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The Impact of Green Tea on Blood Fluidity Improvement and Weight Loss

Efek Teh Hijau Terhadap Peningkatan Fluiditas Darah Dan Penurunan Berat Badan

Author :
Dr. Djoko Agus Purwanto, Apt., M.Si. | djokoagus@ff.unair.ac.id
Fakultas Farmasi
Dra. Asri Darmawati, MS. | -
Fakultas Farmasi
Purwaningsih, S.Kp., M.Kes. | purwaningsih1@gmail.com
Fakultas Keperawatan

Abstract

Introduction: Various studies have shown the beneficial effects of green tea, not only on cardiovascular diseases but also on type 2 diabetes. Method: In this study, the preparation of green tea water extract has been standardized to (-)-epigalocatechin gallate (EGCG), the major component of green tea. The role of green tea water extract on blood fluidity and diabetes diseases has been studied in 13 Fructose-Fed Rat (FFR). The rats were given high fructose diet ad libitum for one week and then combination with green tea water extract every day for 6 days. Results: The results show, green tea water extract can reduces 100 µL blood passage times of wistar rat significantly ($p<0.01$) by Micro-Channel Array Flow Analyzer (MC-FAN) instrument. Green tea water extract also had strong effect in reducing abdominal fat ($p<0.05$), blood glucose level ($p<0.01$) and body weight ($p<0.01$). Discussion: These results suggest that green tea water extract may has beneficial effects for the treatment of diabetes and reduce blood viscosity

Keyword : green, tea, epigallocatechin, gallate, HPLC, blood, fluidity, fructose-fed, rat,

Daftar Pustaka :

1. **Beltz LA, Bayer DK, Moss AL, Simet IM, (2006).** Mechanisms of cancer prevention by green tea and black tea polyphenols. - : Anticancer Agent Med Chem
2. **Lin YS, Tsai YJ, Tsay JS, Lin JK, (2003).** Factors affecting the levels of tea polyphenols and caffeine in tea leaves. - : J Agric Food Chem

EFEK TEH HIJAU TERHADAP PENINGKATAN FLUIDITAS DARAH DAN PENURUNAN BERAT BADAN

(The Impact of Green Tea on Blood Fluidity Improvement and Weight Loss)

Djoko Agus Purwanto,* Asri Darmawati*, Purwaningsih**

*Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Airlangga University, Campus B, Jl. Dharmawangsa Dalam, Surabaya 60286, Indonesia
**Department of Fundamental Nursing, Faculty of Nursing, Airlangga University, Surabaya, Indonesia
E-mail: djokoagus@ff.unair.ac.id

ABSTRAK

Pendahuluan: Berbagai penelitian telah menunjukkan efek menguntungkan dari teh hijau, tidak hanya pada penyakit jantung tetapi juga pada diabetes tipe 2. **Metode:** Dalam penelitian ini, penyusunan ekstrak air teh hijau sudah terstandarisasi untuk (-) - epigalocatechin gallate (EGCG), komponen utama teh hijau. Efek ekstrak air teh hijau terhadap fluiditas darah dan diabetes dipelajari dalam *13 Fruktosa - Fed Rat* (FFR). Tikus diberi diet tinggi fruktosa *ad libitum* selama satu minggu dan kemudian kombinasi dengan ekstrak air teh hijau setiap hari selama 6 hari. **Hasil:** Hasil penelitian menunjukkan, air ekstrak teh hijau dapat mengurangi 100 uL darah dari wistar tikus secara signifikan ($p<0,01$) dengan alat *Micro-Channel Array Arus Analyzer (MC - FAN)*. Ekstrak air teh hijau juga memiliki efek yang kuat dalam mengurangi lemak perut ($p< 0,05$), kadar glukosa darah ($p<0,01$) dan berat badan ($p<0,01$). **Diskusi:** Hasil ini menunjukkan bahwa ekstrak air teh hijau memiliki potensi menguntungkan untuk pengobatan diabetes dan mengurangi kekentalan darah.

Kata kunci : teh hijau , *(-) - epigallocatechin gallate*, HPLC, fluiditas darah , fruktosa-fed rat

ABSTRACT

Introduction: *Various studies have shown the beneficial effects of green tea, not only on cardiovascular diseases but also on type 2 diabetes.* ***Method:*** *In this study, the preparation of green tea water extract has been standardized to (-)-epigalocatechin gallate (EGCG), the major component of green tea. The role of green tea water extract on blood fluidity and diabetes diseases has been studied in 13 Fructose-Fed Rat (FFR). The rats were given high fructose diet ad libitum for one week and then combination with green tea water extract every day for 6 days.* ***Results:*** *The results show, green tea water extract can reduces 100 µL blood passage times of wistar rat significantly ($p<0.01$) by Micro-Channel Array Flow Analyzer (MC-FAN) instrument. Green tea water extract also had strong effect in reducing abdominal fat ($p<0.05$), blood glucose level ($p<0.01$) and body weight ($p<0.01$).* ***Discussion:*** *These results suggest that green tea water extract may has beneficial effects for the treatment of diabetes and reduce blood viscosity.*

