Songklanakarin J. Sci. Technol.
31 (2), 133-137, Mar. - Apr. 2009

Songklanakarin Journal of Science and Technology
http://rdo.psu.ac.th/sjst

Original Article

The hypoglycemic effect of water extract from leaves of *Lagerstroemia speciosa* L. in streptozotocin-induced diabetic rats

Amornnat Thuppia[1]*, Pornrut Rabintossaporn[1], Suphaket Saenthaweesuk[1], Kornkanok Ingkaninan[2] and Seewaboon Sireeratawong[1]

[1] *Department of Preclinical Science, Faculty of Medicine, Thammasat University, Rungsit Campus, Khlong Luang, Pathum Thani, 12120 Thailand.*

[2] *Department of Pharmaceutical Chemistry and Pharmacognosy, Faculty of Pharmaceutical Science, Naresuan University, Muang, Phitsanulok, 65000 Thailand.*

Received 4 June 2008; Accepted 11 February 2009

Abstract

The purpose of this study was to evaluate the hypoglycemic effect of the extract from the leaves of *Lagerstroemia speciosa* L. in normal and streptozotocin (STZ)-induced diabetic rats. Diabetes mellitus (DM) was induced in rats by intraperitoneal injection of streptozotocin at a dose of 45 mg/kg. Extract at the doses of 500, 1000 and 2000 mg/kg, glibenclamide (3 mg/kg) and vehicle were administered orally for 12 days. Blood glucose levels were measured on the 5th and 12th days of the experiment and at three days after ceasing the extract administration. A significant ($p<0.05$) decrease in fasting blood glucose level was observed in diabetic rats that received the extract at the doses of 1000 and 2000 mg/kg on the 5th and 12th days of administration when compared to the control diabetic group. However, in an oral glucose tolerance test study, none of the doses of the extract showed any effects on blood glucose level in either diabetic or normal rats. The histological examination of liver showed some vacuoles in the cytoplasm of hepatocytes of normal rats receiving the extract at a dose of 2000 mg/kg. This result demonstrates that the water extract from leaves of *L. speciosa* can reduce fasting blood glucose of STZ-induced diabetic rats. Future studies are needed to identify its mechanism for controlling DM and to investigate its toxicity.

Keywords: Hypoglycemic effect, *Lagerstroemia speciosa* L.

1. Introduction

Lagerstroemia speciosa L., commonly known as "intaninnam" or "tabakdam" in Thailand, has been used for treating diabetes mellitus (DM) and kidney-related disease (Klein *et al.*, 2007). The effective compounds from the methanol extract of *L. speciosa* leaves were identified as corosolic acid and triterpene, which have been previously reported to have antidiabetic activity as well as being an activator of glucose uptake in Ehrlich ascites tumor cells (Murakami *et al.*, 1993). The corosolic acid from the extract of *L. speciosa* leaves was found to reduce blood glucose level and plasma insulin level in KK-Ay diabetic mice (a model of type II diabetes) (Kakuda *et al.*, 1996; Miura *et al.*, 2006) as well as to stimulate glucose uptake in 3T3-L1 cells (Liu *et al.*, 2001). Furthermore, ellagitannins, which are found in the water extract of *L. speciosa*, acted as an activator of the glucose transportation in fat cells (Hayashi *et al.*, 2002). The most potent ellagitannin was Lagerstroemin which could enhance the glucose uptaking process in rat adipocytes both intracellularly (by increasing the [3H]2-deoxy-*D*-glucose) and extracellulary (by binding to the insulin receptor). Moreover, Lagerstroemin has been shown

*Corresponding author.
Email address: phungamornnat@hotmail.com

to possess insulin-like actions such as the activation of insulin receptor and tyrosine kinase, the inhibition of tyrosine phosphatases, as well as effects on lipolysis and the Erk activity (Hayashi *et al.*, 2002; Hattori *et al.*, 2003).

