

## Effects of Bioactive Component of Kiwi Fruit and Avocado (Fruit and Seed) on Hypercholesterolemic Rats

<sup>1</sup>Manal M.S.M. Shehata and <sup>2</sup>Sahar S.A. Soltan

<sup>1</sup>Department of Food Science, Faculty of Agriculture, Zagazig University, Zagazig, Egypt

<sup>2</sup>Department of Home Economics (Nutrition and Food Science),  
Faculty of Specific Education, Fayoum University, Egypt

**Abstract:** This study evaluated the effect of kiwi fruit and avocado (fruit and seed) on diet-induced hypercholesterolemia in rats. The results showed that avocado seed content was the highest in phenolic compounds (285.43 mg/100g GAE); flavonoids (3.21mg/100g CE) and soluble dietary fiber (38%) followed by avocado and kiwi fruits. Kiwi fruit is rich in ascorbic acid (15.52 mg/100g). Hypercholesterolemia was induced using a diet containing 1% cholesterol and 16%fat. Sixty six male of Albino rats weighing 115-120g were randomly distributed into eleven groups of six rats each-Group 1: control rats fed on the standard diet, group 2 rats fed on hypercholesterolemic diet, while the other groups of rats fed hypercholesterolemic diet supplemented with kiwi fruit and avocado (fruit and seed) (10%, 20% and 30%).The present study showed that 1% cholesterol and 16% fat administration for 6 weeks caused a significant increase  $P<0.05$  in total cholesterol and triglyceride in both serum and liver. In serum, the levels of total cholesterol, triglyceride and LDL-C significantly decreased for the groups fed kiwi fruit and avocado (fruit and seed) in comparison with the hypercholesterolemic group (HC group). The activities of AST and ALT enzymes decreased significantly for the groups fed the kiwi fruit and avocado (fruit, seed) in comparison with the HC group. Atherogenic index (AI) increased significantly compared to control group. Regarding liver tissue, the levels of total cholesterol and triglyceride decreased significantly for the kiwi fruit and avocado (fruit, seed) fed rats compared to the HC group. The liver content of reduced glutathione increased significantly in comparison with the hypercholesterolemic group. In this study, the lowering effect of avocado seed on lipid profiles in serum and liver was more observable than that of avocado fruit or kiwi fruit. The results suggest that consumption of kiwi fruit and avocado (fruit and seed) might have some cardiovascular protective properties and beneficial effects on atherosclerosis, CVD risks in hypercholesterolemic rats. So, we recommended consuming avocado seed because the avocado seed have strong antioxidant activity and lower effect of lipid profile.

**Key words:** Kiwifruit • Avocado fruit • Avocado seed • Lipid profiles • Diet-hypercholesterolemia • Risk factors • Cardiovascular disease

### INTRODUCTION

Hypercholesterolemia is a lipoprotein metabolic disorder characterized by high serum low density lipoprotein and blood cholesterol. It is major risk factors in the development and progression of atherosclerosis that leads to cardiovascular disease [1]. Hypercholesterolemia is a major problem to many societies especially the health professionals because of

the close correlation between cardiovascular diseases and lipid abnormalities [2, 3]. Dietary factors such as continuous ingestion of high amounts of saturated fats and cholesterol are believed to be directly related to hypercholesterolemia and susceptibility to atherosclerosis [4]. Clinical trials have demonstrated that intensive reduction of plasma low density lipoprotein (LDL-C) levels could reverse atherosclerosis and decrease the incidence of cardiovascular diseases [5].

Plants in general and fruits particularly have several compounds with antioxidant properties, which include ascorbic acid, carotenoids and polyphenols. Increased consumption of fruits protects cardiovascular diseases [6, 7]. Kiwi fruit (*Actinidia deliciosa planch*) is one of the most popular fruits worldwide and is cultivated in many countries, such as New Zealand, Italy, Japan, Greece and France [8]. There are many Kiwi fruit cultivars and the most known is 'Hayward'. 'Biden' is less spread than "Hayward" and both cultivars belong to the *Actinidia deliciosa* species and are known for good taste [9-11]. Kiwi fruit is a highly nutritional fruit due to its high level of vitamin C and its strong antioxidant including carotenoids, lutein, phenolics, flavonoids and chlorophyll [12]. Kiwi fruit is a rich source of vitamins E, fructose, galactose and minerals, its contains isoflavones and flavonoids which are important phytochemical in kiwi extract and represent the major class of phytosterogen, which has an important function as anti-carcinogenic, neuroprotective and cardio protective activity [13, 14]. Recent studies have shown that kiwi fruit have antioxidant [9], cardiovascular protective [15]. Extracts of kiwi fruit inhibit cancer cell growth and exhibit cell protection against oxidative DNA damage *in vitro* [16, 17].

Avocado (*Persea americana* Mill) is an important commercial tropical fruit. Avocado fruit content high levels of bioactive compounds and including vitamin E, Ascorbic acid, carotenoids and soluble phenolics [18]. It contains one to two times more protein than any other fruit, is high in manganese, phosphorous, iron and potassium, but low in sodium and also contains vitamin C,  $\beta$ -carotene, thiamin, riboflavin, nicotinic acid and folate [19]. Avocado is a good source of the essential linoleic acid. The amount of simple sugars in the avocado fruit is low, but in contrast, it contains appreciable levels of dietary fiber and is the highest in fiber among fruits [20]. In the folk medicine of Latin America and Africa, it has been used as a remedy for hypertension [21], renal diseases [22] and diabetes [23, 24] and for antipyretic and analgesic purposes [25]. Some studies in rats have demonstrated the hypotensive [26, 27], antioxidant [28] and hypocholesterolemic properties [29] of the extract of *Persea americana* leaves, partially confirming the popular belief. The present study was designed to determination bioactive component and hypocholesterolemic effect of kiwifruit and avocado (fruit and seed) in rats.

## MATERIALS AND METHODS

**Materials:** The mature fresh avocado (*Persea Americana* Mill.) and kiwi fruit (*Actinidia deliciosa*) Hayward strain were purchased from local markets in Egypt.

**Chemicals:** Folin-Ciocalteu reagent, methanol, 2, 2 diphenyl-1-picrylhydrazyl (DPPH), Gallic acid, NaCO<sub>3</sub>, AlCl<sub>3</sub>, NaOK tartarate and purified cholesterol were purchased from sigma- Chemical Company, USA. Kits for blood analysis were purchased from Biodiagnostic Co. Dokki, Giza, Egypt. All other reagents used were analytical grade. All solvent used (isopropanol and methanol) were obtained from El-Goumhouria Co. Cairo, Egypt.

### Methods:

**Preparation of Fruit and Seed Extract:** The kiwi fruit, avocado (fruit and seed) were extracted according the method of Biglari *et al.* [30], this method modified to obtained the maximum yield as follows: 500 gram fruits and seed was pitted, cut to small pieces and dry-blended for 3min. The blended fruits were extracted with 1 liter methanol: water (50:50 v/v), at room temperature 25°C for 10 hours using an orbital shaker. The extracts were then filtered and centrifuged and the supernatant was concentrated under reduced pressure at 40°C using rotary evaporator to obtain the methanol crude extracts of the fruit and seed. The extracts were kept in dark glass bottles and stored at -18°C until used.

**Determination of Crude Fibers, Dietary Fiber (Insoluble and Soluble) and Ascorbic Acid:** Crude fibers, dietary fiber (insoluble and soluble) and Ascorbic acid in kiwi fruit, avocado fruit and seed were analyzed by the method of AOAC [31].

**Determination of Total Phenolic Compounds:** Total phenolics compounds of kiwi fruit and avocado (fruit and seed) were determined spectrophotometer using Folin-Ciocalteu colorimetric method [32]. Briefly 5ml of distilled water, 0.5-1.0 ml of each sample of extract, 1.0 ml of Folin-Ciocalteu reagent was added to a 25ml volumetric flask. The contents were mixed and allowed to stand at room temperature for 5-8 min. Then 10 ml of 7% NaCO<sub>3</sub> solution was added, followed by the addition of distilled water filled to volume. Solutions were mixed and allowed to stand at room temperature for 2hours. Sample aliquots were filtered through a Whatman 0.54 in polytetrafluoro ethylene filter prior to the determination of total phenols concentration using a spectronic 2000 spectrophotometer monitoring 750nm. Total phenolic compounds was standardized against Gallic acid and expressed as milligram per liter of Gallic acid equivalents (GAE).