Keywords: *green tea, (-)-epigallocatechin gallate, HPLC, blood fluidity, fructose-fed rat*

INTRODUCTION

Tea has been consumed by many people in the world since ancient times. In various countries, green tea has been processed into a variety of foods and beverages such as cakes, ice cream and candy. Beside water, tea product also consumed in varous type. At least there are four basic form of tea product: green tea, oolong tea, pu'erh tea and black tea. These forms are different in process of production. Black tea was made from fermentation of tea leaf. The main componen of green tea is (-)-epigalocatechin gallate (EGCG). During production process, oxidation of EGCG and another substance are promoted, so that most of these substances are oxidized. Because of that reason, level of EGCG and total catechin in tea were depend on type of tea. In old leaf of green tea, EGCG contain is higher than young leaf, and higher than oolong tea, and also higher than black tea and pu'erh tea (Lyn, et al, 2003).

There is good evidence from in vitro studies that green tea catechins have an important role in protection against degenerative diseases. EGCG from green tea extract has many activities such as anti-carcinogenic (Beltz et al, 2006, Spinela, et al, 2006), antioxidant (Luximon et al, 2006), anti-microbial activities (Paul, et al., 2006, Watson, et al., 2005) as well as anti-diabetic activity (Kim et al., 2013). Feeding of tea catechins has beneficial effect for the reduction of high fat diet induced obesity by increasing lipid metabolism. EGCG may modulate the distribution of Rstn protein, an adipocyte-specific

secretory hormone that can cause insulin resistance (Liu. et al, 2006). In the present study, the effect of standardized of green tea extract on EGCG as the active compounds, on blood fluidity, body weight and diabetes impact of fructose-fed Wistar rat were investigated. In addition, further investigations of the effect of this extract on some organs were also examined.

MATERIALS AND METHODS

Green tea was obtained from commersial product of PTPN 12, Indonesia. Heparin is included on Venoject tube 5 ml sterile from Kruuse. Fructose, casein, vitamins and minerals were purchase from Wako Pure Chemical Industry.

Male Wistar Rats were obtained from Clea Inc. Japan at 7 weeks of age. The fructose-fed diet mixed in Laboratory of Nutrition Physiology Kobe Women's University. The components of the fructose fed (100 g) were follows: 66% fructose, 22% casein, 12% lard plus essential vitamins and minerals. Samples experiment were given green tea extract 300 mg/kg body weight in water solution The control was made by given the same volume of distilled water. Wistar rat were kept in an animal room maintained at constant temperature (22 $\pm$ 2^{o}C). The rat consumed a fructose fed ad libitum and had free access to distilled water for 2 weeks. Body weight was measured every day. All procedures were performed in accordance with standards related to the care and management of experimental animals (including ethics) of the Kobe Women's University.

Wistar rat (8-10 weeks old) were fasted 12 h before the starting point of experiments. The rats were per orally administered with either a suspension of green tea powder on hot water at 9:20 a.m. Ten minute after the administration (at 9:30 a.m.), the rats were perorally administered with 1.0 ml of glucose solution containing 350 mg glucose. Blood glucose levels were measured before and 30, 60, and 120 min after the administration with glucose solution.

Mix 2.0 ml of fresh blood with 100 μl heparin. Blood passage time was measured for 100 μl of blood through an artificial capillary using MC-FAN (Micro-Channel Array Flow Analyzer). The Micro-Channel array consisting of 8,736 capillaries with 4.5 μm in diameter and 30 μm in length. The time needed was determined and expressed against 100 μl saline solution adjusted to 12 seconds.

For statistical evaluation of the data in rat, repeated measures Student's t test were used. Difference of $p < 0.05$ were considered significant. SPSS 14.0 for Windows software was used for all statistical analysis. Values in the text are means $\pm$ SD.

RESULTS

In the chromatograms of the EGCG standard solution analyzed by HPLC, the peak of standard EGCG was identified as the single peak and separated completely from other compound in sample of green tea extract. Standard EGCG concentration has linier correlation ($r = 0.9994$, $p<0.01$) with HPLC peak area. Concentration of EGCG in sample extract green tea was found 4.53 $\pm$ 0.03 % and calculated from dry weight. This standardized sample was used for further examination in blood fluidity and diabetic impact of fructose fed rat.

Effect of green tea extract on blood parameter, fat and organs can be seen in table 1. Abdominal fat decreases significantly after drinking green tea. The average abdominal fat for the control group being 70% greater than in the animals with green tea. All of the blood parameter did not change significantly between two groups. Organs weight also has no significantly different except pancreas and kidney.

