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ARTICLE

Effects of Ramie Leaf Extract on Blood Glucose and Lipid Metabolism in *db/db* Mice

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Abstract Hypoglycemic and hypolipidemic effects of ramie leaf ethanol extract (RLE) on C75BL/KsJ-*db/db* mice were determined. The *db/db* mice were divided into diabetic control group (C), two experimental groups orally treated with low dose (200 mg/k, RLEL) and high dose (400 mg/kg, RLEH) of RLE. After 6 weeks, fasting blood glucose, serum insulin, and glycosylated hemoglobin levels decreased in RLE groups compared to those in the control group. The glucose levels in the oral glucose tolerance test and area under the curve for glucose in the RLE groups were also significantly lower than those in the control group (p <0.05). The serum total cholesterol and low-density lipoprotein cholesterol levels were significantly decreased in the RLEH group, whereas the serum HDL-cholesterol level was significantly increased in the RLEH group (p <0.05). These data suggest that RLE may improve blood glucose and lipid metabolism in mice with type 2 diabetes.

Keywords anti-diabetic effect · blood glucose · *db/db* mice · ramie leaf · serum lipid

Introduction

Diabetes is a chronic metabolic disease with the highest reported prevalence in the world. According to the International Diabetes Federation, 366 million people aged 20–79 years are known to be afflicted with this condition worldwide (International Diabetes Federation, 2012).

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In Korea, 3 out of 10 people are believed to have a risk of hyperglycemia. Recently there is an increasing trend of earlier development of this disease in younger generation (20–30 years old) and mostly would be prevalent after middle age. Diabetes is also associated with high mortality and is ranked as the 5th cause of death (NSOK, 2011; KDA, 2012). Type 2 diabetes that comprises most of the diabetic population in Korea is also known as non-insulin-dependent diabetes (NIDD), and it occurs due to the dysfunction of beta cells in pancreas and decreased insulin sensitivity. Type 2 diabetes has been known as disease of the Western population, but recently its occurrence has increased in Asian countries. This is largely attributed to the influence of Western diet and lifestyle. Furthermore, type 2 diabetes is an important risk factor of cerebrovascular and cardiovascular diseases, which are ranked as 2nd and 3rd leading causes of death, respectively. About 3/4 of cases with cardiovascular mortality are known to have type 2 diabetes as a concomitant disease, and it has been reported that type 2 diabetic patients have about 2-fold higher risk of cardiovascular disease (Schneider et al., 1998).

Presently, many types of hypoglycemic agents are approved for the treatment of diabetes, but these agents have limitations such as risk of lactic acid accumulation, exacerbation of renal failure symptoms, hepatotoxicity, and cause weight increase (Bailey, 1999). Therefore, there is an unmet need for hypoglycemic agents with a better benefit/low risk profile, which has led to studies on natural agents that have similar effects compared to patented drugs that can safely control blood glucose.

Ramie (*Boehmeria nivea* L.) is a flowering plant belonging to the family *Urticales* and has been widely used as a folk remedy or an emergency food (Institute of Drug and Plant, 1998). Roots and leaves of ramie are used as medicine, and the stem is used to make traditional clothes called *Mo-shi*. The young and soft parts of ramie leaves are used in dishes such as blanched and seasoned vegetables (*namul*), pickles, Kimchi, and rice cake. Leaf of ramie is a rich source of fiber, amino acids, vitamin C, calcium, potassium, and magnesium (Yoon and Jang, 1998), and has a

number of polyphenol and flavonoids that are antioxidants (Kim, 2010). In addition, chlorophyll is reported to have a strong inhibitory effect on free radicals (Kim et al., 1994). The studies on biological activity of ramie leaf include; antimicrobial activity (Son, 2007), antioxidant and anticancer effects (Kim et al., 2009b), preventive effect on constipation induced by loperamide in rats (Oh, 2012), improvement of lipid metabolism and anti-obesity effect in rats fed a high-fat/high-cholesterol diet (Lee et al., 2011). Many biological activity studies of ramie leaf have been reported from *in vitro* and *in vivo* trials; however, there has been no study on the anti-diabetic effects of ramie leaf.