**Determination of Total Flavonoids:** Total flavonoids content of kiwi fruit and avocado (fruit and seed) extracts were determined by using spectrophotometer according to the method described by Khatiwara *et al.* [33]. Aliquots

extract solution 1 ml, were taken and made up the volume 3ml with methanol, then 0.01 ml AlCl<sub>3</sub> (10%), 0.1 ml Na k tartarate and 2.8 ml distilled water were added sequentially. The test solution was vigorously shaken. Absorbance at 415 nm was recorded after 30 min of incubation. A standard calibration was generated at 415 nm using the concentration of flavonoid in the test sample were calculated from calibration plot and expressed as mg quercetin equivalent/g sample.

#### **Identification of Individual Phenolic Compounds by High Performance Liquid Chromatography (HPLC):**

Phenolic compounds in kiwi fruit and avocado (fruit and seed) were determined by HPLC according to the method by Goupy *et al.* [34] as follows: 5g of samples were mixed with methanol and centrifugated at 1000 rpm for 10 min and the supernatant was filtered through 0.2 Mm Millipore membrane filter than 1-3 ml was collected in avial for injection into HPLC Hewllet Puckerred (Series 1050) equipped with auto sampling injector, solvent degasser, ultraviolet UV detector set at 280 nm and quaternary HP pump (series 1100) packed column Hypesil BDs-C18 4.0 X 250 nm was used to separation phenolic compound. The column temperature was maintained at 35°C. Gradient separation was carried out with methanol and acetonitrile as a mobile phase at flow rate of 1 ml/min. Phenolic acid standard from Sigma Company were dissolved in mobile phase and injected into HPLC. Retention time and peal area were used to calculation of phenolic compounds concentration by data analysis of Hewllet software, Germany.

#### **Identification of Individual Flavonoids by HPLC:**

Flavonoids compounds were determined by HPLC according to the method of Mattila *et al.* [35] as follow: 5g of sample were mixed with methanol and centrifuged at 1000 rpm for 10 min and the supernatant was filtered through a 0.2 µm Millipore membrane filter than 1-3 ml was collected in a vial for injected into Hewllet Puckerred (Series 1050) equipped with auto sampling injector, solvent degasser, ultraviolet UV detector set at 330 nm and quarter HP pump (series 1050).The column was used to separate flavonoid was Zorba X ODS 5 µm (4.6 x 250 nm). The column temperature was maintained at 35°C. Gradient separation was carried out with methanol and acetonitrile as a mobile phase at flow rate of 1 ml/min. Flavonoid standard from Sigma Company were dissolved in mobile phase and injected into HPLC. Retention time and peal area were used to calculation of flavonoid compounds concentration by data of Hewllet Packared software. Germany.

#### **Determiration of Antioxidant Activity Using 2,2-diphenyle-1-picrylhydrazyl (DPPH) Radical Scavenging Method:**

Antioxidant activity of fruits and seed was determined using the stable radical (DPPH) according to Brand-Williams *et al.* [36]. Aliquot of 0.1 ml methanol solution containing different concentration from kiwi fruit and avocado (fruit and seed) was added to 3.9 ml of 6 x 10<sup>-5</sup> methanolic solution of freshly prepared DPPH. After 30 min incubation at room temperature, the absorbance was read at 515 nm by Perkin Elr spectrophotometer for all tested samples. DPPH solution without fruit and seed extracts were used as control. Percentage inhibition % of DPPH free radical was calculated according to the following equation:

$$\% \text{ Initiation} = \frac{\text{Absorbance control} - \text{Absorbance sample}}{\text{Absorbance control}} \times 100$$

$$\text{Antiradical efficiencies (AE)} = \frac{1}{\text{EC}_{50}}$$

EC<sub>50</sub>=extraction concentration providing 50% inhibition of the DPPH.

**Animals:** Sixty-six male Albino rats weighing 115-120g were purchased from animal house of El-Salam Farm, Giza, Egypt. The animals were placed in individual cages at 25°C under a 12 h light/dark cycle. Water was provided *ad libitum*. After one week of acclimatization, the rats were divided randomly into eleven groups of six rats in each (n = 6) and treated as follows:

**Group 1(G1):** Rats fed basal diet assigned as normal control group. Basal diet was preparing according to Reeves *et al.* [37].

**Group 2 (G2):** Rats fed basal diet contained 1% cholesterol + 16% fat and 0.2% cholic acid and assigned as a hypercholesterolemic (HC) rats according to Harnafi *et al.* [38].

**Groups 3, 4 and 5:** Rats fed HC diet supplemented with 10%, 20% and 30% kiwi fruit.

**Groups 6, 7 and 8:** Rats fed HC diet supplemented with 10%, 20% and 30% avocado fruit.

**Groups 9, 10 and 11:** Rats fed HC diet supplemented with 10%, 20% and 30% avocado seed. Feed intake was measured daily for each rat. Body weight was measured at baseline and at 6 weeks and feed efficiency ratio

(FER) was calculated. At the end of experimental period (6 weeks) animals were sacrificed after overnight fasting. The serum was separated by allowing blood samples left for 15 minutes at temperature of 25°C then centrifuged at 300 rpm for 20 minutes, then kept in plastic vials at -20°C until analysis. Main of organs was carefully separated and washing with cold saline and dry on filter paper and the weight was recorded.

**Biochemical Analysis:** Serum was analyzed for the following biochemical parameter: total cholesterol by the method of Richmond [39], HDL-cholesterol by Burstein *et al.* [40], triglyceride by Jacobs and Vander mark [41] and AST and ALT by Reitman and Frankel [42]. Calculation of LDL-Cholesterol fraction and atherogenic index (AI) and HTR ratio involves equations developed by Friedewald *et al.* [43]:

$$\begin{aligned} \text{Atherogenic index (AI)} &= \\ (\text{Serum total cholesterol} - \text{HDL-c}) / \text{HDL-c} \\ \text{HTR ratio} &= \text{HDL-c} / \text{TC} \times 100 \end{aligned}$$

**Extraction and Determination of TG and TC and GSH in Liver Tissue:** At the end of the experimental, Liver was removed, rinsed in ice chilled normal saline and blotted on filter paper and then tissues were cut into small portion and stored at -20°C before use. Extraction of liver analysis of total cholesterol and triglycerides was carried out according to the method by Hostmark [44]: 1g of liver portion from each rat was homogenized in 10 ml isopropanol. The liver homogenate was allowed to stand for 48 h at 4°C. The mixture was centrifugated 15 min at 2500 rpm and the supernatant was used for lipid analysis. Total cholesterol and triglyceride were quantified using enzymatic as described above. Glutathione reduced activity (GSH) of liver was determined according to the method by Beulter *et al.* [45].

**Statistical Analysis:** The results obtained were analyzed using SPSS program (version 17.0) and expressed as mean and standard deviations (SD). Statistical significance ( $p < 0.05$ ) among the groups were determined by one-way ANOVA followed by Duncan's multiple range test according to the method by Bailey [46].

## RESULTS AND DISCUSSION

Bioactive component in kiwi fruit and avocado (fruit and seed) was illustrated in Table 1. The data in Table 1 revealed that avocado fruit was the highest in crude fiber (12.84%) followed by kiwi fruit (11.22%) and

avocado seed (9.42%). Also avocado fruit and seed have higher dietary fiber (6.56 and 7.61 mg/100g) and insoluble dietary fiber (64 and 62%) and soluble dietary fiber (36 and 38%) compared to kiwi fruit (3.7 mg/100g, 56.2% and 24.1%) respectively. Also data in Table 1 showed the avocado fruit and seed have higher content flavonoids and lower level of ascorbic acid compared to kiwi fruit. These results agreement with Reyes-Caudello [47] reported avocado seed have higher total dietary fiber (39.9 and 36.9%). Park *et al.* [48] reported the amount ascorbic acid in kiwi fruit ranged from 6.56 to 152 mg/100g.

The yield, total phenolic compounds, efficient concentration and antiradical efficiencies of kiwi fruit and avocado (fruit and seed) are shown in Table 2. The maximum yield percentage was obtained for kiwi fruit (18.1%) followed by avocado fruit (14.6%) and avocado seed (12.5%). The results showed the highest concentration of phenolic compound was obtained of avocado seed extract (285.43 mg/100g GAE) followed by avocado fruit (259.15 mg/100g GAE) and kiwi fruit (258.55 mg/100g GAE). These results indicated that the kiwi fruit and avocado (fruit and seed) has higher levels of phenolic compounds. These results are accordance with those obtained by Gorinstein *et al.* [49], who reported that phenolic compound of kiwi fruit extract was higher. Wang *et al.* [50] indicated that avocado seed content the highest total phenolic content and antioxidant capacities whereas the pulp had the lowest. Phenolic content of avocado seed was 88.2 mg/g compared to flesh 1.3 GAE [51]. Among the sample investigated, avocado seed extract showed the highest antiradical efficiency (0.122) and the lowest amount of extract required to scavenge 50% DPPH radical (8.21  $\mu\text{g sample}/\mu\text{g DPPH}$ ) (Table 2). The amount of kiwi fruit, avocado fruit and avocado seed extracts required to scavenging 50% of DPPH radical was 30.12, 24.73 and 8.21  $\mu\text{g sample}/\mu\text{g DPPH}$  respectively. Soong and Barlow [51] reported antioxidant activity of avocado seeds was much higher than edible portion.