Effect of green tea water extract treatment on glucose metabolism in FFR was observed by oral glucose tolerance test (OGTT) at 2 week. Figure 2 shows that no significant differences of OGTT were observed between control and green tea extract groups.

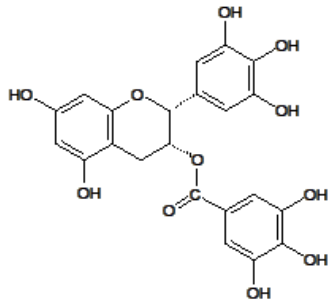

Figure 1. Structure of (-)-epigallocatechin gallate (EGCG) from green tea.

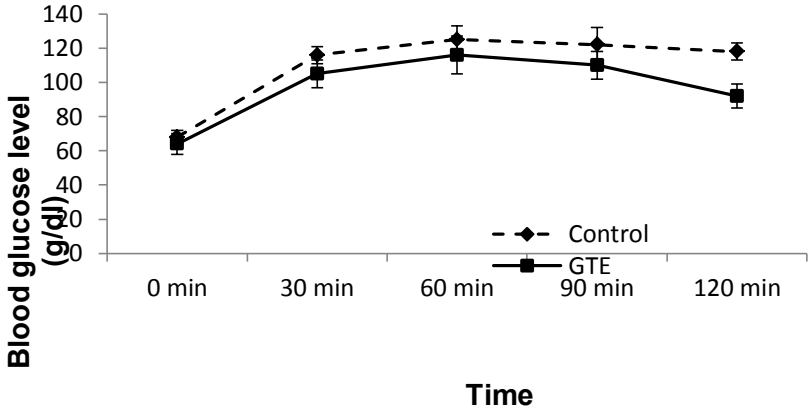

Figure 2. Oral glucose tolerance test of green tea extract (GTE) and control on Fructose-Fed Rats.

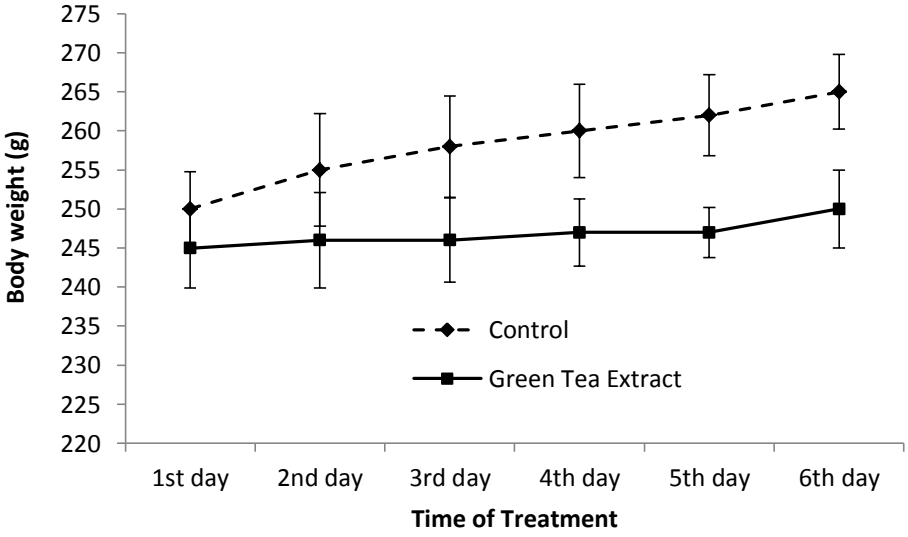

Figure 3. Green tea extract can reduces FFR body weight significantly for 6 days of treatment ($p < 0.01$).

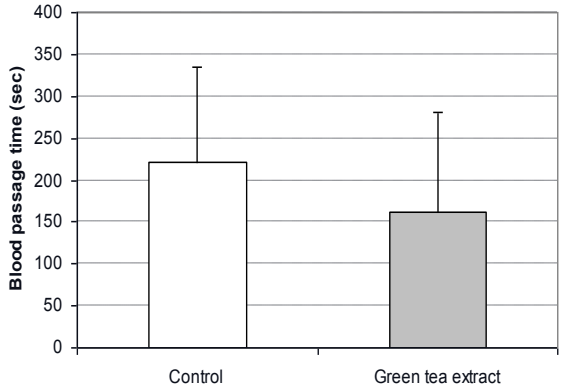

Figure 4. Effect of green tea extract on FFR blood fluidity measured by Micro-channel array flow analyzer (MC-FAN). Green tea extract lowers blood passage time significantly ($p < 0.01$).