Streptozotocin [STZ or 2-deoxy-2-(3-(methyl-3-nitrosoureido)-D-glucopyranose] is widely used to induce DM under experimental conditions (Szkudelski, 2001). The diabetogenic action of STZ was shown with a highly specific cytotoxic action on the pancreatic β-cells (Hirotani *et al.*, 2007). STZ has also caused rapid and irreversible necrosis of pancreatic islets of Langerhans leading to an impaired glucose tolerance in rats and insulin deficiency (Hirotani *et al.*, 2007). Pancreatic β-cells play a major role in insulin secretion to keep blood glucose in normal range and force cells of the body to absorb and use glucose, thereby lowering the blood glucose level.

In this study, we observed the hypoglycemic effect of the water extract from leaves of *L. speciosa* in normal and streptozotocin-induced diabetic rats.

2. Materials and Methods

2.1 Plant material

Leaves of *L. speciosa* were collected from Phitsanuloke, Thailand. The voucher specimen (Tothium 020) was kept by PBM herbarium, Faculty of Pharmaceutical Sciences, Mahidol University, Nakorn Pathom, Thailand.

2.2 Preparation of plant extract

The 2254 g oven-dried leaves of *L. speciosa* were finely chopped and boiled in a stainless boiler for 2 hours and then filtered. The filtrate was collected and freeze dried using the FTS system evaporator model Dura Dry (Science Engineer International CO., LTD., USA). The weight and percentage yield of the water extract of *L. speciosa* were recorded. The percentage yield was 6.34.

2.3 Experimental animals

Male Sprague-Dawley rats weighing 250-270 g were obtained from the National Laboratory Animal Center, Nakorn Pathom, Thailand. They were all maintained in environmentally controlled condition at 25±1°C under a 12-hour-dark-light cycle and given a standard diet and water throughout the experimental period. All experimental protocols for animal studies were approved by the Animal Ethics Committee of Faculty of Medicine, Thammasat University (No.0008/2007).

After a one-week stabilization period, STZ was injected into rats at a dose of 45 mg/kg. Blood samples were taken from rat tail for blood glucose analysis on the eighth and eleventh days after STZ injection. Rats with blood glucose levels of 180 mg/dl or more on both days were categorized as a diabetic group.

2.4 Hypoglycemic test

2.4.1 Diabetic rats

Thirty male diabetic rats were divided into five groups (n = 6). Group 1 (control group) was given the vehicle whereas Group 2 given 3 mg/kg glibenclamide. Groups 3, 4 and 5 served as the experimental groups and received the extract at the doses of 500, 1000 and 2000 mg/kg, respectively.

2.4.2 Normal rats

Twenty-four male normal rats were divided into four groups (n = 6). Group 1 (control group) was given the vehicle. Groups 2, 3 and 4 served as the experimental groups and received the extract at the doses of 500, 1000 and 2000 mg/kg, respectively.

A basal blood glucose level was recorded on day 0 prior to the extract administration. All rats were administered the extract once a day for 12 days. All rats were fasted for 18 hours before they were tested for the blood glucose level. All rats were tested for the fasting blood glucose level on the 5^{th} and 12^{th} days of the experimental period and at three days after ceasing the extract administration.

2.5 Oral glucose tolerance test (OGTT)

2.5.1 Diabetic rats

Thirty male diabetic rats were divided into five groups (n = 6). Group 1 (control group) was given the vehicle whereas Group 2 given 3 mg/kg glibenclamide. Groups 3, 4 and 5 served as the experimental groups and received the extract at the doses of 500, 1000 and 2000 mg/kg, respectively.

2.5.2 Normal rats

Twenty-four male normal rats were divided into four groups (n = 6). Group 1 (control group) was given the vehicle. Groups 2, 3 and 4 served as the experimental groups and received the extract at the doses of 500, 1000 and 2000 mg/kg, respectively.

All rats were fasted for 18 hours. After that, all subjects in each group were orally administered vehicle, glibenclamide or the leaf extract, and their blood glucose levels were measured and defined as the basal blood glucose levels. After 30 minutes, each rat was given glucose orally at a dose of 3 g/kg, and their blood glucose levels were measured at the following 30, 60, 90, 120, 150 and 180 minutes.

