±0.12^{d}	115±0.14^{d}
LSD	**5.420**	**3.461**

Means in the same column with different superscript letters is significantly different at ($P \leq 0.05$)

Table (3): Effect of the red dragon fruit as powder on the serum lipoproteins and atherogenic index of hypercholesterolemic rats

Parameters / Treatments	(HDL_{-C}) (g/dl)	(LDL_{-C}) (g/dl)	($VLDL_{-C}$) (g/dl)	Atherogenic index (%)
	Mean ±SD	Mean ±SD	Mean ±SD	Mean ±SD
Control group (-)	60.0 ± 0.13^{a}	14.0 ± 0.12^{e}	21.0 ± 0.12^{e}	0.58 ± 0.10^{d}
Control group (+)	32.0 ± 0.11^{e}	170.0 ± 0.14^{a}	38.0 ± 0.14^{a}	6.50 ± 0.13^{a}
Hypercholesterolemic rats + 2% red dragon fruit	38.0 ± 0.15^{d}	98.6 ± 0.10^{b}	30.4 ± 0.10^{b}	$3.39\pm.016^{b}$
Hypercholesterolemic rats + 4% red dragon fruit	50.0 ± 0.15^{c}	48.8 ± 0.13^{c}	26.2 ± 0.13^{c}	$1.50\pm.011^{c}$
Hypercholesterolemic rats + 6% red dragon fruit	54.0 ± 0.16^{b}	35.0 ± 0.16^{d}	23.0 ± 0.16^{d}	1.07 ± 0.13^{c}
LSD	**2.303**	**4.101**	**1.280**	**0.810**

Means in the same column with different superscript letters is significantly different at ($P\leq0.05$)

Table (4): Effect of the red dragon fruit as powder on the serum glucose level of hypercholesterolemic rats

Parameters / Treatments	Glucose level (mg/dl)
	Mean ±SD
Control group (-)	96.0 ± 0.12^{e}
Control group (+)	180.0 ± 0.15^{a}
Hypercholesterolemic rats + 2% red dragon fruit	140.0 ± 0.12^{b}
Hypercholesterolemic rats + 4% red dragon fruit	125.0 ± 0.10^{c}
Hypercholesterolemic rats + 6% red dragon fruit	119.0 ± 0.13^{d}
LSD	**3.850**

Means in the same column with different superscript letters is significantly different at ($P\leq0.05$)

Table (5): Effect of the red dragon fruit as powder on the liver enzymes of hypercholesterolemic rats

Parameters / Treatments	ALT (U/L)	AST (U/L)	ALP (U/L)
	Mean ±SD	Mean ±SD	Mean ±SD
Control group (-)	12.0±0.10^{e}	42.0±0.15^{d}	90±0.16^{e}
Control group (+)	23.0±0.13^{a}	95.0±0.11^{a}	135±0.10^{a}
Hypercholesterolemic rats + 2% red dragon fruit	19.0±0.11^{b}	61.0±0.16^{b}	119±0.12^{b}
Hypercholesterolemic rats + 4% red dragon fruit	17.0±0.12^{c}	49.0±0.14^{c}	110±0.15^{c}
Hypercholesterolemic rats + 6% red dragon fruit	15.0±0.11^{d}	41.0±0.10^{d}	104±0.12^{d}
LSD	**0.750**	**1.852**	**3.503**

Means in the same column with different superscript letters is significantly different at ($P \leq 0.05$)

Table (6): Effect of the red dragon fruit as powder on the kidney functions of hypercholesterolemic rats

Parameters / Treatments	Uric acid (mg/dl)	Urea (mg/dl)	Creatinine (mg/dl)
	Mean ±SD	Mean ±SD	Mean ±SD
Control group (-)	2.50±0.10^{c}	24.0±0.10^{e}	0.71±0.10^{b}
Control group (+)	6.90±0.13^{a}	60.0±0.13^{a}	1.73±0.13^{a}
Hypercholesterolemic rats + 2% red dragon fruit	4.60±0.13^{b}	41.0±0.13^{b}	1.58±0.13^{a}
Hypercholesterolemic rats + 4% red dragon fruit	4.00±.016^{b}	36.0±.016^{c}	0.90±.016^{a}
Hypercholesterolemic rats + 6% red dragon fruit	2.70±.011^{c}	30.0±.011^{d}	0.82±.011^{b}
LSD	**1.151**	**3.402**	**0.852**

