American Scientific Research Journal for Engineering, Technology, and Sciences (ASRJETS)

ISSN (Print) 2313-4410, ISSN (Online) 2313-4402

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ttp://asrjetsjournal.org

Decreased of Levels of Triglyceride in Subjects Drinking Garcinia atroviridis Leaf Tea from Sijunjung - West Sumatra, Indonesia

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Abstract

The objective of this study was to evaluate the effect of *Garcinia atroviridis* on triglyceride levels in obese subjects. Obesity and related non-communicable diseases are largely preventable. The household industry in Lubuk Tarok District of Sijunjung West Sumatra produces tea of *G.atroviridis* leaves packed in tea bags and people drink GA-tea to reduce triglyceride levels and waist circumference. Fruit rind of GA or asam glugur contains hydroxycitric acid (HCA, or (-)-HCA) as principal acid. HAC is a potent inhibitor of ATP-citrate lyase. The inhibition of this enzyme limits the availability of acetyl-CoA units required for fatty acid synthesis and lipogenesis. Participants in this study were obese male and female adults, who were divided into *G.atroviridis* tea group and control group. Anthropometric measurement and laboratory test were performed in pre- and post-treatment. The result showed that the reduction of triglyceride levels was found to be higher in tea group than control groups.

Keywords: Hydroxycitric acid; triglyceride; obesity; Garcinia atroviridis.

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1. Introduction

Heretofore, the research has still been ongoing to find useful food and supplements to prevent and cope with obesity and lipid profile disorders. Obesity is defined as abnormal or excessive fat accumulation that may impair health. Most of the world's population resides in countries where overweight. The fundamental cause of obesity and overweight is an energy imbalance between calories consumed and calories expended. Raised BMI is a major risk factor for non-communicable diseases (NCDs) such as; cardiovascular diseases, diabetes, musculoskeletal disorders, and some cancers. The risk for these NCDs increases in body mass index (BMI). Overweight and obesity, and the related NCDs, are largely preventable. Healthful foods and regular physical activity are the easiest choice to prevent overweight and obesity [1]. Body mass index (BMI) is a simple index of weight-for-height that is commonly used to classify obesity in adults; it is defined as a person's weight in kilograms divided by the square of body height in meters (kg/m²) [1]. An individual adult can limit energy intake and engage in regular physical activity, while local food industry ensuring healthy choices available for community[1]. In western Sumatra, Garcinia atroviridis leaves are dried, graded, and packed in tea bag or sachets and taken for the purpose of weight loss and reduce body fat. Garcinia atroviridis, known as asam gelugur, grows to the height of 20 m, with shiny dark green leaves [2]. The ripe fruits of G.atroviridis used by Indonesian natives as a flavoring ingredients in Indonesian cuisine, and as acidity to cooked dishes[2]. The fruit of G.atroviridis contains fruiting acids such as citric acid, tartaric acid, malic acid, and ascorbic acid. The principal acid of the fruit rinds is hydroxycitric acid (HCA, or (-)-HCA)[2] which is also found in the fruits and in the rind of certain garcinia fruit [3]. Hydroxycitric acid was shown to be a potent inhibitor of ATP citrate lyase which is one of the three citrate enzymes which catalyzes the same bond-making and -breaking reaction which involves the equilibrium of citrate with oxalacetate and an acetyl moiety [4]. ATP citrate lyase, a cytosolic enzyme, cleaves citrate so that acetyl CoA may be used for fatty acid and other biosyntheses. In the presence of ATP and CoenzymeA, ATP Citrate (pro-3S)-lyase catalyzes the cleavage of citrate to yield acetyl CoA, oxaloacetate, ADP, and orthophosphate. This reaction represents an important step in FA biosynthesis [4]. The inhibition of this reaction limits the availability of acetyl-CoA units required for fatty acid (FA) synthesis and lipogenesis during a lipogenic diet, that is, a diet high in carbohydrate. Animal studies indicated that HCA suppresses the FA synthesis, lypogenesis, food intake, and induced weight loss. Vitro studies revealed the inhibition of FA synthesis and lipogenesis from various precursors [3].