2.6 Statistical analysis

Level of blood glucose were reported as mean ± standard error of mean (SEM). Statistical significance was tested by one-way ANOVA and post hoc least-significant difference

(LSD) test. *P* values less than 0.05 were considered significant.

3. Results

3.1 Hypoglycemic test

Fasting blood glucose levels of the normal and diabetic rats in the hypoglycemic effect study are as shown in Table 1. There was no significant difference in the fasting blood glucose levels between the normal rats receiving the extract and the control group. The diabetic rats receiving the extract at doses of 1000 and 2000 mg/kg body weight had significantly decreased blood glucose levels on the 5th and 12th days of administration when compared with the control group, whereas the extract at a dose of 500 mg/kg did not show any significant effect on lowering the fasting blood glucose level. At three days after the end of experimental period, the fasting blood glucose of diabetic rats from all groups except those receiving glibenclamide returned to approximate basal blood glucose levels.

3.2 Oral glucose tolerance test

As shown in Figure 1, blood glucose levels of normal rats in all groups reached their peaks at the 30th minute after taking D-glucose and gradually declined until the end of the experiment. There were no significant differences when compared to the control group.

The results of the OGTT study in diabetic rats are shown in Figure 2. The blood glucose level of the diabetic rat group receiving glibenclamide reached its peak at the 30th minute and significantly decreased until the end of the experimental period. The blood glucose levels in control and treated group at doses of 500 and 1000 mg/kg body weight reached their peaks at the 60th minute and gradually decreased until the end of the experimental period. The extract at a dose of 2000 mg/kg body weight also produced a blood glucose level peak at the 30th minute, and the level gradually decreased until the end of the experiment, but this not significant when compared to the control group.

4. Discussion

The present study was designed to evaluate the hypoglycemic effect of the water extract from *L. speciosa* in normal and STZ-induced diabetic rats.

In the hypoglycemic test, none of the doses of the *L. speciosa* extract showed any effects in lowering the fasting blood glucose level in normal rats, whereas the extract at the doses of 1000 and 2000 mg/kg body weight could reduce the blood glucose level in STZ-induced diabetic rats. The demonstrated antihyperglycemic effects of the water extract of *L. speciosa* in diabetic rats may be because the extract could recover or repair the pancreatic β-cells and hepatocytes from STZ-induced damage. Furthermore, the period of administration of the extract and the amounts of the active ingredients may be suitable for lowering of blood glucose levels. On the other hand, the water extract of *L. speciosa* did not show any effects in lowering the level of fasting blood glucose in normal rats. This may be caused by the pancreatic β-cells and hepatocytes not being destroyed in the first place, enabling them to maintain normal function in regulating the blood glucose level. Moreover, this result may be explained by the ability of the intact pancreatic β-cells to regulate glucose homeostasis, and that target tissues may still

Table 1. Effect of a 12-day oral administration of the water extract from *Lagerstroemia speciosa* L. (*L. speciosa*) leaves on the fasting blood glucose level in normal rats and STZ-induced diabetic rats.

Experimental groups	Fasting Blood glucose (mg/dl)			
	Day 0	Day 5	Day 12	3 day withdrawn
Normal rats				
Vehicle	121.00±3.34	99.83±2.86	112.00±3.82	107.83±2.90
L. speciosa 500 mg/kg	122.67±3.18	100.17±2.44	108.00±7.26	140.50±22.32
L. speciosa 1000 mg/kg	124.83±4.83	99.83±3.81	117.17±3.90	118.83±1.72
L. speciosa 2000 mg/kg	107.83±6.33	100.33±4.22	113.67±6.70	111.50±1.84
Diabetic rats				
Vehicle	296.83±25.27	298.17±58.82	320.50±36.54	343.83±28.16
Glibenclamide	344.67±19.76	178.83±44.56 *	166.00±12.80*	198.83±30.32*
L. speciosa 500 mg/kg	325.00±12.45	264.83±7.78	282.00±51.26	276.17±15.88
L. speciosa 1000 mg/kg	302.67±14.66	191.83±25.30*	170.50±11.69*	303.67±40.80
L. speciosa 2000 mg/kg	317.67±19.93	194.33±12.08*	186.00±26.43*	301.50±33.77