Data in Table 3 showed that remaining DPPH % of extracts of kiwi fruit and avocado (fruit and seed). The results showed concentration 5  $\mu\text{g sample}/\mu\text{g DPPH}$ , the remaining DPPH% was 91.6%, 89.9% and 69.5 of kiwi fruit, avocado fruit and avocado seed extracts. When the concentration increased to 15  $\mu\text{g sample}/\mu\text{g DPPH}$  the remaining percent of DPPH decreased to 75.5%, 70.2% and 10%, respectively for kiwi fruit, avocado fruit and avocado seed. Antioxidant activity of avocado seeds was much higher than fruit and kiwi fruit. The effects of antioxidant of avocado seed on DPPH radical scavenging was thought to be due to their hydrogen donating ability or radical scavenging activity [52]. Strong activity of

Table 1: Crude fiber, flavonoids and ascorbic acid in kiwi fruit and avocado (fruit and seed).

Bioactive component	Kiwifruit	Avocado fruit	Avocado seed
Crude fiber %	11.22	12.84	9.42
Dietary fiber mg/100g	3.7	6.56	7.6
Insoluble dietary fiber %	56.2	64	62
Soluble dietary fiber %	24.1	36	38
Total flavonoid mg/100g	1.68	2.96	3.21
Ascorbic acid mg/100g	15.52	9.37	5.24

Table 2: Yield, total phenolic compounds, efficient concentration and antiradical efficiencies of kiwi fruit and avocado (fruit and seed).

Extracts	Yield g/100g	Total phenolic mg/100g	Efficient concentration EC <sub>50</sub>	Antiradical efficient AE
Kiwi fruit	18.1	258.55	30.12	0.033
Avocado fruit	14.6	259.15	24.73	0.040
Avocado seed	12.5	285.43	8.21	0.122

Table 3: Scavenging activity of methanol extract of Kiwi fruit and avocado (fruit and seed) determined by the scavenging of DPPH radial.

Concentration of methanol extract µg sample/µg DPPH	% Scavenging activity of kiwi fruit	% Scavenging activity of avocado fruit	% Scavenging activity of avocado seed
5	91.6	89.9	69.5
10	80.1	83.6	40
12	79.6	75.2	25
15	75.5	70.2	10

Table 4: Phenolic compounds of kiwi fruit and avocado (fruit and seed) (ppm).

Phenolic compounds	Kiwi fruit	Avocado fruit	Avocado seed
Pyrogallol	224.51	217.57	226.93
Syringic	--	--	768.59
Protocatechuic	13.85	23.34	54.32
Vanillic	4.32	3.88	50.89
Chlorogenic	57	--	154.71
Catechol	5.33	21.36	28.94
Caffeine	2.28	--	5.58
Catechin	48.62	47.78	196.20
Ferulic	--	2.06	14.60
Cinnamic	3.41	--	0.35
Coumarin	3.86	0.99	--
Ellagic	--	73.41	--

Table 5: Flavonoids compounds of kiwi fruit and avocado (fruit and seed) (µg/100g).

Flavonoid compounds	Kiwi fruit	Avocado fruit	Avocado seed
Rutin	--	82.03	53.21
Rosmarinic	901.80	60.71	114.19
Quercitrinic	--	87.40	112.79
Quercetin	--	222.24	61.98
Narenginin	66.63	-	--
Hesperetin	--	176.52	54.62
Apigenin	--	--	84.36
Kampferol	--	60.77	--
Hesperetin	826.20	--	387.52

avocado seed may be due to higher content phenolic compounds and flavonoids compounds. Other researchers have found that strong correlation between antioxidant activity as assessed by DPPH and total phenolics [53].

Data in Table 4 showed identification of phenolic compound of kiwi fruit and avocado (fruit and seed) by HPLC. Phenolic acid (ppm) was the following: pyrogallol (224.51, 217.57 and 226.93ppm); protocatechuic (13.85, 23.34 and 54.32 ppm); vanillic acid (4.32, 3.88 and 50.89 ppm); catechol (5.33, 21.36 and 28.94 ppm); catechin (48.62, 47.78 and 196.20 ppm). Syringic was only detected in avocado seed. Ellagic acid only detected in avocado fruit. The major concentration of phenolic acid was present in avocado seed. These results are in line with those obtained by Park *et al.* [48], who reported that the protocatechuic of two type kiwi was (23.4 and 25.7 mg), vanillic (6.18 and 4.56) and caffeic (45.3 and 17.1 ppm). Rodriguez-Carpena *et al.* [54] reported that phenolic substances are widely distributed in flesh and seed avocado (caffeic acid, P-Hydroxy benzoic, chroogenic, ferulic and epicatechin).

Identification of flavonoid compounds of kiwi fruit and avocado (fruit and seed) are summarized in Table 5. Rosmarinic and hesperidin was the major flavonoid in kiwi fruit (901.80 and 826.20 µg/100g) and avocado seed (114.19 and 387.52 µg/100g). Rutin, quercitrinic and quercetin was detectable in avocado fruit and seed and not detectable in kiwi fruit. Apigenin was only detectable in avocado seed. Meanwhile, narenginin was only detectable in kiwi fruit. From these results it could be noticed that the flavonoid compounds in avocado fruit and seed were more than kiwi fruit. Kosinska *et al.* [55] indicated that the avocado seed and peel are rich in flavonoid (quercetin). Pahlavani *et al.* [56] reported protocatechuic acid was the main phenolic compound in avocado seed powder followed by kaempferol and vanillic acid.

Data in Table 6 showed that induced hypercholesterolemia caused a significant increase P<0.05 in body weight gain, feed intake and feed efficiency ratio (FER) in HC group G2 as compared with healthy control group G1. These results are in agreement with the findings of Lecumberri *et al.* [57] and Barakat and Lamiaa [58], they reported that rats fed high cholesterol diet showed significant increase in body weight gain. Administration of kiwi fruit and avocado (fruit and seed) to hypercholesterolemic rats caused a significant decrease in body weight gain and feed efficiency ratio at all levels except for diet supplemented 30% avocado fruit.

Table 6: Changes of body weight, feed intake and FER in the hypercholesterolemic rats fed diet supplemented with kiwi fruit and avocado (fruit and seed).

Groups	°IBW (g)	§FBW (g)	*BWG (g)	†FI (g/day)	‡FER (g)
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
G1 control (-)	115.5±2.83 <sup>b</sup>	166.3±2.28 <sup>c</sup>	50.8±2.00 <sup>c</sup>	13.00±0.84 <sup>d</sup>	0.093±0.25 <sup>bcd</sup>
G2 (HC) (+)	116.2±3.21 <sup>b</sup>	185.9±3.69 <sup>a</sup>	69.7±2.61 <sup>a</sup>	14.48±0.24 <sup>a</sup>	0.115±0.04 <sup>a</sup>
G3(HC)+10% Kiwi fruit	115.9±2.82 <sup>b</sup>	169±2.61 <sup>de</sup>	53.1±2.28 <sup>bc</sup>	13.98±0.53 <sup>ab</sup>	0.090±0.13 <sup>c</sup>
G4 (HC) +20% Kiwi fruit	117.1±1.41 <sup>ab</sup>	171.7±3.85 <sup>cd</sup>	54.6±2.28 <sup>b</sup>	14.32±0.26 <sup>a</sup>	0.091±0.08 <sup>cd</sup>
G5 (HC) +30% Kiwi fruit	117.4±1.41 <sup>ab</sup>	172.7±2.83 <sup>bc</sup>	55.3±3.69 <sup>b</sup>	14.4±0.22 <sup>a</sup>	0.091±0.05 <sup>cd</sup>
G6 (HC) +10% avocado fruit	119±1.26 <sup>a</sup>	175.6±3.16 <sup>b</sup>	54.6±2.61 <sup>b</sup>	14.17±0.42 <sup>ab</sup>	0.092±0.1 <sup>bcd</sup>
G7 (HC) +20% avocado fruit	116.8±2.28 <sup>ab</sup>	172.3±1.41 <sup>bcd</sup>	55.5±3.16 <sup>b</sup>	14.23±0.24 <sup>a</sup>	0.093±0.06 <sup>bcd</sup>
G8- (HC) +30% avocado fruit	117.9±1.41 <sup>ab</sup>	184.3±2.61 <sup>a</sup>	66.4±3.41 <sup>a</sup>	14.37±0.44 <sup>a</sup>	0.110±0.15 <sup>a</sup>
G9 (HC) +10% avocado seeds	118±1.41 <sup>ab</sup>	171.3±3.22 <sup>cd</sup>	53.3±3.69 <sup>bc</sup>	13.38±0.57 <sup>cd</sup>	0.095±0.18 <sup>bc</sup>
G10(HC) +20% avocado seeds	119±1.26 <sup>a</sup>	173.5±2.28 <sup>bc</sup>	54.5±2.28 <sup>b</sup>	13.63±0.4 <sup>bc</sup>	0.095±0.13 <sup>bc</sup>
G11(HC) +30% avocado seeds	117.8±1.41 <sup>ab</sup>	173.3±2.00 <sup>bc</sup>	55.2±2.28 <sup>b</sup>	13.93±0.42 <sup>abc</sup>	0.094±0.12 <sup>bc</sup>

°Initial body weight, § Final body weight, \*Body weight gain, †Food intake, ‡Feed efficiency ratio  
Mean with the same letters in the same horizontal column are not significantly different at P<0.05.