2. Materials and Methods

This study was designed as a case-control study to compare the TG levels lowering effect of *G.aroviridis* leaf tea in Tea group and Control group. It recruited 30 obese adults as the participants in Medan, from January 2016 until December 2016. The criteria for participants recruited in this study were male and female adults with obesity while the diagnostic criteria of obesity and waist circumference used were according to the classification of BMI categories for Asia. Subjects with obesity were classified with BMI \geq 25 kg/m², with waist circumference (WC) \geq 80 cm for women and \geq 90 cm for men [5,6]. Selected thirty participants (15 men, 15 women) were divided into two groups by randomization; control group (consisted of 15 subjects) and tea group (15 subjects). Randomization was performed by random number generation, and group assignment was placed in a sealed envelope.

All anthropometric measurements were done with standard techniques: body weight was measured with digital scales with arms on side, legs straight, knees together, and without heavy clothing; body height was measured barefoot by stadiometer within 0.5 cm ; circumferences within 1 mm with stretch resistant plastic tapes, with waist mid-way between the lowest rib and the iliac crest with the subject standing at the end of gentle expiration [7, 8]. The exclusion criteria were the history of heart disease, diabetes, the use of lipid-lowering therapy, drugs affecting insulin resistance, oral hypoglycemic pills or insulin, and pregnancy. All participants gave written informed consent.

One sachet of *G.atroviridis* leaf tea was brewed into 200 mL of boiled water (100°C) and allowed until it decreased in temperature; this tea could be drunk immediately or any time all day long by the participants in tea group. All of participants attended walking activity three times a week, 45 minutes each time, for 4 weeks. Walk duration was measured by a stop-watch, and the distance was also measured in each activity. Subjects were educated about healthful food menu, and they were allowed to consume daily menu that had been adjusted to the education given.

The pre-treatment physical examination performed were measurement for height (measured in cm), weight (in kg), and waist circumference (cm). The laboratory test was performed after an overnight of 12 hours, blood samples were drawn for measurements of total cholesterol (TC, mg/dL), triglyceride (TG, mg/dL) and fasting blood glucose (FBG, mg/dL). The post-treatment measurement was performed after completing a 4-week treatment, Laboratory test was done in fasting blood specimen.

Fasting was referred to 12 hours or overnight complete dietary restriction with the exception of water [9]. Fresh blood samples were analyzed to measure plasma levels of fasting plasma glucose, total cholesterol, and triglycerides.

For the measurement of fasting blood glucose level, the preparation and procedure were similar to that described elsewhere [10, 11]. For the measurement of lipid profile, the procedure was similar to that described by Syukur et. al. (2017) [12].

The primary endpoint of this study was to compare the lowering effect of *G.atroviridis* leaf tea on triglyceride levels of obese subjects after 4 weeks of tea consumption against control. The secondary endpoint was body indices (body weight and body height to measure BMI), waist circumferences (WC), and laboratory values. The study was approved by Ethical Committee of Faculty of Medicine University of Andalas, Padang - Sumatra Barat, and conducted according to the Declaration of Helsinki.

For statistical analysis, characteristic data not normally distributed were given as medians (with min-max), and data normally distributed were given as mean \pm SD. Mann-Whitney U test was employed in data analyses.

3. Results

The characteristics of the subjects were shown in Table 1. Fifteen males and fifteen females were recruited in this study and separated into control (15 subjects) and tea group (15 subjects). All subjects were categorized as

obese according to their body mass indices and waist circumferential measurements (Table 2).

An example of a column heading	Control Group	Tea Group (<i>T</i>)
Ν	15	15
Male	10	4
Female	5	11
Age (y)	47.7 (30.9 - 59.1)	50.3 (31.4-58.3)

Table 1: Baseline characteristic of anthropometric indices of subjects

After the treatment period was completed, anthropometric measurements and biochemistry levels compared with baseline measurement (Table 2).