Values are expressed as mean ± S.E.M., n= 6
*Significantly different from vehicle, $p<0.05$

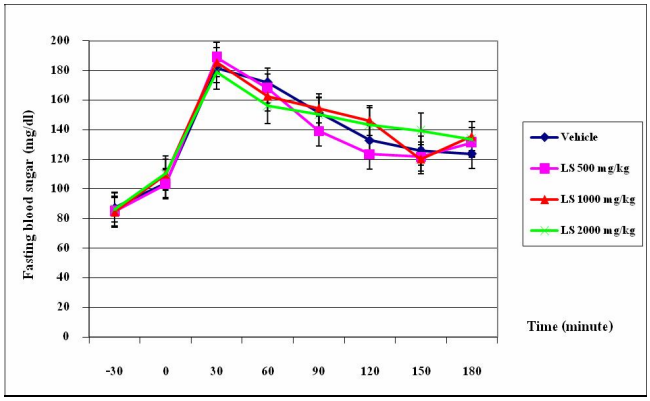

Figure 1. The fasting blood glucose level of the normal rats in OGTT.
* Significantly different from vehicle, $p<0.05$.

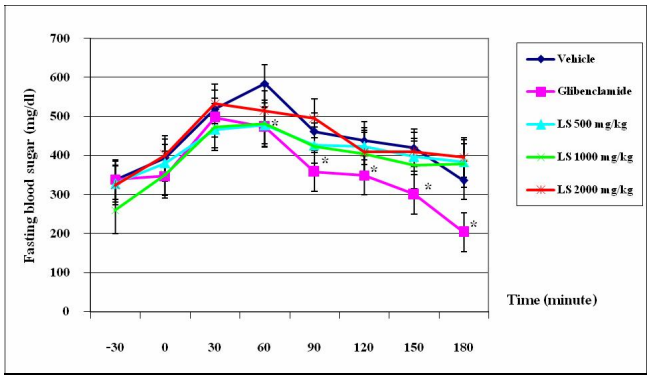

Figure 2. The fasting blood glucose level of the STZ-induced diabetic rats in OGTT.
* Significantly different from vehicle, $p<0.05$.

have the normal response to insulin.

Hayashi *et al.* (2002) reported that the water extract from leaves of *L. speciosa* contains active compounds such as tannin, reginin A, ellagitannins, Lagerstroemin, flosin B, triterpene and corosolic acid; some of which lower the blood glucose level. These active compounds might cause glucose lowering by increasing glucose transport to peripheral tissue (Hayashi *et al.*, 2002; Liu *et al.*, 2005); in particular, the corosolic acid also improves glucose metabolism by reducing insulin resistance (Miura *et al.*, 2006; Yamada *et al.*, 2008).

In the OGTT study, none of the doses of the extract reduced the blood glucose levels in normal or diabetic rats. This result may be due to the period of receiving the extract. The diabetic rats in OGTT study only received a single dose of the extract, which may not be enough for repairing or improving the pancreatic β-cells or hepatocytes and may not directly stimulate insulin secretion from pancreatic β-cells. However, Fukushima *et al.* (2006) reported that corosolic acid, the active compound of *L. speciosa*, could suppress the postchallenge plasma glucose level at 90 minutes. The explanation for this difference may be that this study used the crude extract in which the efficacy of the active compound may not be enough to reduce the blood glucose levels in OGTT.

In conclusion, the present study suggests that the crude extract from leaves of *L. speciosa* administered orally at the doses of 1000 and 2000 mg/kg could reduce the blood glucose level. It is possible that the extract could increase glucose transport to peripheral tissue and improve glucose metabolism by reduced insulin resistance. Further investigation should evaluate the b-cells function, pancreatic insulin level and the effective dose as well as the longer period of administration to better elucidate the mechanism for controlling DM and the toxicity of this extract.

