EFFECTS OF *GARCINIA CAMBOGIA* PLANT EXTRACTS ON SOME BIOCHEMICAL PARAMETERS OF EXPERIMENTAL RATS BLOOD

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Abstract

Garcinia plant is extensively used in herbal medicine and as food in the tropical rain forest region. The present study determined chemical composition of dried *Garcinia cambogia* fruits, hydroxyl citric acid. The antioxidant activity of their hot and cold (10%) *Garcinia cambogia* extracts was determined by using 1, 1-diphenyl-2-picrylhydrazyl (DPPH). The study also, investigated the effects of *Garcinia cambogia* extract (10%) hot and cold on some blood parameters of experimental rats to show their effects on health. The biological experiments consisted of 18 male rats, divided into 3 groups (control, hot and cold 10% *Garcinia cambogia* extracts). The effects of extracts on liver and kidney functions as well as lipid profile of blood were estimated. *Garcinia cambogia* (10%) extracts (2ml w/v) were given twice daily by using stomach tube. Chemical composition of dried *Garcinia cambogia* showed that fruits contained 19.8 % of fat and 66% carbohydrates while possessed 0.22 % protein. Meanwhile it contained some minerals such as calcium (180 mg/100g) iron (150 mg/100g), magnesium (80 mg/100g) as well as few amounts of zinc (2 mg/100g) and copper (2mg/100g). Hydroxycitric acid (HCA) amount was 8%, while showed high antioxidant activity in cold extract than that found in hot. The effect of G*arcinia cambogia* extracts on blood parameters of rats showed a significant decreament (at $P<0.05$) in total lipid, triglycerides, cholesterol, LDL-C, vLDL-C, glucose, albumin, creatinine, urea, ALT and AST activity, while HDL increased compared with control. Total protein and uric acid, showed no significant difference in all groups. It could be concluded that *Garcinia cambogia* plants has a high antioxidant property and has positive effects on different items of investigated biochemical blood tests. The optained natural extracts possess some benefits effect on health and can be used as a natural preservative substance in food and beverages.

Key words: biochemical analysis, antioxidant, *Garcinia cambogia* and hydroxycitric acid.

INTRODUCTION

Garcinia plant is extensively used in herbal medicine and as food, usually found in tropical rain forest region. It prevails as multipurpose tree crops in the home gardens of southern Nigeria. With the shifting of attention from synthetic drugs to natural plant products, the use of plant extracts for disease treatment is now on the increase. Plants that were once developed into drugs had little or no side effects.

(Nzegbule and Mbakwe, 2001). Some studies have indicated that phenolic substances such as flavonoids and phenolic acids are considerably more potent antioxidants than vitamins C and E. The bioactive components can effectively inhibit LDL oxidation and may prevent atherosclerosis by reducing and slowing down the progression to the advance stage (Hodzic *et al*., 2009). Recently, hydroxyl citric acid has been found to be used as a potent metabolic regulator of obesity and lipid abnormalities in mammalian system. *Garcinia* is loaded with B-complex vitamins, and minerals like potassium, manganese and magnesium, that help in controlling heart rate and blood pressure, offering protection against stroke and coronary heart diseases. This *Garcinia* fruit has been used to counter digestive problems such as indigestion, flatulence, acidity and constipation. The fruit possess useful antioxidant, chelating, anti-cancer, anti-fungal, anti-inflammatory, antibacterial, cardio protective and anti-ulcer activities (Manikanta, 2004). *Garcinia atroviridis* or commonly named as 'Asam Gelugur' among locals is extensively used as flavoring agent to provide sour sensation. Apart from being used as flavoring agent, *G. atroviridis* is also used in many ways to traditionally promote health (Hamidon *et al.,* 2017).

The present study aimed to evaluate the chemical composition of dried *Garcinia cambogia*, determined the amounts of hydroxycitric acid and antioxidant activity. The study will estimate the effects of 10% hot and cold *Garcinia* extracts on lipid profile, liver and kidney functions of experimental rats to can be used safely as a natural healthy substance in food.