Therefore, in the present study, ramie leaf ethanol extract was administered to *db/db* mice for 6 weeks to determine its effect on the lipid profile and change in blood glucose level, as well as to find out the applicability of ramie leaf as a functional food source for the improvement of dyslipidemia and as a hypoglycemic agent.

Material and Methods

Sample material. Ramie leaves were purchased from Ramie Association, Jeonam, Korea in August 2011. The leaves were washed three times with tap water in order to remove the sand and dust attached to the surface. The leaves were then rinsed carefully with fresh water and excess water was removed using a salad spinner (Caous, WINDAX, Korea). The rinsed leaves were dried for 40 min at 60°C by using a heat drier (GNO12, Hanil GNCO Co., Ltd., Korea). Finally, the dried leaves were triturated with a grinder and kept in a deep freezer at −70°C.

Preparation of ramie leaf extracts. Powdered ramie leaves (100 g) were added to 1,500 mL of 80% ethanol. The powder was extracted three times for 3 h at 65°C with heating mantle and reflux cooler, and then filtered with Whatman filter paper (No. 2, GE Healthcare, UK). The solvent was removed from the remaining extract in a 40°C water bath using a rotary vacuum evaporator (EYELA VACUUM NVC-1100, Japan). The extract was then decompressed, concentrated, followed by lyophilization, and stored at −70°C.

Determination of total polyphenolics, flavonoids and chlorogenic acid contents. Total polyphenolic content was determined with Folin-cicocalteu's reagent and tannic acid as an external standard (Folin and Denis, 1912). The results were expressed as mg tannic acid equivalent (TAE) per gram of sample. Contents of total flavonoids were measured according to the Davis method with slight modifications (Chae et al., 2002) with rutin as an external standard. The results were expressed as mg rutin equivalent (RE) per gram of sample. Total chlorogenic acid content was measured according to the Coseteng method (1987) with slight modifications with chlorogenic acid as an external standard.

Animal experiments. Thirty male C75BL/KsJ-*db/db* mice were purchased from Central Laboratory Animal Inc. (SLC Inc., Japan). The animals were housed in individual stainless steel cages under controlled conditions at temperatures between 21–3°C with a 12:12 h light-dark cycle (08:00–20:00 h light). Five-week-old *db/db* mice were provided with unrestricted access to rat chow and water in our animal facility for at least 2 weeks prior to the experiments. The *db/db* mice were randomly divided into three groups (n =8): control group, treated with vehicle (distilled water); RLEL group treated with RLE low dose, 200 mg/kg/day; and RLEH group treated with high dose of RLE, 400 mg/kg/day.

Animal diets were formulated based on AIN-93G (Reeves et al., 1993). The experimental groups were treated daily by oral administration of the same volume of RLE for 6 weeks. Food intake and body weight were measured on a daily and weekly basis. All experiments involving mice were performed according to the guidelines for the care and use of laboratory animals approved by Chosun University.

Collection of blood and organ. After 6 weeks of treatments, all mice were fasted for 12 h and then sacrificed under light anesthesia in carbon dioxide (CO_2)-saturated chamber. For whole blood and serum biomarker determination, the blood sample was withdrawn from the inferior vena cava. The blood was centrifuged at 3,000 rpm for 20 min to separate serum, which was then stored at −70°C. Liver and adipose tissues from subcutaneous, epididymal, retroperitoneal, and mesenteric adipose tissues were removed, washed in saline buffer, weighed, and immediately stored at −70 °C until further analysis.