Table 7: Changes weight of organs in the hypercholesterolemic rats fed diet supplemented with kiwi fruit and avocado (fruit and seed).

Groups	Liver (g) Mean ± SD	Heart (g) Mean ± SD	Kidney (g) Mean ± SD	Spleen (g) Mean ± SD
	G1 control (-)	6.1±0.67 <sup>a</sup>	0.500±0.08 <sup>ab</sup>	1.1250±0.19 <sup>a</sup>
G2 (HC) (+)	7.12±0.91 <sup>b</sup>	0.550±0.05 <sup>a</sup>	1.25±0.17 <sup>a</sup>	0.325±0.05 <sup>ab</sup>
G3(HC)+10% Kiwi	6.42±0.92 <sup>a</sup>	0.475±0.05 <sup>ab</sup>	1.125±0.26 <sup>a</sup>	0.300±0.41 <sup>ab</sup>
G4 (HC) +20% Kiwi	6.62±0.45 <sup>a</sup>	0.450±0.05 <sup>ab</sup>	0.975±0.15 <sup>a</sup>	0.275±0.05 <sup>ab</sup>
G5 (HC) +30% Kiwi	6.22±0.96 <sup>a</sup>	0.450±0.05 <sup>ab</sup>	1.125±0.45 <sup>a</sup>	0.350±0.01 <sup>a</sup>
G6 (HC) +10% avocado	5.72±0.76 <sup>a</sup>	0.450±0.05 <sup>ab</sup>	1.025±0.15 <sup>a</sup>	0.275±0.05 <sup>ab</sup>
G7 (HC) +20% avocado	6.47±0.71 <sup>a</sup>	0.450±0.129 <sup>ab</sup>	1.050±0.05 <sup>a</sup>	0.300±0.1 <sup>ab</sup>
G8- (HC) +30% avocado	6.17±1.22 <sup>a</sup>	0.450±0.057 <sup>ab</sup>	1.00±0.18 <sup>a</sup>	0.300±0.1 <sup>ab</sup>
G9 (HC) +10% avocado seeds	5.60±1.26 <sup>a</sup>	0.450±0.057 <sup>ab</sup>	1.025±0.15 <sup>a</sup>	0.275±0.05 <sup>ab</sup>
G10(HC) +20% avocado seeds	5.60±1.26 <sup>a</sup>	0.440±0.050 <sup>ab</sup>	1.025±0.16 <sup>a</sup>	0.275±0.05 <sup>ab</sup>
G11(HC) +30% avocado seeds	6.05±0.085 <sup>a</sup>	0.475±0.050 <sup>ab</sup>	1.10±0.16 <sup>a</sup>	0.275±0.05 <sup>ab</sup>

Mean with the same letters in the same horizontal column are not significantly different at P<0.05.

Table 8: Effect of kiwi fruit and avocado (fruit and seed) on the serum lipid profiles of hypercholesterolemic rats.

Groups	*TG mg/dl Mean ± SD	°TC mg/dl Mean ± SD	†HDL-C mg/dl Mean ± SD	‡LDL-C mg/dl Mean ± SD
	G1 control (-)	131.3±2.61 <sup>k</sup>	83.5±2.28 <sup>h</sup>	33.3±2.61 <sup>a</sup>
G2 (HC) (+)	273.2±2.83 <sup>a</sup>	137.0±3.69 <sup>a</sup>	23.3±3.16 <sup>c</sup>	59.1±2.28 <sup>a</sup>
G3(HC)+10% Kiwi fruit	219.1±2.83 <sup>c</sup>	122.6±3.41 <sup>b</sup>	27.3±2.99 <sup>b</sup>	51.5±2.98 <sup>b</sup>
G4 (HC) +20% Kiwi fruit	203.5±3.16 <sup>c</sup>	117.5±2.61 <sup>c</sup>	28.5±3.42 <sup>b</sup>	48.3±2.28 <sup>b</sup>
G5 (HC) +30% Kiwi fruit	195.9±3.22 <sup>e</sup>	117.6±3.35 <sup>c</sup>	29.7±2.61 <sup>ab</sup>	42.8±2.28 <sup>c</sup>
G6 (HC) +10% avocado fruit	212.3±2.83 <sup>d</sup>	119.9±3.41 <sup>bc</sup>	27.8±3.16 <sup>b</sup>	49.6±2.83 <sup>b</sup>
G7 (HC) +20% avocado fruit	199.2±2.61 <sup>f</sup>	113.6±2.28 <sup>d</sup>	29.3±2.18 <sup>b</sup>	44.5±2.98 <sup>c</sup>
G8- (HC) +30% avocado fruit	168.8±2.83 <sup>i</sup>	106.2±3.69 <sup>e</sup>	30.6±3.32 <sup>ab</sup>	41.8±2.00 <sup>c</sup>
G9 (HC) +10% avocado seeds	232.2±2.83 <sup>b</sup>	112.1±3.41 <sup>d</sup>	28.5±2.83 <sup>b</sup>	37.2±3.25 <sup>d</sup>
G10(HC) +20% avocado seeds	181.7±2.0 <sup>h</sup>	99.5±2.83 <sup>f</sup>	29.9±3.22 <sup>ab</sup>	33.3±3.31 <sup>e</sup>
G11(HC) +30% avocado seeds	155.2±2.0 <sup>j</sup>	93.2±2.28 <sup>g</sup>	31.3±2.92 <sup>ab</sup>	30.9±2.83 <sup>e</sup>

\*Triacylglycerol, °Total Cholesterol, †High density lipoprotein Cholesterol, ‡Low density lipoprotein Cholesterol  
Mean with the same letters in the same horizontal column are not significantly different at P<0.05

Meanwhile, there are no significant difference was observed in feed intake of all groups. The reduced in body weight gain of kiwi and avocado fruit may be due to higher content of crude fiber (11.22% and 12.84%). The fiber reduced the gastric emptying rate and makes it possible for rats to feel full, which delaying the absorption and digestion of nutrients and reduced feed intake which lead to decrease body weight gain [59]. Our results are in agreement with Kim *et al.* [60], Shehata and Soltan [61], they reported hypercholesterolemic diet was supplemented with food rich in fiber (mulberry leaf

powder, purslane and celery fresh) lead to decreases in body weight gain and the reduction of FER were depended on the levels of dietary fiber.

Effects of different concentration of kiwi fruit and avocado (fruit and seed) on weight of the main organs of hypercholesterolemic rat are shown in Table 7. Results revealed that liver weight was markedly increased in rats fed hypercholesterolemic diet compared to normal control group. Meanwhile, there are no changed in different other weight of organs. Supplemented diet of hypercholesterolemic rat with kiwi fruit and avocado

(fruit and seed) reduced liver weight compared to HC group. The lowering effect of avocado fruit and seed may be due to increases catabolism of lipid accumulated in adipose tissue causing a decrease in body weight. These results are in line with those obtained by Ojewele and Amabeark [62], who reported that the methanol extract of avocado leave provoked animal 8% decrease in mean liver weight compared to the hyperlipidemic control group.