MATERIALS AND METHODS

Materials:

Garcinia combogia dried fruits were purchased from the folk medicine market in Cairo. Starch and sun flower oil were purchased from local market in Cairo. Casein, salt and vitamin mixtures were obtained from El- Gomhoria Campany, Cairo, Egypt. Hydroxycitric acid (HCA) standard was purchased from Sigma Chemical Company.

Methods of *Garcinia Cambogia* extract preparation:

The whole dried *Garcinia cambogia* fruits were cleaned and milled by Moulinex caba (type 843, code 243, 220 vac 50 Hz 750W) at 3000 RCF for 4 minutes at room temperature to obtain a homogenate powder.

Cold extract:

Exactly 20 gm of *Garcinia cambogia* powder were added to 200 ml of distilled water in conical flask to prepare 10% concentration (w/v) cold extract, and left for 24 hours. The extract was filtered and kept in washed and sterilised glass bottle at refrigerator at (5±1C°) until used.

Hot extract:

Exactly 20 gm of *Garcinia cambogia* powder in conical flask were added to 200 ml of boiled water to prepare 10% ratio (w/v) hot extract. The flask contains extract was covered and left to cool. The prepared extract was filtered and kept in washed and sterilised glass bottles and kept at a refrigerator at (5±1C°) until used (Abd El Hafez*,* 2012).

Analytical methods:

Protein, ash, fat, fibre and mineral contents were determined according to the method of AOAC, (2005) while total carbohydrates were estimated by difference. Hydroxy citric acid (HCA) in *Garcinia combogia was* determined by using HPLC apparatus according to Wodecki, *et al.,* (1991) method. HCA was extracted by using phosphoric acid (0.1% v/v) from *Garcinia* dried fruits. The clear filtrate was injected into Helwellet Packared 1050 HPLC fitted with C18 column (250X 4.6 mm). Ultraviolet (UV) detector set and quarter HP (series 1050). The column temperature was 55C° throughout the analysis. Retention time of standard HCA was used to characterize HCA of *Garcinia*. A calibration curve of HCA was used to quantify the level of HCA in the fruit sample by the data analysis of HPLC apparatus. Antioxidant activity of sample extracts was studied through the evaluation of the free radical-scavenging effect on the 1, 1-diphenyl-2-picrylhydrazyl (DPPH) radical. The results were expressed as percentage of inhibition of the DPPH radical. Percentage of inhibition of the DPPH radical was calculated according to Alothman *et al.,* (2009).

Biological Study:

The experiment was performed in the animal house of Research Institute of Ophthalmology, Giza, Egypt. The experiment was operated by using 18 normal Sprague Dawley male rats (weighted 160 – 170 gm) to study the safety effects of *Garcinia cambogia* extracts (10% cold and hot extracts) on some blood parameters such as lipid profile, liver function and kidney functions. The rats were adapted by feeding on basal diet for one week before the initiation of the experiment Zamora *et al*., (1991). The experimental rats were divided into 3 groups; each group contained 6 rats**.** Group 1 (control) was fed on basal diet for along all the experiment period (6 weeks). While the other two groups (group 2 and 3) fed on basal diet and 2 ml w/v of *Garcinia cambogia* (10 % cold and hot extracts) were taken twice daily by using stomach tube.

Blood Samples Preparation:

At the end of experiment, blood samples were withdrawn and biochemical blood analysis of rats were studied. The blood samples of the experiment rats were

taken from orbital plexus venous by using fine capillary glass tubes. Blood samples were allowed to clot for 10 min at 37 ℃ and centrifuged at 3000 RCF for 10 minutes, and then the separated serum was frozen and kept at -18 ℃ until assayed.

Biological Assays

Aspartate aminotransferase (AST) and Alanine aminotransferase (ALT) activities were colorimetric estimated according to Reitman and Frankel (1957). Total cholesterol, triglycerides and glucose were determined by the colorimetric method according to Trinder (1969). HDL was determined according to Assman (1979). Total lipid, LDL and vLDL were determined according to Fruchart (1982). Total protein and albumin concentrations were determined by the method of Doumas *et al.,* (1971). Creatinine, urea and uric acid were determined according to Thomas, (1998) methods.