Serum parameters. The levels of serum triglyceride (TG), total cholesterol (TC) and high-density lipoprotein cholesterol (HDL-C) were measured by using Fuji Dri-Chem 3500 analyzer (Fuji Photo film Co, Japan). Serum low-density lipoprotein cholesterol (LDL-C) was calculated using the Friedwald formula (1972) {total cholesterol – (HDL-cholesterol – TG/5)}. Serum insulin level was measured by using the insulin radioimmunoassay kit (EIKEN Chemical Co., Ltd., Japan). The blood sample was added with anticoagulant and then hemolyzed. HbA1c (glycosylated hemoglobin) level was analyzed with MicroMat™ II Hemoglobin Analyzer (Bio-Rad Laboratories, USA).

Blood glucose level, oral glucose tolerance test (OGTT) and area under the curve (AUC). Glucose levels of the animals were measured after a 12-h fasting period at week 0 (before treatment), week 2, week 4, and week 6 (last day of treatment). Blood glucose levels were determined from vein blood of tails and analyzed with Glucose Analyzer (Accu-Chek Active, Roche, Germany). The oral glucose tolerance test was performed at the last week of the experiment. The mice were given oral administration of glucose (2 g/kg of BW). After glucose administration, blood was taken from the tail vein at intervals of 0, 30, 60, 90, and 120 min to measure the blood glucose level. From the OGTT value, the AUC was calculated using the following formula: AUC = [(M2+M1)/2+(M3+M2)/2+(M4+M3)/2+(M5+M4)/2], where M1 is blood glucose level at 0 min, M2=30 min, M3=60 min, M4=90 min, and M5=120 min. This procedure was performed as suggested by Pruessner et al. (2003).

Statistical analysis. The statistical analysis was carried out using Statistical Package for the Social Sciences (SPSS Inc., USA, version12.0). All data were expressed as the means ± standard error (SE). Statistical analysis was performed using an analysis of variance followed by Tukey's test. Student's t test was used to confirm comparisons between the groups. Statistical significance was considered to be at the p <0.05 level.

Results and Discussion

Total polyphenolics, flavonoids, and chlorogenic acid contents. Phenolic compounds such as flavonoids and chlorogenic acid are considered to be the major contributors to the antioxidant ability of plants (Ong et al., 2013). Thus, the flavonoids and chlorogenic acid of RLE were evaluated. Total polyphenolics, flavonoids, and chlorogenic acid contents in RLE were found to be 132.50±2.76 TAE, 119.00±1.15 RE, and 15.24±0.65 mg/g, respectively (Table 1). In the study of Lee et al. (2009), total polyphenol content in RLE was149.58±1.62 TAE mg/g DW, similar to the results of the present study. However, total flavonoid content of RLE was 49.24±0.38 TAE mg/g DW (Lee et al., 2009). A higher content of total flavonoids was shown in the present study. It was reported that RLE had the highest antioxidant capacity, due to being rich in total polyphnolics, flavonoids, and chlorogenic acid. Furthermore, chlorogenic acid is the major phenolic compounds in RLE (Tan et al., 2014) and has many biological effects such as antioxidant, antidiabetic, and antilipidemic (Ong et al., 2013).

Table 1 Contents of total polyphenol, total flavonoids and chlorogenic acid of 80% ethanol extracts of ramie leaf

Sample	Total polyphenol (TAE mg/g DM)	Total flavonoids (RE mg/g DM)	Chlorogenic acid (mg/g DM)
RLE	132.50±2.76	119.00±1.15	15.24±0.65

Body weight, food intake, and water consumption. We studied the changes in body weight, food intake, and water consumption after RLE administration for 6 weeks in *db/db* mice (Table 2). The body weight increased gradually during the 6-week experimental period. There were no significant differences in body weights between the treatment groups and the control group. The food intake and water consumption were slightly lower in the treatment groups than in control group, but the difference was not found to be significant. These results were similar to the ones reported by Kim et al. (2009a), which showed that body weight, food intake, and water intake were not significantly reduced by the administration of loquat leaf methanol extract to *db/db* mice.