The results of lipids profile which are presented in Table 8. Data clarified that hypercholesterolemic rats HC group recorded significant increase  $P < 0.05$  in TC, TG and LDL-c concentration as well as significant decrease in the level of good cholesterol HDL-c compared to healthy control group. These results are in agreement with Harnafi *et al.* [38] and Kumer *et al.* [63], they reported TC, TG and LDL-c showed significantly higher in hypercholesterolemic group than normal control group. Supplemented diet of hypercholesterolemic rats with different concentration of kiwi fruit and avocado (fruit and seed) lead to significantly decrease  $P < 0.05$  in TG, TC and LDL-c concentration as well as significant increase in level of good cholesterol HDL-c compared to HC group. The decrease in TC, TG and LDL-c level and increases in HDL-c were increases with increasing concentration of kiwi and avocado (fruit and seed). The present study indicated that the best results of lipid profile were observed of high concentration (30%) of kiwi fruit and avocado (fruit and seed). Feeding hypercholesterolemic rat on diet supplemented with 30% kiwi fruit reduced TC, TG and LDL-c by 14.16%, 28.29% and 27.58%, respectively and increased HDL-c by 27.46%. These results may be due to the micronutrient such as vitamin C (15.52%) and present phenolic compounds (258.55 mg/100g GAE), act as antioxidant to scavenge free radical and can delay or inhibit the oxidation of both lipid and other molecules [9]. These results are in line with those obtained by Maria *et al.* [64], who reported that diet containing kiwi fruit decrease of TG (61%), TC (29%) and LDL-c (38%). Chang and Liu [65] and Lim [66], they mentioned that regular consumption of kiwi fruit (Two kiwi fruit per day) might exert beneficial in hyperlipidemic subject.

Treatment hypercholesterolemic rat with 30% avocado fruit reduced TC, TG and LDL-c by 22.48%, 38.21% and 29.27%, respectively and increases HDL-c by 31.33% as compared to the HC group. These results may be due to avocado fruit are rich in monounsaturated fatty acid, fiber (12.84%), flavonoids (2.96 mg/100g CE), phenolic compound (259.15mg/100g GAE) and sterols.

These results are accordance with those reported by Mohammed [67], who revealed that the avocado fruit pulp administrated at doses 1 and 2ml/day/rat for ten week caused a significant decrease in the serum lipid including TC and TG levels and increased in HDL-c. Treatment hypercholesterolemic rat with 30% avocado seed reduced TC, TG and LDL-c by 31.97%, 43.19% and 47.72% respectively and increased HDL-c by 34.33% compared to HC group. The lowering effect of avocado seed of TC, TG and LDL-c may be due to the avocado seed had highest phenolic compound (285.43 mg/100g GAE) and flavonoid compounds (3.21mg/100g CE), soluble dietary fiber (38%) compared to kiwi and avocado fruit. These results agreement with Anderson [68] mentioned that dietary fiber, especially soluble, effectively decrease serum cholesterol and LDL-c concentration. Soluble fiber principle effects on cholesterol metabolism through decrease in bile acid absorption. These soluble fiber bind bile acids in the small intestine alter micelle formation and decrease their absorption, in the small intestine. Consequently, more bile acid is excreted with the feces. Flavonoid (Rosmarinic, Quercetin, Apigenin and Hesperdine) and phenolic compounds (pyrogallol, syringic, catchin, procatechuic and catecal) which highly content in avocado seed acts as antioxidant protect of lipid peroxidation and scavenging to free radicals. Hung *et al.* [69] reported that flavonoid (quercetin, hesperidins' and narnigin) have been shown to inhibit the generation or release of free radicals derived from lipoxygenase.

These results are in agreement with Asaolu *et al.* [70], who reported treatment hypercholesterolemic rat with various doses of methanolic of avocado seed (50, 100 and 200 ml) caused a significant reduction in the levels of TC, TG and LDL-c.

Data in Table 9 showed that the Levels of AI, LDL-C/HDL-C Ratio and HTR ratio of hypercholesterolemic rats administered diet supplemented with kiwi fruit, avocado fruit and avocado seed. Atherogenic index and LDL-C/HDL-C ratio increased significantly (4.9 and 2.5), while HTR% decreased significantly (17.0) in the HC group in comparison with the control group (1.5, 0.72 and 39.9), respectively. The atherogenic index and LDL-C/HDL-C ratio decreased according to the amount of kiwi fruit and avocado (fruit, seed) added in comparison with HC group and HTR% increased according to the amount of kiwi fruit and avocado (fruit, seed) added in comparison with HC group. The best level of kiwi fruit and avocado (fruit, seed) was 30% administration for hypercholesterolemic rats as compared to the HC group. Supplementation of the

Table 9: Levels of AI, LDL-C/HDL-C ratio and HTR of hypercholesterolemic rats fed diet supplemented with kiwi fruit and avocado (fruit and seed).

Groups	*AI	LDL/HDL	HTR
	Mean± SD	Ratio	Ratio
G1 control (-)	1.5±0.77 <sup>c</sup>	0.72±0.28 <sup>c</sup>	39.9±2.83 <sup>a</sup>
G2 (HC) (+)	4.9±1.77 <sup>a</sup>	2.5±1.14 <sup>a</sup>	17.0±2.83 <sup>f</sup>
G3(HC)+10% Kiwi fruit	3.5±1.58 <sup>b</sup>	1.9±0.85 <sup>ab</sup>	22.3±2.92 <sup>e</sup>
G4 (HC) )+20% Kiwi fruit	3.1±0.79 <sup>b</sup>	1.7±0.46 <sup>abc</sup>	24.3±2.79 <sup>e</sup>
G5 (HC) +30% Kiwi fruit	3.00±1.14 <sup>bc</sup>	1.4±0.62 <sup>bcd</sup>	25.3±3.22 <sup>de</sup>
G6 (HC) +10% avocado fruit	3.3±1.04 <sup>b</sup>	1.8±0.77 <sup>abc</sup>	23.2±3.24 <sup>e</sup>
G7 (HC) )+20% avocado fruit	2.9±1.36 <sup>bc</sup>	1.5±0.54 <sup>bcd</sup>	25.8±2.83 <sup>cd</sup>
G8- (HC) )+30% avocado fruit	2.5±0.71 <sup>bc</sup>	1.4±0.51 <sup>bcd</sup>	28.8±2.83 <sup>cd</sup>
G9 (HC) )+10% avocado seeds	2.9±1.49 <sup>bc</sup>	1.3±0.54 <sup>bcd</sup>	25.4±3.53 <sup>de</sup>
G10(HC) )+20% avocado seeds	2.3±1.04 <sup>bc</sup>	1.1±0.46 <sup>bcd</sup>	30.1±2.91 <sup>e</sup>
G11(HC) )+30% avocado seeds	2.0±0.85 <sup>bc</sup>	1.0±0.49 <sup>cd</sup>	33.6±2.28 <sup>b</sup>

\*Atherogenic index

Mean with the same letters in the same horizontal column are not significantly different at P<0.05

Table 10: Activities of AST and ALT in serum of hypercholesterolemic rats fed diet supplemented with kiwi fruit, avocado (fruit and seed).

Groups	*AST U/mL	°ALTU/mL
	Mean± SD	Mean± SD
G1 control (-)	79.5±2.61 <sup>g</sup>	32.3±3.41 <sup>e</sup>
G2 (HC) (+)	123.5±2.28 <sup>a</sup>	56.5±2.61 <sup>a</sup>
G3(HC)+10% Kiwi fruit	113.6±3.16 <sup>b</sup>	47.9±3.41 <sup>b</sup>
G4 (HC) )+20% Kiwi fruit	104.5±2.28 <sup>c</sup>	43.8±2.28 <sup>c</sup>
G5 (HC) +30% Kiwi fruit	92.0±3.22 <sup>e</sup>	39.5±3.41 <sup>d</sup>
G6 (HC) +10% avocado fruit	104.1±2.61 <sup>c</sup>	44.9±3.16 <sup>c</sup>
G7 (HC) )+20% avocado fruit	97.6±2.83 <sup>d</sup>	39.4±2.61 <sup>d</sup>
G8- (HC) )+30% avocado fruit	85.6±1.41 <sup>f</sup>	34.6±3.16 <sup>e</sup>
G9 (HC) )+10% avocado seeds	110.8±2.28 <sup>b</sup>	44.4±2.28 <sup>c</sup>
G10(HC) )+20% avocado seeds	95.5±3.22 <sup>d</sup>	38.5±2.0 <sup>d</sup>
G11(HC) )+30% avocado seeds	82.2±2.83 <sup>g</sup>	33.8±1.79 <sup>e</sup>

\*Aspartate aminotransferase, °Alanine aminotransferase.

Mean with the same letters in the same horizontal column are not significantly different at P<0.05.

Table 11: Levels of total cholesterol, triglyceride and GSH in liver tissue of hypercholesterolemic rats fed diet supplemented with kiwi fruit and avocado (fruit and seed).