Statistical analysis:

Collected data were subjected to the analysis of variance by using Duncan multiple range test procedure at $P<0.05$ as described in the SAS soft ware (SAS, 2002).

RESULTS AND DISCUSSION

Chemical composition of *Garcinia cambogia*:

Chemical composition of dried *Garcinia cambogia* fruits is presented in Table (1). The data showed that *Garcinia cambogia* contained 19.8 % crude fat (ether extract), 0.22 % protein, 3.1 % ash 10.88 % crude fibre and 66% carbohydrates, on dry basis. Meanwhile Table (2) showed that it was contained a reasonable amount of minerals such as calcium (180mg/100g) and iron (150 mg/100g) as well as few amounts of zinc (2.0 mg/100g), copper (2.0 mg/100g) and magnesium (80 mg/100g). These data are in accordance with those found by Abd El Hafez, (2012) who mentioned that *Garcinia cambogia* is characterized by low protein contents, high carbohydrates and moderate amounts of crude fibres and ash also, it contains considerable amounts of minerals.

Table 1. Chemical composition of *Garcinia cambogia* dried fruits (g/100g) on dry basis:

Raw material	Crud Fat %	Protein %	Crude fibre %	Ash %	Total Carbohydrates %
Garcinia	19.8	0.22	10.88	3.1	66

*Total carbohydrate was estimated by difference.

Table 2. Mineral contents in raw *Garcinia cambogia* (mg/100g) on dry basis:

Raw material	Calcium (Ca)	Iron (Fe)	Zinc (Zn)	Copper (Cu)	Magnesium (Mg)
Garcinia	180	150	2.0	2.0	80

The data in Table (3) showed that the hydroxycitric acid (HCA) contents and DPPH activity of cold and hot *Garcinia cambogia* extracts. The hydroxycitric acid (HCA) was recorded 8%. Manikanta, (2004) mentioned that hydroxycitric acid (HCA) is a principle constituent (10–30%) of the dried fruit rind of *Garcinia cambogia*. HCA has undergone considerable formal safety, without the evidence of toxicity appearing.

The decreasing of HCA under study may be return to storage of plants at folk medicine.

The same table data showed high antioxidant activity of 10% *Garcinia cambogia* cold extract (69.8% of inhibition of DPPH radical) than that found in the 10% hot extract (36.36 % of inhibition of DPPH radical). The polyphenol contents of extracts were quantified by Folin–Ciocalteau's reagent assay and expressed as gallic acid equivalents (µg GAE/g). Ritthiwigrom *et al.,* (2013) mentioned that higher antioxidant activity (DPPH) of some fruits exhibited perhaps are due to the linear correlation between antioxidant activity and total soluble phenols which could be useful to consumers to plan antioxidant rich diets and their impact in health and disease.

Table 3. Hydroxycitric acid (HCA) contents and (DPPH) activity in *Garcinia cambogia* on dry basis:

Raw material	hydroxycitric acid (HCA)*	DPPH activity % of cold *Garcinia* extract	DPPH activty % of hot *Garcinia* extract
Garcinia cambogia	8	69.8	36.36

*HCA: g/100g

Total lipids, triglycerides, total cholesterol, HDL-c, LDL-c and vLDL-c (mg/dl) in serum rats fed on (10%cold and hot) *Garcinia cambogia* extracts:

The effect of *Garcinia cambogia* (10% cold and hot) extracts on total lipids, triglycerides, total cholesterol, HDL-c, LDL-c and vLDL-c in serum rats were recorded in Table (4). The data showed that a slightly significantly decreasing was detected in total lipid, triglycerides and vLDL-c at ($P<0.05$) compared with control. Total cholesterol and LDL-c, showed a significantly difference decreasing in groups fed on *Garcinia cambogia* extracts (10% cold and hot). While HDL-c showed a significantly difference increasing in groups fed on 10% cold and hot *Garcinia cambogia* extracts comparing with control. Al-Mansoub *et al.,* (2013) mentioned that *Garcinia* fruit juice affected on body weight and cholesterol reduction ability in rats. The treatment was successfully reduced the LDL-c cholesterol level. Darji *et al*., (2010) illustrated that the methanolic extract of the dried fruit of *Garcinia* type kokum showed remarkable antihyperlipidemic activity in rats, using cholesterol induced hyperlipidemic model

diets. A significant decrease in total cholesterol, triglycerides, LDL-c, VLDL-c levels and increase in HDL-c was reported.