Weights of liver and adipose tissue. The weight of liver of *db/db* mice are shown in Table 3. Kim et al. (2012) described that the surface of liver tissue of diabetic *db/db* mice was noticeable less shiny with low elasticity. This was due to fat disposition in this organ. In a severe case of diabetic, high fat disposition, hepatocytes necrosis, and sedimentation of inflammatory cells in portal vein were increased. In accordance with that, Lee et al. (2010) reported that the weight of liver in diabetic *db/db* mice increased by 3.36 times than that of normal non-diabetic mice. In the present study, the weight of liver from treatment groups (RLEL and RLEH) was slightly lower than in control group, but the difference was not significant.

The *db/db* mice are known to suffer with obesity due to the

Table 2 Weight gain, food intake, and water consumption of *db/db* mice after treated orally with ramie leaf ethanol extract (RLE) for 6 weeks

Groups[1)]	Body weight (g)			Food intake (g/day)	Water consumption (mL/day)
	initial	final	gain		
Control	31.57±0.21[2)NS3)]	36.12±0.38[NS]	4.55±0.18[NS]	6.66±0.54[NS]	9.01±0.47[NS]
RLEL	31.57±0.23	35.97±0.41	4.46±0.25	6.40±0.43	8.85±0.24
RLEH	31.59±0.34	35.56±0.16	4.17±0.28	6.15±0.25	8.70±0.19

[1)]RLEL: Ramie leaf ethanol extract-low (200 mg/kg/day), RLEH: Ramie leaf ethanol extract-high (400 mg/kg/day).
[2)]Values are mean ± SE of 8 rats in each group.
[3)]NS: not significantly different among groups.

Table 3 Weights of liver, subcutaneous, mesenteric, epididymal, retroperitoneal, and total adipose tissue in *db/db* mice after treated orally with ramie leaf ethanol extract (RLE) for 6 weeks

Groups[1)]	Liver wt.	Subcutaneous wt.	Mesenteric wt.	Retroperitoneal wt.	Epididymal wt.	Total adipose tissue wt.
	(g/100 g body wt)					
Control	5.98±0.05[2)NS3)]	1.62±0.12[NS]	1.15±0.05[a4)]	0.61±0.08[NS]	1.75±0.13[NS]	5.13±0.07[a]
RLEL	5.82±0.07	1.55±0.11	0.94±0.03[ab]	0.49±0.06	1.48±0.15	4.46±0.08[b]
RLEH	5.67±0.07	1.52±0.13	0.85±0.04[b]	0.47±0.12	1.41±0.13	4.25±0.16[b]

[1)]See the legend of Table 1.
[2)]The results are mean ± SE of 8 mice in each group.
[3)]NS: not significantly different among groups.
[4)]Values with different superscripts in the same column are significantly different (p <0.05) between groups by Tukey's test.

Table 4 Change in fasting blood glucose level in *db/db* mice after treated orally with ramie leaf ethanol extract (RLE) for 6 weeks

Groups[1)]	week-0	week-2	week-4	week-6
	(mg/dL)			
Control	325.65±12.12[2)NS3)]	412.75±6.54[NS]	454.65±8.09[a4)]	482.21±13.27[a]
RLEL	318.23±10.07	404.36±7.53	421.13±10.43[ab]	408.33±9.54[b]
RLEH	322.26±11.54	391.86±11.70	387.08±13.11[b]	391.34±10.21[b]

[1)]See the legend of Table 1.
[2)]The results are mean ± SE of 8 rats in each group.
[3)]NS: not significantly different among groups.
[4)]Values with different superscripts in the same column are significantly different (p <0.05) between groups by Tukey's test.

metabolic disorders related to diabetes and resistance to leptin (Considine et al., 1996); hence, the effect of administration of RLE on the weights of white adipose tissues including subcutaneous, epididymal, and retroperitoneal as well as mesenteric adipose tissue was investigated (Table 3). The weights of subcutaneous, epididymal, and retroperitoneal adipose tissues were decreased in the RLEL and RLEH groups as compared to control group, but there was no statistical significance observed. The weight of mesenteric adipose tissue, decreased in a dose-dependent manner in relation to the administration of RLE (p <0.05). Furthermore, administration with RLE effectively reduced the weight of total adipose tissue (p <0.05). In another study by Lee et al. (2011), obesity-induced rats were fed high-fat/high-cholesterol diet with ramie leaf powder for 4 weeks. They reported that mesenteric and epididymal adipose tissue weights in treatment group decreased significantly compared to that of control group.