Groups	*TG	°TC	†GSH
	Mean ± SD	Mean ± SD	Mean ± SD
G1 control (-)	12.2±2.35 <sup>d</sup>	2.42±1.16 <sup>d</sup>	4.9±1.41 <sup>a</sup>
G2 (HC) (+)	18.9±3.35 <sup>a</sup>	3.8±1.25 <sup>a</sup>	3.0±1.14 <sup>c</sup>
G3(HC)+10% Kiwi fruit	17.1±3.31 <sup>ab</sup>	3.61±1.56 <sup>ab</sup>	3.69±1.43 <sup>b</sup>
G4 (HC) )+20% Kiwi fruit	16.4±3.51 <sup>abc</sup>	3.41±2.25 <sup>abc</sup>	3.75±1.67 <sup>b</sup>
G5 (HC) +30% Kiwi fruit	15.0±2.83 <sup>bcd</sup>	3.2±1.52 <sup>bc</sup>	4.08±1.45 <sup>b</sup>
G6 (HC) +10% avocado fruit	16.7±3.41 <sup>abc</sup>	3.42±2.55 <sup>abc</sup>	3.84±1.42 <sup>b</sup>
G7 (HC) )+20% avocado fruit	15.0±2.28 <sup>bcd</sup>	3.19±3.21 <sup>bc</sup>	4.17±2.02 <sup>b</sup>
G8- (HC) )+30% avocado fruit	13.6±2.6 <sup>bcd</sup>	3.04±2.12 <sup>bc</sup>	4.35±2.2 <sup>ab</sup>
G9 (HC) )+10% avocado seeds	15.0±3.22 <sup>bcd</sup>	3.34±1.92 <sup>abc</sup>	3.87±1.51 <sup>b</sup>
G10(HC) )+20% avocado seeds	14.6±2.61 <sup>bcd</sup>	3.15±2.31 <sup>bc</sup>	4.11±1.44 <sup>b</sup>
G11(HC) )+30% avocado seeds	12.8±1.88 <sup>cd</sup>	2.89±1.71 <sup>cd</sup>	4.4±1.76 <sup>ab</sup>

\*Triglycerides, °Total Cholesterol, †Reduced Glutathione.

Mean with the same letters in the same horizontal column are not significantly different at P<0.05.

Cholesterol diet with 30% kiwi fruit and avocado (fruit, seed) improved the atherogenic index (AI) by about 38.8%, 49% and 59.2%, respectively. These results are in line with those obtained by Maria *et al.* [64], who reported that diets containing kiwi fruits significantly decreased in the value of the atherogenic index by about 32%. These results also are accordance with the findings reported by Chang and Liu [65] and Lim [66], who indicated that after 8 weeks kiwi fruit consumption LDL-C/HDL-C ratio was significantly decreased in hyperlipidemic suffered subjects compared with hyperlipidemic subjects not consumed kiwi fruit. These results are in agreement with Maria *et al.* [71], who reported that treatment of mice with 125 mg avocado seed flour/kg BW significantly reduced the elevated level of AI by 50.6% compared to hypercholesterolemic mice. The atherogenic index markedly decreased causing a reduction in LDL-C/HDL-C ratio in all groups fed diet supplemented with kiwi fruit and avocado (fruit, seed). Our results agree with Makni *et al.* [72], who stated that the increase in HDL-C or HTR ratio is one of the most important criteria of anti-hypercholesterolemic agent.

Effect supplemented diet of hypercholesterolemic rats with kiwi fruit and avocado (fruit and seed) on activities of AST and ALT are summarized in Table 10. Supplemented diet with different concentration of kiwi fruit and avocado (fruit and seed) reduced the AST and ALT activities compared to HC group. The best recorded of liver function was observed of higher concentration (30%) of all treatment. Supplemented diet with 30% kiwi fruit and avocado (fruit and seed) reduced AST and ALT by (25.51% and 30.09%, 30.69% and 38.76%, 33.44% and 40.18%) respectively. In this study, it is expected that the adding kiwi fruit and avocado (fruit and seed) to hypercholesterolemic diet well be effective for the recovery the hepatic function by improvement of lipid metabolism or delaying the hepatic disease. These results are in agreement with those reported by Mohammed [67], who indicated that rats which consumed 1 or 2 ml/day/rat avocado extract for 70 days showed decrease in AST and ALT activity compared to the control group. Also, Mohamed and Amr [73] reported that administration of dried avocado fruit at the three tests levels (5, 10 and 15%) caused lower of serum AST and ALT content compared to the control group.

Data in Table 11 illustrated that lipid level of liver tissue. High cholesterol diet caused significant increase p<0.05 of hepatic cholesterol (54.92%) and triacylglycerol (57.02%) compared to the healthy control group. Administration hypercholesterolemic rat diet

supplemented with kiwi fruit and avocado (fruit and seed) at all levels reduced hepatic TC and TG. Meanwhile, the diet supplemented with avocado seed at all three concentration was the best to reduced hepatic lipid. Polyphenolics compounds of plants decrease the cholesterol level of liver tissue [74]. Liver cholesterol lowering effect may be due to avocado seed reduced absorption of cholesterol and fat or increases the fecal excretion of fat and cholesterol [75]. The hypercholesterolemic rat's administration diet supplemented with 30% kiwi fruit and avocado (fruit and seed) had 20.6%, 28% and 32.2%, respectively, reduction in TG level in liver tissue. These results are in accordance with Imafidon and Amaectina [76], who reported that the hypertensive rats treated with 500 mg/kg avocado seed extract reduced TG by 36.19% compared to the control group.

Data in Table 11 also showed that the content of liver reduced glutathione (GSH) of hypercholesterolemic rats administered diet supplemented with kiwi fruit and avocado (fruit, seed). The content of GSH decreased significantly in the HC group in comparison with the normal group, while the groups fed the kiwi fruit and avocado (fruit, seed) GSH significantly increased with increases levels of kiwi fruit and avocado (fruit and seed) compared to HC group. Our results are in accordance with the data reported by Sadek *et al.* [77], who indicated that rats which consumed kiwi fruit caused a significant increase in glutathione content when compared to control rats.

## CONCLUSIONS

The present study demonstrated that consumption kiwi fruit, avocado (fruit and seed) can modulate the risk factors of CVD (Cardiovascular diseases) by reducing the LDL-C, LDL-C/HDL-C and increasing HDL-C and HTR ratio. The results suggested that consumption of kiwifruit, avocado fruit and avocado seed might have some cardiovascular protective properties and beneficial effects on atherosclerosis, CVD risks in hypercholesterolemic rats.

## REFERENCES

1. Rerkasan, K., P.J. Gallagher, R.F. Grimble, P.C. Calder and C.P. Shearman, 2008. Managing hypercholesterolemia and its correlation with carotid plaque morphology in patients undergoing carotid endarterectomy (A Review). *Vascular Health and Risk Management*, 4(6):1259-1264.
2. Ramachandran, H.D., K. Narasimhamurthy and P.L. Raina, 2003. Modulation of Cholesterol induced hypercholesterolemia through dietary factors in Indian desert gerbils (*Merionesurriace*). *Nutrition Research*, 23: 245-256.
3. Matos, S.L. H. Paula, M.L. Pedrosa, R.C. Santos, E.L. Oliveira, J.D. Chianca and M.E. Silva, 2005. Dietary Models for inducing Hypercholesterolemia in rats. *Brazilian Archives of Biology and Technology*, 48(2): 203-209.
4. Asashina, M., M. Sato and K. Imaizumi, 2005. Genetic analysis of diet induced hypercholesterolemia in exogenously hypercholesterolemic (ExHC) rats. *Journal of Lipid Research*, 46: 2289-2294.
5. Ichihashi, T., M. Izawa, K. Miyata, T. Mizui, K. Hirano and Y. Takagishi, 1998. Mechanisms of hypercholesterolemic action of S-89211 in rats: S-8921 inhibits ileal bile acid absorption. *Journal of Pharmacology and Experimental Therapeutics (JPET)*, 284(1): 43-50.
6. Paredes-López, O., M. Cervantes-Ceja, M. Vigna-Pérez and T. Hernandez-Perez, 2010. Berries: improving human health and healthy aging and promoting quality life-A review. *Plant Foods Hum Nutr.*, 65: 299-308.
7. Duttaroy, A.K. and A. Jorgensen, 2004. Effects of kiwi fruit consumption on platelet aggregation and plasma lipids in healthy human volunteers. *Platelets*, 15: 287-292.
8. Larocea, M., R. Rossano and P. Riccio, 2010. Analysis of green kiwi fruit (*Actinidia deliciosa* cv. Hayward) proteinases by two-dimensional zymography and direct identification of zymographic spots by mass spectrometry. *Journal of the Science of Food and Agriculture*, 90: 2411- 2418.
9. Fiorentino, A., C. Mastellone, B. D'Abrosca, S. Pacifico, M. Scognamiglio, G. Cefarelli, R. Caputo and P. Monaco, 2009.  $\Delta$ -Tocomonoenol: A new vitamin E from kiwi (*Actinidia chinensis*) fruits. *Food Chem.*, 115: 187-192.
10. Sarbu, C., R.D. Masco-Briciu, A. Kot-Wasik, S. Gorinstein, A. Wasik and J. Namiesnik, 2012. Classification and finger printing of kiwi and pomelo fruits by multivariate analysis of chromatographic and spectroscopic data. *Food Chem.*, 130: 994-1002.
11. Park, Y.S., S.H. Kyung, S.G. Kang, Y.K. Park, J. Namiesnik, H. Leontowicz, M. Leontowicz, A. Ezra, S. Trachtenberg and S. Gorinstein, 2012. Organic and conventional kiwifruit, myths versus reality: antioxidant, anti-proliferative and health effects. *J. Agric. Food Chem.*, 60: 6984-6993.