Table 4. Total lipids, triglycerides, total cholesterol, HDL-C, LDL-C and vLDL -c in serum rats fed on 10% (cold and hot) *Garcinia Cambogia* extracts:

Rat Groups	Total lipid*	Triglycerides*	Total cholesterol*	HDL-c*	LDL-c*	v LDL-c*
Control	270±1.15 a	129.52±0.19 a	120.27±0.07 a	49.99±0.01 c	44.81±0.22 a	25.90±0.01 a
10% cold extract	266±1.15 b	127.62±0.13 b	119.22±0.13 b	52.80±0.06 b	40.92±0.17 b	25.50±0.09 b
10% hot extract	268±0.58 ab	129.45±0.25 a	119.50±0.0.06 b	53.50±0.25 a	40.40±0.16 ab	25.80±0.11 b

Each mean value, within the same column, followed by the same letter is not significantly different at 0.05 level. Each value, mean of three replicates, is followed by ± standard deviation. *mg/dl

Albumin, total protein, glucose, ALT and AST activity in serum rats fed on 10% (cold and hot) *Garcinia cambogia* extracts:

Total protein (g/dl), albumin (g/dl), glucose (mg/dl), ALT (U/L) and AST (U/L) activity in serum rats fed on cold and hot *Garcinia cambogia* extracts was recorded in Table (5). Total protein showed that no a significant difference among groups fed on *Garcinia cambogia* extracts (10% cold and hot) and control. The results recorded a slight significant difference decreased in albumin ($P \leq 0.05$) of rats fed on 10% *Garcinia cambogia* cold extracts comparing with hot extract and control. Glucose and ALT activity were showed that a significant difference decreasing among groups fed on *Garcinia cambogia* extracts (10% cold and hot) and control meanwhile, there was no significant difference either hot or cold groups. AST activity also, was showed a significantly difference decreased in groups fed on *Garcinia cambogia* extracts (10% cold and hot) relative control.

From data it was found that *Garcinia cambogia* extracts improved liver enzymes (AST and ALT activity) and glucose while was no effects in total protein. Gogoi *et al.,* (2012) who reported that aqueous and ethanolic extract of *Garcinia indica Linn* fruit has showed a hepatoprotective effect on liver which lower toxicity on wistar albino rats and the degree of liver protection increased. The presented data revealed that biochemical analysis of blood parameters showed decreasing in lipid profile, improving liver functions meanwhile increasing in HDL-c.

Table 5. Total protein, albumin, glucose, ALTand AST activity in serum rats fed on (10% cold and hot) *Garcinia cambogia* extracts:

Rat groups	Total protein*	Albumin*	Glucose***	ALT**	AST**
Control	5.17±0.15 a	3.77±0.09 a	84.22±0.13 a	38.12±0.07 a	48.22±0.13 a
10% cold E.	5.03±0.19 a	3.23±0.09 b	82.33±0.06 b	35.65±0.13 b	44.52±0.19 c
10% hot E.	5.23±0.20 a	3.40±0.17 ab	82.31±0.07 b	35.99±0.06 b	45.11±0.06 b

Each mean value, within the same column, followed by the same letter is not significantly different at 0.05 level. Each value, mean of three replicates, is followed by ± standard deviation.

* g/dl ** U/L ***mg/dl

Urea, createnine and uric acid (mg/dl) in serum rats fed on cold and hot *Garcinia cambogia* extracts was recorded in Table (6). The results showed a slightly significant difference decreased in urea and creatinine of rats fed on 10% *Garcinia cambogia* extracts (10% cold and hot) relative control. Uric acid was showed no a significant difference in all groups comparing to control. The data revealed that *Garcinia cambogi* extracts 10% cold more effective on Urea and createnine than hot while was no effects on uric acid.