Means in the same column with different superscript letters is significantly different at ($P \leq 0.05$)

Table (7): Effect of the red dragon fruit as powder on *hematological* parameters of hypercholesterolemic rats

Parameters / Treatments	RBC 10^6/mm^3	WBC 10^3/mm^3	Hemoglobin g/dl	Platelet 10^6/mm^3
	Mean ±SD	Mean ±SD	Mean ±SD	Mean ±SD
Control group (-)	6.5±0.12^a	10.5±0.10^a	16.8±0.11^a	215±0.14^d
Control group (+)	3.6±0.13^c	7.0±0.14^c	11.4±0.12^c	760±0.16^a
Hypercholesterolemic rats + 2% red dragon fruit	4.7±0.12^b	11.1±.010^a	14.9±0.10^b	265±0.60^c
Hypercholesterolemic rats + 4% red dragon fruit	5.2±.010^b	10.7±.012^a	16.5±.015^a	360±0.13^c
Hypercholesterolemic rats + 6% red dragon fruit	5.6±.015^a	8.8±0.14^b	16.6±.013^a	485±0.15^b
LSD	**1.151**	**1.062**	**1.220**	**5.120**

Means in the same column with different superscript letters is significantly different at ($P \leq 0.05$)

Conclusion

The present study demonstrates that red dragon fruits is rich in phenolic content and antioxidant properties that has a significant influence on altering rats lipid metabolism. The red dragon powder fortified diet has potential in reducing TC, TG, LDL-c, VLDL-c, liver enzymes, kidney functions and increasing HDL-c levels. The diet fortified with red dragon powder especially 6% contribute the prevention of dyslipidemia.

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التأثير المحتمل لثمار فاكهة التنين الأحمر علي الفئران المصابة بارتفاع الكوليسترول

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الملخص العربي

أجريت هذه الدراسة لمعرفة تأثير المستويات المختلفة (2% ، 4%، 6%) من مسحوق فاكهة التنين الأحمر على الفئران المصابة بارتفاع الكوليسترول. واشتملت الدراسة على ثلاثون من ذكور الفئران البيضاء وزنها حوالي (140 ± 10 جم) وتم تقسيمها إلى (٥) مجموعات، كل مجموعة تحتوي على (٦) فئران. تم الاحتفاظ بمجموعة كمجموعة ضابطة سالبة، بينما تم اصابة المجموعات الأربع الأخرى بارتفاع مستوى الكوليسترول في الدم بواسطة حقنهم مادة تريتون إكس ١٠٠ بجرعة (١٠٠ مجم/ كجم من وزن الجسم). كذلك تم تقدير بعض التحاليل الكيميائية الحيوية والتي أشتملت على دهون الدم، معامل التصلب(AI) ، مستوى الجلوكوز ، وإنزيمات الكبد ووظائف الكلى. كما تم التعرف على المركبات الفينولية لثمار فاكهة التنين الأحمر باستخدام جهاز الكروماتوجرافى الغازي عالي الأداء. وأظهرت النتائج أن مسحوق فاكهة التنين الأحمر تحتوي على العديد من المركبات الفينولية، حيث سجلت المستويات الأعلى لحمض الكاتشين وحمض الكلوروجينيك وحمض السيرينجك وحمض بى كيوماريك وحمض الفيرلك. أظهرت النتائج أيضًا أن المجاميع المعاملة بمسحوق فاكهة التنين الأحمر حدث لها انخفاضًا كبيرًا في إنزيمات الكبد ووظائف الكلى ومستوى الجلوكوز ومؤشرات الدم وقيم دهون الدم (TC، TG، LDL-c، VLDL-c) مقارنة بالمجموعة الضابطة الموجبة وزيادة في مستوى HDL-c وبناءً على ذلك فإن مسحوق فاكهة التنين الأحمر خاصة مستوى ٦٪ قد حسن جميع القياسات البيوكيميائية المختبرة ويمكن اعتباره وسيلة علاجية لعلاج ارتفاع مستوى كوليسترول الدم .

الكلمات المفتاحية: الفاكهة الغير شائعة، ارتفاع مستوى الكوليسترول في الدم، المركبات الفينولية، التحاليل الكيميائية الحيوية.