12. Cassano, A., A. Figoli, A. Tagarelli, G. Sindono and E. Drioli, 2006. Integrated membrane process for the production of highly nutritional kiwi fruit juice. *Desalination*, 189: 21-30.
13. Dehghani, F., T. Talael-Khozani, M.R. Panjehshahin and Z. Panahi, 2006. Toxic effects of hydroalcoholic extract of kiwi (*Actinidia chinensis*) on histological structure of the male Sprague-Dawley rat reproductive tissues. *Iranian J. Sci. and Technol. Transduction A*, 30(A1): 19-25.
14. Hunter, D.C., M.A. Skinner, A.R. Ferguson and L.M. Stevenson, 2010. *Kiwi Fruit and Health*. The New Zealand Institute for Plant and Food Research Ltd, Auckland, New Zealand, 2<sup>nd</sup> Edition, pp: 565 -580.
15. Jung, K.A., T.C. Song, D. Hand, D. Kim, I.H. Kim, Y.E. Kim and C.H. Lee, 2005. Cardiovascular protective properties of kiwi extract *in vivo*. *Biological and Pharmaceutical Bulletin*, 28: 1782-1785
16. Collins, B.H., A. Horsk and P.M. Hotten, 2001. Kiwi fruit protects against oxidative DNA damage in human cell and *in vitro*. *Nutr. Cancer*, 39(1): 148- 153.
17. Bekhradina, S., S.M. Nabavi, S.F. Nabavi and M.A. Ebrahimza, 2011. The extract of kiwi fruit (*Actinidia chinensis* L. *syn. A. delicious*) exhibited different levels of antioxidant activity. *Pharmacology on Line*, 1: 758- 764.
18. Lee, J., N. Koo and D. Minm, 2004. Reactive oxygen species, aging and antioxidative nutraceuticals. *Comprehensive Review in Food Science and Food Safety*, 3: 21- 33.
19. Rainey, C., M. Affleck, K. Bretschger and R.B. Alfin-Slater, 1994. The California avocado, a new look. *Nutr. Today*, 29: 23-27.
20. Bergh, B., 1992. *Nutritious Value of Avocado*. California Avocado Society Book, CA. pp: 123- 135.
21. Lans, C.A., 2006. Ethnomedicines used in Trinidad and Tobago for urinary problems and diabetes mellitus. *Journal of Ethnobiology and Ethnomedicine*, 2: 45-55.
22. Agra, M.F., P.F. Ferities and J.M. Barbosa-Filho, 2007. Synopsis of the plants known as medicinal and poisonous in Northeast of Brazil. *Brazilian Journal of Pharmacognosy*, 17: 114-140.
23. Andrade-Cetto, A. and M. Heinrich, 2005. Mexican plants with hypoglycemic effect used in the treatment of diabetes. *Journal of Ethnopharmacology*, 99: 325-348.
24. Andrade-Cetto, A., J. Becerra-Jimenez, E. Martinez-Zurita, E.P. Ortega-Larrocea and M. Heinrich, 2006. Disease -consensus index as a tool of selecting potential hypoglycemic plants in Chikindzonot, Yucatan, Mexico. *Journal of Ethnopharmacology*, 107: 199-204.
25. Adeboye, J.O., M.O. Fajonyomia, J.M. Makindeb and O.B. Taiwob, 1999. A preliminary study on the hypertensive activity of *Persea americana* leaf extracts in anaesthetize dnormotensive rats. *Fitoterapia*, 70: 15-20.
26. Di' Stasi, L.C., G.P. Oliveira, M.A. Carvalhaes, M. Queiroz-Junior, O.S. Tien, S.H. Kakinami and M.S. Reis, 2002. Medicinal plants popularly used in the Brazilian tropical Atlantic forest. *Fitoterapia*, 73: 69-91.
27. Owolabi, M.A., S.I. Jaja and H.A.B. Coker, 2005. Vasorelaxant action of aqueous extract of the leaves of *Persea americana* on isolated thoracic rat aorta. *Fitoterapia*, 76: 567-573.
28. Owolabi, M.A., H.A.B. Coker and S.I. Jaja, 2011. Bioactivity of the phytoconstituent of the leaves of *Persea americana*. *Journal of Medicinal Plant Research*, 4: 1130-1135.
29. Brai, B.I., A.A. Odetola and P.U. Agomo, 2007. Hypoglycemic and hypocholesterolemic potential of *Persea americana* leaf extract. *Journal of Medicinal Food*, 10: 356-360.
30. Biglari, F., F.M. Abbas and M.E. Azhar, 2008. Antioxidant activity and phenolic content of various date palm (*Phoenix dactylifera*) fruit from Iran. *Food Chemistry*, 107: 1636- 1641.
31. A.O.A.C. 2000. *Official Methods of Analysis*. 17<sup>th</sup> Ed. of AOAC International published by AOAC International suit 500, 481 North Frederick Avenue, Gaithersburg. Maryland.
32. Asami, K.D., Y.J. Hong, M.D. Barrett and E.A. Mitchell, 2003. Composition of total phenolic and ascorbic acid content of freeze dried and air-dried marinoberry, strawberry and corn grown using conventional organic and sustainable agricultural practices. *J Agric and Food Chem.*, 51: 237-1241.
33. Khatiwara, E., V.B. Adsul, M.M. Kulkarzi, N.R. Deshpande and R.V. Kashalkar, 2010. Spectroscopic determination of total phenol and flavonoid content of *Ipomoea caruea*. *International Journal Chem. Tech Research*, 2(3): 698-1701.