The observations presented above indicate that the administration of RLE does not have a considerable effect on the reduction of liver weight; however, there was a significant decrease in mesenteric adipose tissue and total adipose tissue weights. This suggests that the ramie leaf inhibits the accumulation of adipose tissues and therefore is effective in reducing obesity.

Levels of glucose in whole blood. The alteration of fasting blood glucose levels after administration with RLE are presented in Table 4. The result revealed that the blood glucose levels of RLEL and RLEH groups decreased similarly to that of control group, although up to 2 weeks, and there were no significant differences among groups. However, from week 4 onwards, there was a significant decrease of glucose levels in the RLEH group compared to that of the control group (p <0.05). On week 6, both RLEL and RLEH groups showed significantly lower blood glucose levels compared to the control group. There was a slight increase of the final blood glucose levels in all groups compared to its initial values. Interestingly, the increasing rates were becoming lower in longer treatment period, thus the longer RLE administration, the lower of blood glucose levels (p <0.05). Lawes et al. (2004) reported that there was 21% decrease in the risk of seizure and 23% decrease in the risk of ischemic heart disease per mM/L decrease in fasting blood glucose level. This clearly suggests the importance of blood glucose management in diabetes. Also, the oxidative stress due to high blood glucose levels plays an important role in the prevalence of diabetic complications and causes diabetic retinopathy, neuropathy, and nephrosis, hence the blood glucose management is an important factor in the treatment of diabetes (Mullarkey et al., 1990). Study of Kim et al. (2006) had results similar to those of our study. They administered pine leaf extract to *db/db* mice for 6 weeks, and found no difference in blood glucose levels up to week 2, but from week 3 onwards, they observed significant decrease in blood glucose levels in pine leaf extract high concentration group compared to that in control group. At weeks 5 and 6, there was a significant decrease in blood glucose levels in both the low and high concentration groups compared to that in the control group. They suggested that the reason for this decrease in blood glucose level could be due to the increase in serum leptin levels, which may have affected the insulin-related sensitivity. Similar study was reported by Lee et al. (2012). They administered chlorogenic acid, which is rich in ramie leaf, to high-fat diet and streptozotocin-induced diabetic mice for 6 weeks. Although the blood glucose level between the test groups was comparable at the start of the study, treatment groups showed a significant decrease of blood glucose levels compared to control group. Blood glucose levels decreases were also reported by Jung et al. (2006) on caffeic acid treatment to *db/db* mice and Jeon and Choi (2010) on naragin 0.02%, a type of flavonoid, to mice fed a high fat diet. Polyphenols and flavonoids are known to be therapeutically useful in diabetic rats due to their ability to act on multiple targets associated with the pathology of diabetes (Chang and Jhonson, 1980; Shanmugasundaram et al., 1990). These reports suggest that polyphenolic compounds and flavonoid are responsible for the hypoglycemic effects of ramie leaf extract (Kim, 2010). However, it is not clear whether the mechanism of action is affected by increasing insulin secretion or by overcoming insulin resistance. Even though the mechanism is not yet clear, our study has found that the RLE inhibits the increase in blood glucose.

Levels of insulin and HbA1c. Serum insulin and blood HbA1c levels are shown in Table 5. NIDD patients are the most common of type 2 diabetics, which is associated with hyperinsulinemia that results from insulin resistance. NIDD is primarily caused by several factors such as obesity, overeating, lack of exercise, age, alcohol intake, mental stress, and genetics. It also causes glucose intolerance and hyperinsulinemia, which play important roles in the etiology of lipid metabolism dysfunction, high blood pressure, and coronary artery disease (Choi and Koo, 2000). Our study

Table 5 Serum insulin and blood HbAlc levels in *db/db* mice after treated orally with ramie leaf ethanol extract (RLE) for 6 weeks

Groups[1)]	Serum insulin (μIU/mL)	Blood HbAlc (%)
Control	57.98±1.25[2)a3)]	9.51±0.15[a]
RLEL	48.37±1.83[b]	9.12±0.13[ab]
RLEH	45.98±0.89[b]	8.65±0.17[b]

[1)]See the legend of Table 1.
[2)]The results are mean ± SE of 8 rats in each group.
[3)]Values with different superscripts in the same column are significantly different (p <0.05) between groups by Tukey's test.