Acknowledgements

This project was financially supported by Thammasat University Research Fund. The authors would like to acknowledge Assistant Professor Dr.Auragun Poomkokruk, Dr. Pholawat Tingpej, Dr. Praphaporn Stewart and Michael James Stewart for their help in proofing and editing the manuscript.

References

Fukushima, M., Matsuyama, F., Ueda, N., Egawa, K., Takemoto, J., Kajimoto, Y., Yonaha, N., Miura, T., Kaneko, T., Nishi, Y., Mitsui, R., Fujita, Y., Yamada, Y. and Seino, Y. 2006. Effect of corosolic acid on postchallenge plasma glucose levels. Diabetes Research and Clinical Practice. 73(2), 174-177.

Hayashi, T., Maruyama, H., Kasai, R., Hattori, K., Takasuga, S. and Hazeki, O. 2002. Ellagitannins from *Lagerstroemia speciosa* as activators of glucose transport in fat cell. Planta Medica. 68, 171-173.

Hattori, K., Sukenobu, N., Sasaki, T., Takasuga, S., Hayashi, T., Kasai, R., Yamasaki, K. and Hazeki, O. 2003. Activation of insulin receptors by Lagerstroemin. Journal of Pharmacological Sciences. 93, 69-73.

Hirotani, Y., Ikeda, T., Yamamoto, K. and Kurokawa, N. 2007. Effects of Hachimi-jio-gan (Ba-Wei-Di-Huang-Wan) on Hyperglycemia in Streptozotocin-Induced Diabetic Rats. Biological and Pharmaceutical Bulletin 30(5), 1015-1020.

Kakuda, T., Sakane, I., Takihara, T., Ozaki, Y., Takeuchi, H. and Kuroyanagi, M. 1996. Hypoglycemic effect of extracts from *Lagerstroemia speciosa* L. leaves in genetically diabetic KK-AY mice. Bioscience, Biotechnology, and Biochemistry. 60, 204-208.

Klein, G., Kim, J., Himmeldrik, K., Cao, Y. and Chen, X. 2007. Antidiabetes and anti-obesity activity of *Lagerstroemia speciosa*. Evidence-based Complementary and Alternative Medicine. 4, 401-407.

Liu, F., Kim, Y., Li, Y., Liu, X., Li, J. and Chen, X. 2001. An extract of *Lagerstroemia speciosa* L. has insulin-like glucose uptake-stimulatory and adipocyte differentiation-inhibitory activities in 3T3-L1 cells. Journal of

Nutrition. 131, 2242-2247.

Liu, X., Kim, J.K., Li, Y., Liu, F. and Chen, S. 2005. Tannic acid stimulates glucose transport and inhibits adipocyte differentiation in 3T3-L1 cells. Journal of Nutrition. 135, 165-171.

Miura, T., Ueda, N., Yamada, K., Fukushima, M., Ishida, T., Kaneko, T., Matsuyama, F. and Seino, Y. 2006. Antidiabetic effects of corosolic acid in KK-Ay diabetic mice. Biological and Pharmaceutical Bulletin. 29, 585-587.

Murakami, C., Myoga, K., Kasai, R., Ohtani, K., Kurokawa, T. and Ishibashi, S. 1993. Screening of plant constituents for effect on glucose transport activity in Ehrlich ascites tumour cells. Chemical and Pharmaceutical Bulletin (Tokyo). 41, 2129-2131.

Szkudelski, T. 2001. The mechanism of alloxan and streptozotocin action in β cells of the rat pancreas. Physiological Research. 50, 536-546.

Yamada, K., Hosokawa, M., Fujimoto, S., Fujiwara, H., Fujita, Y., Harada, N., Yamada, C., Fukushima, M., Ueda, N., Kaneko, T., Matsuyama, F., Yamada, Y., Seino, Y. and Inagaki, N. 2008. Effect of corosolic acid on gluconeogenesis in rat liver. Diabetes Research and Clinical Practice. 80, 48-55.