Table 2: Anthropometric and biochemistry indices at baseline and after 4 weeks treatment

	Baseline			After 4 weeks		
	Control Group	Tea Group	р	Control Group	Tea Group	р
BMI (kg/m ²)	32.60 (4.64)	31.32(3.49)	ns	31.82 (4.80)	30.85 (3.46)	ns
WC (cm)	104.00 (7.23)	104.27(7.40)	ns	102.07(7.43)	102.87 (7.02)	ns
FBG (mg/dL)	103.00 (78-171)	116.00(84-581)	ns	85.00(73-139)	89.00 (79-268)	ns
TC (mg/dL)	209.40 (38.78)	205.27(19.37)	ns	201.93(45.65)	193.87(24.03)	ns
TG (mg/dL)	179.00 (79-550)	175.00(74-843)	ns	136.00 (74-331)	119.00(66-367)	ns

At baseline, all subjects were obese by BMI criteria. Central obesity with measured waist circumference were 104.00 (\pm 7.23) cm and 104.27 (\pm 7.40) cm in control and *G.atroviridis* tea groups, respectively. Meanwhile, FBG and TG levels were 103.00 (78-171) mg/dL, and 179.00 (79 - 550) mg/dL for control group, and 116.00(84-581) mg/dL and 175.00 (74 - 843) for tea group. The measurements between the two groups were not statistically different. After four weeks, all anthropometric and biochemical measurements demonstrated a decrease in control group and in tea group. Fasting blood glucose decreased to 85.00 (73-139) mg/dL and 89.00 (79-268) mg/dL, and triglyceride level declined to 136.00 (74-331) mg/dL and 119.00 (66-367) mg/dL for control and tea groups, consecutively.

Statistic analysis using Mann-Whitney U, resulted with p value 0.72 (>0.05) (two tailed) (Table 3), showed that the changes of TG levels between the two groups were not statistically different; hence the null hypothesis was retained. The median of decreased change of the TG levels was presented as median (min-max); 30.00 (-96 - 219) in control group, and 31.00 (-147 - 617) in tea group, with an average tendency to decrease TG levels were greater in tea group than in control group. The mean difference in total length of walking track (12 walking activities in four weeks) between groups were not different significantly (mean difference 0.67, p 0.75, 95% CI

(-3.67 - 5.06), and also, the mean of total walking track did not differ in the two groups (mean difference 0.04, p 0.75, and 95%CI (-0.23 - 0.31).

	Mean \pm SD	Median (Min-Max)	Ranks of Change
Control Group (n=15)) 37.13 ± 71.88	30.00 (-96 - 210)	14.93
Tea Group (n=15)	75.40 ± 176.09	31.00 (-147 - 167)	16.07

Table 1: Changes of triglyceride levels in the two groups after 4 weeks

Fourteen percent of obesity was associated with high or very high triglyceride, and one of associated factors with chronic NCD was triglyceride levels and obesity [10]. All subjects involved in this study were central obese, with waist circumferences greater than 80 cm in females, and over 90 cm in males. Non-communicable disease is known to be associated with obesity, high total cholesterol, and triglycerides [13]. According to a consensus statement all subjects in both groups showed borderline - high levels of triglyceride at baseline [14]. Fasting glucose levels of both groups were in the criteria of fasting glucose levels of the metabolic syndrome and/or as pre-diabetes (impaired fasting glucose), at baseline[15]. It is well known that among subjects with metabolic syndrome, waist circumference and triglyceride levels are independently associated with intra-abdominal fat and subcutaneous fat areas. Waist circumference and TG are suggested to be the best factors of insulin resistance and visceral adiposity [16].

Regular physical activity is one of the easiest choices to prevent obesity and regular walking is one of the easiest physical activities. Previous study reported that physical activity like endurance exercise training was beneficial for health [16], while other study reported decreased plasma free fatty acid turnover and oxidation during submaximal exercise [17]. In this study, all subjects in both groups were involved in regular walking as exercise for 45 minutes, three times a week for 4 weeks, and then all indicators - BMI, WC, FBG levels, total cholesterol, and triglyceride levels were examined.

Researcher had been conducted studies on local plant-originating foods beneficial to improve lipid profiles, and there were local food product of Smtrb demonstrated to be beneficial in lowering triglyceride levels and increase HDL levels [12], while this study used tea from dried leaves of *G.atroviridis* produced in west Sumatra to prove its efficacy to decrease triglyceride levels among obese subjects. Hydroxycitric acid is a derivative of citric acid. The principal acid of *G.atroviridis* is hydroxycitric acid [2] which is found in the fruits and rind [3].