34. Goupy, P., M. Hugues, P. Biovin and M.J. Amiot, 1999. Antioxidant composition and activity of barley (*Hordeum vulgare*) and malt extracts and isolated phenolic compounds. J. Sci. Food Agric., 79: 1265-1634.
35. Mattila, P., J. Astola and J. Kumpulainen, 2000. Determination of flavonoids in plant material by HPLC with diode- array and electro- array detection. J. Agric. Food Chem., 48: 5834- 5841.
36. Brand-Williams, S.W., M.E. Cuvelier and C. Berset, 1995. Use of a free radical method to evaluate antioxidant activity. Food Science and Technology, 28: 25-30.
37. Reeves, R., G. F.H. Nielsen and G.C. Fahey, 1993. AIN-93 purified diets for laboratory rodents. J. Nutr., 123: 1939.
38. Harnafi, H., M. Aziz and S. Amrani, 2009. Sweet basil (*Ocimum basilicum* L.) improves lipid metabolism in hypercholesterolemic rats. Span. The European Journal of Clinical Nutrition and Metabolism, 4: e181- e186.
39. Richmond, N., 1973. Preparation properties of a cholesterol oxidase from *Nocardia* sp. enzymatic assay of total cholesterol in serum. Clin. Chem., 19: 1350-1356.
40. Burstein, M.H., R. Scholnick and R. Haarfin, 1970. Rapid method for isolation of lipoprotein from human serum by precipitation with polyamine. Lipid Research, 11: 385- 395.
41. Jacobs, N.J. and P.K. Vander mark, 1960. Determination of serum triglycerol. Arch. Biochem. Biophys. 88: 250.
42. Reitman, S. and S. Frankel, 1957. A calorimetric Method for Determination of Serum AST. Am. J. Clin. Path., 18: 26.
43. Friedewald, W.T., K.T. Levy and D.S. Fredrickson, 1972. Estimation of the Concentration of low density lipoprotein Cholesterol in Plasma Without use of the Preparative Ultracentrifuge. Clin. Chem., 226: 499-504.
44. Hostmark, H.A., 1987. Lipoprotein lipases, lipoprotein and tissue lipids rats feed fish oil and or coconut oil. Journal of Nutrition, 117: 1011-1017.
45. Beutler, E., O. Duran and B.M. Kelly, 1963. Improved method for the determination of blood glutathione. J. Lab. and Clin. Med., 61(5): 882-888.
46. Bailey, N.T., 1995. Statistical Method in Biology. 3<sup>rd</sup> Cambridge Univ. Press Cambridge.
47. Reyes-Caudello, E., A. Tecante and M.A. Valdiria-Lopez, 2008. Dietary fiber content and antioxidant activity of phenolic present in Mexican chia (*Salvia hispanica* L.) seeds. Food Chem., 107(2): 656- 663.
48. Park, Y.S., H. Leontowicz, M. Leontowicz, J. Namiesnik, M. Suhaj, M. Cvikrova, O. Martincova, M. Weiss and S. Gorinstein, 2011. Comparison of the contents of bioactive compounds and the level of antioxidant activity in different kiwifruit cultivars. J. Food Comp. Anal., 24: 963-970.
49. Gorinstein, S., P. Sumitra, L. Hanna, L. Maria, N. Jacek, V. Suchada, H. Ratiporn, R. Pramoj, K. Elena and T. Zer, 2011. Antioxidant properties and bioactive constituents of some rare exotic Thai fruits and comparison with conventional fruits in vitro and in vivo studies. Food Research International, 44: 2222-2232.
50. Wang, W., T.R. Bostic and L. Gu, 2010. Antioxidant capacities, procyanidin and pigments in avocados of different strains and cultivars. Food Chem., 122: 1193-1198.
51. Soong, Y.Y. and P.J. Barlow, 2004. Antioxidant activity and phenolic content of selected fruit seeds. Food Chemistry, 88: 411- 417.
52. Der-Duh, P. and G. Chin Yen, 1997. Antioxidant activity of three herbal water extracts. Food Chemistry, 60(4): 645- 645.
53. Yuka, L., K. Yumiko, N. Miyo and K. Takashi, 2003. Antioxidant activity of tropical fruit. Feijae sellowanaberg. Nippon Kasei Gakkaishi, 54: 945- 949.
54. Rodriguez-Carpena, J.G., D. Morcuende and M. Esterez, 2011. Avocado by products as inhibitors of color determination and lipid and protein oxidation in raw porcine patties subjected to chilled storage. Meat Science, 89: 166-173.
55. Kosinska, A., M. Karamae, I. Estrella, T. Hernandez, B. Bartolome and G.A. Dykes, 2012. Phenolic compound profiles and antioxidant capacity of *Persea americana* Mill. Peels and seeds of two varieties. J. Agric. and Food Chem., 60(18): 4613-4619.
56. Pahlua-Romos, M.E., A. Certiz-Moreno, G. Chanorro-Cevallos, M.D. Hernandez- Navarro, L.G. Garduno-Siciliano, H. Necochea-Mondragon and M. Hernandez-Ortega, 2012. Hypolipidemic effect of avocado (*Persea americana* Mill) seed in a hypercholesterolemic mouse model. Plant Food Hum. Nutr., 67: 10-16.
57. Lecumberri, E., L. Goya and R. Mateos, 2007. A diet rich in dietary fiber from cocoa improves lipid profile and reduced malondialdehyde in hypercholesterolemic rats. Nutrition, 23: 332-341.
58. Baraket, A. and A. Lamiaa, 2011. Hypolipidemic and anti atherogenic effects of dietary chitosan and wheat bran in high fat-high cholesterol fed rats. Australian Journal of Basic and Applied Science, 5(10): 30-37.

59. Torsdottir, I., M. Alpsten, G. Holm, A.S. Sandberg and T.J. Llim, 1991. A small dose of soluble alginate- fiber affects postprandial glycemia and gastric emptying in human with diabetes. *Journal of Nutrition*, 121: 795-799.
60. Kim, A.J., S.Y. Kim, M.K. Choi, M.H. Kim and K.S. Chang, 2005. Effects of mulberry leave powder on lipid metabolism in high cholesterol fed rats. *Korean J. Food Sci. and Technol.*, 37: 636- 641.
61. Shetata, M.S.M. and S.S.A. Soltan, 2012. The effects of purslane and celery on hypercholesterolemic mice. *World Journal of Dairy and Food Sciences*, 7(2): 212-221.
62. Ojewele, J.A.O. and G.J. Amabeark, 2006. Anticonvulsant effect of *Persea americana* Mill. (Lauraceae) avocado leaf aqueous extract in mice. *Phytother Res.*, 20(8): 696-700.
63. Kumar, D., V. Parcha, F. Dhulia and A. Maithani, 2011. Evaluation of anti-hyperlipidemic activity of method extract *Salvador olooides* (Linn) leaves in Triton WR- 1339 (Tyloxaol) induced hyperlipidemic rats. *J. of Pharmacy Res.*, 4: 512- 513.
64. Maria, L., J. Iowan, L. Hanna, P. Yong-Seo, A. Jacek, D.R. Adamo, W. Moshe and G. Sheila, 2013. Health-promoting effects of ethylene-treated kiwifruit 'Hayward' from conventional and organic crops in rats fed an atherogenic diet. *J. Agric. Food Chem.*, 61: 3661-3668.
65. Chang, W. and J. Liu, 2009. Effects of kiwi fruit consumption on serum lipids profiles and anti-oxidative status in hyperlipidemic subjects. *Intern. J. Food Sci. and Nutr.*, 60(8): 709-716.
66. Lim, T.K., 2012. *Edible Medicinal and Non-medicinal Plants. (Fruit)*. 1<sup>st</sup> Edition. New York. pp: 20-29.
67. Mohammed, S.A., 2011. Hypolipdemic and antioxidant activities of avocado fruit pulp on high cholesterol fed diet in rats. *African Journal of Pharmacy and Pharmacology*, 5(12): 1475-1483.
68. Anderson, J.W., 1995. Cholesterol-lowering Effects of Soluble Fiber in Human. In "Dietary Fiber in Health Disease" (Kritchevsky D. and Bonfield C. eds), Eagan Press, St. Paul, MN, pp: 126- 145.
69. Huang, Z., M.J. Fasco and L.S. Kaminsky, 1997. Inhibition of estrone sulfatase in human liver microsomes by quercetin and other flavonoids. *J. Steroid Biochem Mal. Bio.*, 63: 9- 15.
70. Asaolu, M., S.S. Asaolu, A.O. Oyeyemi and B.T. Aluko, 2010. Hypolipidemic effects of methanolic extract of *Persea americana* seed in hypercholesterolemic rats. *Journal of Medicine and Medical Sciences*, 1(4): 126-128.
71. Maria, E.P., O. Alicia, C. German, D.H. Maria, G. Leticia, N. Hugo and H. Marcela, 2012. Hypolipdemic effect of avocado (*Persea americana* Mill.) seed in a hypercholesterolemic mouse model. *Plant Foods Hum. Nutr.*, 67: 10-16.
72. Makni, M., H. Fetoui, N. Gargouri, H. Japer, T. Boudawara and N. Zeghal, 2008. Hypolipidemic and hepatoprotective effects of flaxseed and pumpkin seed mixture in  $\omega$ -3 and  $\omega$ -6 fatty acids in hypercholesterolemic rats. *Food Chem. Toxicol.*, 46: 3714-3720.
73. Mohamed, Y.M. and A.R. Amr, 2013. Hepatoprotective effect of avocado fruits against carbon tetrachloride-induced liver damage in male rats. *World Applied Sciences Journal*, 21(10): 1445-1452.
74. Igarashi, K. and M. Ohmuma, 1995. Effect of isorhamnetin, rhamnetin and quercetin on the concentration of cholesterol and lipoperoxide in the serum and liver and on the blood and liver antioxidative enzyme activities of rats. *Biosci. Biotech Biochem.* 59: 595-598.
75. Kang, S.M., J.Y. Shim, S.I. Hwang, S.G. Hong, H.E. Jang and M.H. Park, 2003. Effects of saengshik supplementation on health improvement in diet-induced hypercholesterolemic rats. *J. Korean Soc. Food Sci. Nutr.*, 32: 906- 912.
76. Imafidon, K.E. and F.C. Amaechina, 2010. Effects of aqueous seed extract of *Persea americana* Mill. (Avocado) on blood pressure and lipid profile in hypertensive rats. *Adv. Biol. Res.*, 4(2): 116-121.
77. Sadek, A.M., I.A. Mohammed, A.K. Fatma, A.A. Lamiaa, N.H.A. Barakat and S.M.S. Basma, 2012. Impact of *Actinidia deliciosa* (Kiwi fruit) consumption on oxidative stress status in carcinogenesis. *African J. Biol. Sci.*, 8(1): 117-127.