Table 6. Urea, creatnien and uric acid levels in serum rats fed on (10% cold and hot) *Garcinia cambogia* extracts:

Rat groups	Urea *	Creatinine*	Uric acid*
Control	25.11±0.07 [a]	1.50±0.10 [a]	1.70±0.15 [a]
10% cold E.	23.11±0.06 [c]	1.02±0.01 [b]	1.37±0.12 [a]
10% hot E.	23.73±0.20 [b]	1.37±0.12 [a]	1.47±0.15 [a]

Each mean value, within the same column, followed by the same letter is not significantly different at 0.05 level. Each value, mean of three replicates, is followed by ± standard deviation. * mg/dl

CONCLUSION

From the above studies, it could be concluded that *Garcinia cambogia* natural plant extract was a good natural antioxidant which acts as a protective agent against free radical substances in the body. It has a protective agent on some health parameters. These potential beneficial effects enable to be used as an additive substance for food quality and health.

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تأثير مستخلص نبات الجارسنيا كمبوجيا على بعض المواد البيوكيميائية ومكونات الدم لفئران التجارب

سامية على العسقلانى

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يستخدم نبات الجارسينا فى طب الاعشاب والطعام فى المناطق الاستوائية الممطره أشتملت هذه الدراسة على تقديرالتركيب الكيماوى لنبات الجارسنيا كمبوجيا الخام وقياس نسبة حامض الهيدروكسى سترك والنشاط التأكسدى بأستخدام (DPPH) للمستخلص المائى الساخن والبارد (١٠%) واشتملت ايضا الدراسة على التاثيرات البيولوجية على بعض مكونات الدم لذكور الفئران بأستخدام تركيز ١٠% للمستخلص المائى الساخن والبارد للنبات لدراسة التأثير الأمن للمستخلص على الصحة.

وقد أجريت التجربة البيولوجية بأستخدام ثلاث مجموعات (الأولى) كنترول و(الثانية) المستخلص المائى للجارسنيا البارد (١٠%) و(الثالثة) المستخلص المائى للجارسنيا الساخن (١٠%) لدراسة تأثير مستخلص الجارسنياعلى الدهون ووظائف الكبد والكلى فى الدم.

وأظهرت نتائج التحاليل الكيميائية لنبات الجارسنيا الجاف أنها تحتوى على (١٩,٨%) من الدهن و(٦٦%) من الكربوهيدرات و(٠,٢٢ %) البروتين ومن جهة أخرى تحتوى على كمية من المعادن يمكن أخذها مثل الكالسيوم (١٨٠ ملى جرام/١٠٠جرام) والحديد (١٥٠ ملى جرام/١٠٠جرام) وكمية قليلة من الزنك (٢ ملى جرام/١٠٠جرام) والنحاس (٢ ملى جرام/١٠٠جرام) والماغنسيوم (٨٠ ملى جرام/١٠٠جرام) وتحتوى على ٨ % من حامض الهيدروكسى سيترك الذى له نشاط تأكسدى عالى خاصة للمستخلص البارد، ومن دراسة التحليلات البيوكيميائية للدم لفئران لوحظ وجود نقص فى دهون الدم والجلسريدات الثلاثية والكولسترول والكولسترول المنخفض الكثافة والمتناهى الصغر فى الكثافة الضار(LDL-C و vLDL-C) و الالبيومين والكرياتتين واليوريا وكذلك أنخفاض نشاط أنزيمات الكبد (ALT) و(AST) مقارنة بالكنترول بينما زادت نسبة الكولسترول عالى الكثافة (HDL-C) المفيد ولم يتأثر البروتين وحامض البوليك.

ويمكن أن نستبط من الدراسة أن نبات الجارسنيا يحتوى على مواد مضادة للاكسدة بنسبة عالية والتى لها تأثير أيجابى على تحليلات الدم المختلفة وأن هذا المستخلص الطبيعى للجارسنيا له فوائد صحية تمكننا من أستخدامه كمادة حافظة طبيعية فى الطعام و المشروبات.