Tabel 1.
Effect of green tea extract on blood parameters, fats and organs weight of Fructose-Fed Rat (FFR)

	Control (n = 5)	*Green Tea (n = 8)*	*P***
Blood parameter			
White blood cell ($x10^2$ /μl)	*51.07 ± 14.25*	*54.04 ± 11.48*	*NS*
Red blood cell ($x10^4$ /μl)	*773.40 ± 15.75*	*792.42 ± 33.36*	*NS*
Hemoglobin (g/dl)	*15.21 ± 0.28*	*15.63 ± 0.46*	*NS*
Hematocrite (%)	*48.35 ± 0.85*	*49.35 ± 1.48*	*NS*
MCV (fl)	*62.53 ± 0.84*	*62.31 ± 0.96*	*NS*
MCH (pg)	*19.66 ± 0.24*	*19.73 ± 0.34*	*NS*
MCHC (g/dl)	*31.47 ± 0.20*	*31.69 ± 0.20*	*NS*
Platelet ($x10^4$ /μl)	*111.86 ± 8.64*	*113.28 ± 15.80*	*NS*
Fat			
Abdominal fat (g)	*3.58 ± 0.68*	*2.03 ± 0.46*	*S*
Abdominal fat (g/100 g b.w)	*1.48 ± 0.23*	*0.87 ± 0.20*	*S*
Organs			
Spleen (g/100 g b.w)	*0.21 ± 0.02*	*0.20 ± 0.04*	*NS*
Pancreas (g/100 g b.w)	*0.20 ± 0.09*	*0.27 ± 0.05*	*S*
Kidney (g/100 g b.w)	*0.42 ± 0.02*	*0.35 ± 0.05*	*S*
Adrenal (g/100 g b.w)	*0.0073 ± 0.0034*	*0.0644 ± 0.1390*	*NS*
Genital (g/100 g b.w)	*0.63 ± 0.07*	*0.69 ± 0.05*	*NS*
Lung (g/100 g b.w)	*0.45 ± 0.04*	*0.48 ± 0.04*	*NS*
Thymus (g/100 g b.w)	*0.12 ± 0.08*	*0.10 ± 0.02*	*NS*
Liver (g/100 g b.w)	*3.88 ± 0.17*	*3.58 ± 0.31*	*NS*
Heart (g/100 g b.w)	*0.33 ± 0.01*	*0.35 ± 0.03*	*NS*

**Results are expressed as means ± SD.*
***Significance of difference between control and green tea ($p<0.05$); S=Significantly; NS=Not Significantly ($p>0.05$)*

DISCUSSION

The oral glucose tolerance test (OGTT) measures the body's ability to use glucose, as an energy source for the body. An OGTT can be used to diagnose prediabetes and diabetes. But, there is no significant differences were observed between control and green tea extract groups, although some researcher expects that high-fructose feeding could induce insulin resistance (Nakagawa et al., 2002). This fact may be due to the treatment is too short so insulin tolerance has not happened.

Green tea extract has been known as chemo preventive agent in various diseases, and EGCG as its major component has important role for this

activity. EGCG contain in some green tea leaf and products are different. Many in vitro studies on green tea report mechanisms consistent with protection against degenerative diseases (Huang et al., 1999, Nakagawa et al., 2002). Nevertheless, many of these studies used various concentrations of catechin and thus do not reflect certain catechin concentrations found in herbal extract. It is difficult to decide these results to EGCG concentration. However, because of the lack information of active compounds role and also biological effects of the conjugates, thus, animal studies are more relevant for investigating the physiological effects of catechins. This research studies the most interesting in vivo animal of the biological effects of standardized green tea extract on EGCG before used as herbal medicine to get sufficient effects.

The results showed that consumption of green tea extract for 6 days caused a suppressive effect on weight gain and visceral fat accumulation in FFR. Green tea extract contains caffeine and EGCG as the principal constituents, and these constituents showed a tendency to suppress body weight gain and visceral fat accumulation. Studies conducted with human subjects report reduced body weight and body fat, as well as increased fat oxidation and thermogenesis (Wolfram et al., 2006). Thus, these constituents (EGCG and coffein) are suggested to be partially involved in the suppressive effect on body weight gain and visceral fat accumulation.

Blood rheology is now receiving increasing attention as an important potential contributory factor to diabetic angiopathy (Le Dévéhat et al., 2004). Therefore, monitoring of the blood fluidity becomes very important in handling of diabetics impact. The use of green tea extract may increase blood fluidity, thereby reducing the impact of diabetic angiopathy.

CONCLUSIONS AND RECOMENDATION

Conclusions

It can be concluded that the water extract of green tea can decrease blood passage time of FFR, suppress body weight and visceral fat accumulation, but have no effect on blood parameter, insulin tolerance and organs weight except kidney and pancreas.

Recommendation

According to the result, it is suggest that green tea water extract may has beneficial effects for the treatment of diabetes and reduce blood viscosity.

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