This study conducted for 4 weeks in male and female subjects, different from a study conducted in 8 weeks, using water soluble calcium HCA as *G.atroviridis* in obese female subjects. Dissolved *G.atroviridis* was taken before meals, three times daily. Followed with significant loss of body weight compared with placebo as a result of the reduction of fat storage [18]. Although the subjects in the tea group were using tea for only 4 weeks, the result showed the decrease in BMI and WC at the end of the study.

Not only hydroxycitric acid, G.atroviridis leaves and fruit extracts have potential use as a source of natural

antioxidants and nutrients for therapeutic purposes[19]. In vivo, hydroxycitric acid is a highly effective inhibitor of FA synthesis by rat liver. ATP-citrate lyase is a lipogenic enzyme that catalyzed the critical reaction linking cellular glucose catabolism and lipogenesis, converting cytosolic citrate to acetyl-coenzyme A (CoA). Acetyl-CoA is further converted to malonyl-CoA, the essential precursor for FA biosynthesis. Liver-specific ACL abrogation inhibits the expression of PPAR γ and lipogenic program in the liver. Hepatic ACL deficiency resulted in enhanced insulin sensitivity in the muscle, leading to improved systemic glucose metabolism. Hence, targeted suppression of ACL in the liver is also to improve in whole-body glucose metabolism[20]. Beside ACL, hydroxycitric acid also inhibits the activities of isocitrate dehydrogenase, malate dehydrogenase and aconitate hydratase [21].

This study used one tea bag of *G.atroviridis* dried leaves brewed in 200 mL of water that should be taken at any time of the day, for four weeks. The concentration of hydroxycitric acid in this tea and its bioavailability were not investigated yet. The results showed that obese subjects in tea group and control group underwent a decrease in TG levels together with insignificant reduction of BMI after a 4 week-consumption of *G.atroviridis* tea, compared with control. The decrease in TG levels in T-group was slightly greater than that in C-group, and the result of Mann-Whitney U test showed that p-value = 0.72 (p > 0.05). This reduced BMI and TG levels, followed by the reduction of WC. This result was supported with the study which demonstrated that *G.cambogia* with its HCA reduced abdominal fat accumulation, subcutaneous fat, and total fat areas significantly[22].

Several studies have shown controversial results of the effects of *G.atroviridis* extraction on trained subjects. A large amount of HCA resulted with the increase in high plasma HCA concentration during the exercise. Ingestion of one dose of HCA before exercise resulted in an increase in plasma HCA concentration during rest, and a dose of HCA during exercise was followed with further increase in plasma HCA concentration without significant differences in total fat and carbohydrate oxidation rates. Increased and large dose of HCA did not increase total fat oxidation in vivo in trained humans [23]. Different from previous study, the subjects in our study were supplemented with one tea bag of *G.atroviridis* which were taken at any time of the day, the subjects themselves used to be sedentary before recruitment, but results of our study supported by previous study that even in untrained subjects, HCA supplementation resulted with the increase in lipid metabolism[24]. Dose and HCA concentration in tea liquid used in this study were not yet determined, while in previous study 0.5 gr of HCA ingestion for five days increased fat oxidation in untrained man who did not control their daily food consumption during experiment [25]. To get the benefit, the concentration of HCA in tea needs to be determined before consumption by community.

Heretofore, there have been some controversial results of previous research on HCA and *G.atroviridis*. This study was not supported by previous research which concluded that a treatment with HCA did not affect energy expenditure, either during rest or during moderately intense exercise [26]. Another previous report revealed that HCA failed to produce significant weight loss and fat mass loss beyond that observed with placebo [27]. Controversies on dose supplementation, bioavailability, and the proper use and benefits of HCA extract derived from dried leaves of *G.atroviridis* grown in west Sumatra required further research to provide recommendation.

4. Conclusion

Theresult of the use of *G.atroviridis* tea demonstrated that clinical decrease in triglyceride levels was greater in the control group. The decrease in triglyceride levels was accompanied by the decrease in body mass index, waist circumference, and fasting blood glucose levels. In order to obtain significant results, the next researches need to continue to obtain certain HCA concentration in tea for a more appropriate product.

Acknowledgement

I would like to express my appreciation to my supervisor Prof.Endang Purwati, whose supervision and attention was invaluable throughout the course of this study. The authors wish to thank PUSPEN 2016-2017, all members of field staff, PT Prodia Widya Husada and all study participants involved in this study.

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