Table 6 Oral glucose tolerance test of *db/db* mice after treated orally with ramie leaf ethanol extract (RLE) for 6 weeks

Time (min)	Control	RLEL	RLEH
	(mg/dL)		
0	489.32±12.23[1)a2)]	416.02±8.98[b]	404.12±11.34[b]
30	632.21±15.58[a]	581.87±10.29[b]	561.76±14.24[b]
60	578.24±10.20[a]	515.35±13.76[b]	495.23±10.62[b]
90	546.21±11.05[a]	480.14±9.45[b]	457.22±12.32[b]
120	529.45±12.43[a]	447.86±11.98[b]	417.64±9.27[b]
AUC_{OGTT}[3)]	2,266.05±67.87[a]	2,009.30±87.05[b]	1,925.04±76.25[b]

[1)]The results are mean ± SE of 8 rats in each group.
[2)]Values with different superscripts in the same row are significantly different (p <0.05) between groups by Tukey's test.
[3)]AUC_{OGTT}: the area under the curve of glucose.

showed that serum insulin levels were significantly lower in the RLEL and RLEH groups than those in the control group (p <0.05). The levels of blood HbA1c was significantly lower in the RLEH group than control group (p <0.05). Park et al. (2007) administered crude polysaccharides from *Grifola frondosa* for 8 weeks to type 2 diabetes model (KK-Ay) mice. They reported that levels of both serum insulin and HbA1c were significantly reduced in the test group than the control group, suggesting that hypoglycemic effect was attributed to the administration of polysaccharides, which would increase insulin sensitivity despite the low level of insulin in diabetic mice. In alliance to Park et al. (2007), our result showed a decreased level of serum insulin and blood glucose as well as HbA1c after administration of the RLE, which suggested that RLE is effective in improving insulin sensitivity, thereby showing hypoglycemic effect.

Oral glucose tolerance test and area under the curve Changes in the levels of blood glucose and AUC after glucose administration during OGTT are shown in Table 6. OGTT measures the postprandial changes in blood glucose levels. The blood glucose levels of mice in the RLEL and RLEH groups were significantly lower than those of the control at 30, 60, 90, and 120 min after glucose administration (p <0.05). The RLE treatment significantly improved glucose tolerance over the entire OGTT monitoring intervals in the *db/db* mice. The AUC for both RLEL and RLEH groups were significantly decreased compared to the control (p <0.05). They were 11.33 and 15.05% for RLEL and RLEH, respectively. Similar result was reported by Lee et al. (2012) on diabetic mice after treated with chlorogenic acid in ramie leaf accompanied by high fat diet. Plant derived phenolic compounds were known to inhibit the activation of glycolytic and fermentation enzymes such as α-amylase, sucrase, and α-glucosidase in the small intestine, resulting in a decrease in blood glucose and insulin levels (Matsumoto et al., 1992). The improvement on OGTT and AUC status on db/db mice after RLE treatment could be attributed to ramie leaf's phenolic contents, chlorogenic acid and soluble fibers. This was supported by reports of Jenkins et al. (1977), Albrink et al. (1979), Kim et al. (1994) and Kim (2010). Jenkins et al. (1977) reported that the feeding of water-soluble fibers, such as guar gum and pectin, reduced the serum levels of insulin, whereas Albrink et al. (1979) stated that increased administration of dietary fiber for the treatment of diabetics would reduce the insulin requirement as well as decreased the glucose levels in urine. Therefore, it can be implied that the hypoglycemic effect observed in our study after administration of RLE is due to the presence of phenolic compounds and soluble dietary fiber in the ramie leaf (Kim et al., 1994; Kim, 2010).

Levels of serum TG, TC, HDL-C and LDL-C. The changes in serum TG, TC, HDL-C, and LDL-C levels after administration of RLE are shown in Table 7. An imbalance in the lipid metabolism is an important indicator in the treatment of diabetes. In the present study, although the level of TG in serum decreased in both RLEL and RLEH groups compared to the control, there was no significant difference. The levels of TC in serum decreased in a dose-dependent manner in both treatment groups. A significant decline was observed in RLEH group (p <0.05); however, the

Table 7 Contents of triglyceride, total cholesterol, HDL-cholesterol, and LDL-cholesterol in serum of *db/db* mice after treated orally with ramie leaf ethanol extract (RLE) for 6 weeks

Groups[1)]	Triglyceride	Total cholesterol	HDL-C	LDL-C
	(mg/dL)			
Control	64.87±6.32[2)NS3)]	138.76±5.94[a4)]	24.36±0.52[b]	73.98±1.22[a]
RLEL	154.76±6.70	127.64±7.65[ab]	26.98±0.47[ab]	64.90±2.07[b]
RLEH	143.45±7.23	108.32±5.87[b]	31.26±0.72[a]	60.14±1.59[c]

[1)]See the legend of Table 1.
[2)]The results are mean ± SE of 8 rats in each group.
[3)]NS: not significantly different among groups.
[4)]Values with different superscripts in the same column are significantly different (p <0.05) between groups by Tukey's test.

decrease in RLEL group was not significant compared to the control group. The serum LDL-C levels were significantly lower in both RLEL and RLEH groups compared to the control group (p <0.05), and there was a significant difference between both treatment groups (p <0.05).

Generally, the diabetic mice are known to inhibit the HDL-C formation due to a low activity of lipoprotein lipase that reduces the breakdown of lipoproteins, mainly the TG (Betteridge, 2001). In the present study, the serum HDL-C level increased in a dose-dependent manner after administration of RLE compared to the control group, with significant difference observed in RLEH group (p <0.05). In a similar study by Lee et al. (2011), administration of 5 and 10% ramie leaf powder for 4 weeks to rats fed a high fat/high-cholesterol diet were found to decrease the serum TG, TC, and LDL-C levels, whereas the level of HDL-C was significantly increased. This result was also confirmed by Roberfroid (1993), who stated that in terms of bioactive compounds related to the improvement in lipid metabolism, the consumption of dietary fiber decreased serum TG and TC levels. On the other hand, study of Jeon and Choi (2010) showed that administration of naringin to mice fed a high fat diet has no effect on the level of serum TG, in contrast there was a significant decrease in the serum TC level as well as a significant increase in the serum HDL-C level. Chlorogenic acid is found as an anti-lipoperoxidant agent that significantly reduces lipid peroxidation in liver tissues and red blood cells of diabetic mice (Shanmugasundaram et al., 1990). In the present study, RLE administration attenuated the serum TC and LDL-C levels and increased serum HDL-C level. This could be due to the presence of major flavonoids namely chlorogenic acid or dietary fiber (Roberfroid, 1993; Yoon and Jang, 2006; Kim, 2010; Lee et al., 2012). Based on our results, RLE would be effective in reducing dyslipidemia in diabetes; therefore, it will help in lowering the risk of cardiovascular disease in patients with diabetes.

In conclusion, the administration of RLE would suppress the raise of postprandial blood glucose, HbA1c, serum insulin, and fasting blood glucose levels. It was also discovered as an effective way to improve serum lipid profile and reducing adipose tissue weight in the *db/db* mice. These results suggest that ramie leaf has the potential as a functional food ingredient to prevent and/or manage type 2 diabetes. However, further studies will be required to elucidate the mechanism underlying the hypoglycemic and hypolipidemic effects of ramie leaf.

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