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Research Article

TOTAL FLAVONOID CONTENT ANALYSIS OF *LAGERSTROEMIA FLORIBUNDA* JACK (KEDAH BUNGOR)

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ABSTRACT

Till date there is no scientific data available regarding total flavonoid content analysis of *Lagerstroemia floribunda* Jack. The purpose of this study was to investigate the total flavonoid content of *Lagerstroemia floribunda* Jack. leaves and fruits. Extraction was carried out by using Soxhlet apparatus. The standard curve for total flavonoids was made using rutin standard. In results, the flavonoid content in methanolic leaves and fruits extract were found as 653.3 RE/g and 220 RE/g respectively. Based on the results, it can be concluded that *Lagerstroemia floribunda* leaves extract showed higher existence of flavonoid constituents than its fruits.

Keywords: Total flavonoid content, *Lagerstroemia floribunda* Jack, fruit, leaves

INTRODUCTION

Lagerstroemia floribunda, also known as Thai crape myrtle and kedah bungor, is a species of flowering plant in the Lythraceae family (Figure 1). This delightful ornamental plant is native to

subtropical and tropical South-East Asia, from southern China to Myanmar, Thailand, Cambodia, Indo-China and Peninsular Malaysia. It's the provincial tree of Saraburi Province in Thailand ¹⁻⁵.

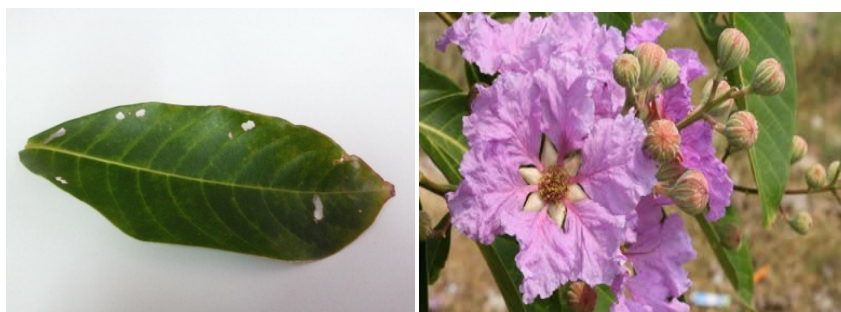


Figure 1: *Lagerstroemia floribunda* leaves and flower

Today, there are many diseases caused by oxidative stress. For examples, atherosclerosis, thrombosis, cardiovascular disease, cancer, asthma, Alzheimer's disease, angina, arrhythmia, anxiety, chronic obstructive pulmonary disease, systemic lupus erythematosus (SLE). Imbalance between the systemic manifestation of free radicals and the biological system's ability to detoxify the reactive oxygen species or to repair the damage cause the symptoms of oxidative stress to occur. Polyphenolic compounds, for example phenolic acid, tannins and also the flavonoid are commonly reported in the plants that exert multiple biological effects, including antioxidant activity. We have carried out this study on *Lagerstroemia floribunda* plant as there are flavonoids and polyphenolic compounds potentially found in this plant. By exploring out this study, we can further explore the antioxidant of this plant.

MATERIAL AND METHOD

Collection and preparation of plant material

Fresh leaves and fruits of *Lagerstroemia floribunda* were collected in and around the campus of AIMST University, Kedah, Malaysia in the month of October 2014. A voucher herbarium specimen was prepared and submitted to Unit of Pharmaceutical chemistry, Faculty of Pharmacy, AIMST University, Malaysia. The leaves and fruits were separated, and shade dried at the room temperature for seven days. Both the leaves and fruits also dried in the hot air oven to increase the speed of drying process. Then, the leaves and fruits were homogenized to fine powder by using the electronic blender. Both the fine powder of leaves and fruits were subsequently sieved to get the coarse powder and stored in the air tight container.

Extraction

Soxhlet apparatus was used for the extraction. It is the hot continuous extraction. In this method, the finely ground crude drug of leaves and fruits were placed in the different porous bags or 'thimble' made of strong filter paper of the Soxhlet apparatus. The 'thimble' was closed with the filter paper. 3 to 4 boiling chips were placed into the solvent vessels, such as round bottom flask or cylindrical flask. The Soxhlet extractor connection was placed on the top of the flasks. The 'thimble' was inserted into the Soxhlet extractor fitting into the thimble compartment. The condenser was connected on the top of the Soxhlet extractor and the water flow was turned on. 250 ml of methanol was added from the top of the Soxhlet apparatus which passed through the 'thimble'. The extracting solvent in the flask was heated and the heat was adjusted to monitor the regular flushing. The Soxhlet apparatus was switched on and maintained at the suitable temperature for about 2 to 3 days continuously. After the continuous extraction, the Soxhlet apparatus was switched off and the content in the round bottom flask was weighed. The extract obtained was concentrated by using rotary evaporator⁶.

Total Flavonoid Content

Rutin stock solution was made by dissolving 10mg of rutin in 10 ml of ethanol. The concentration of the stock solution is 1000

µg/ml. From the stock solution, prepared different dilutions of standard rutin like 10, 20, 40, 60, 80, and 100 µg/ml. Then, 1ml of the standard rutin with different concentration, 0.7 ml of 5 % of NaNO₃ and 10 ml of 30 % (v/v) ethanol were mixed and allowed to stand for 5 minutes. After that, 0.7 ml of 10 % AlCl₃ was added and mixed. After 6 minutes, 5 ml of 1 mol/L NaOH was added into the solution. The resulting solution was diluted to 25 ml with 30 % (v/v) ethanol and allowed to stand for 10 minutes. The absorbance was measured at 430 nm by using spectrophotometer. The stock solution of extract for both the leaves and fruits were prepared by dissolving 10 mg of extract in 10 ml of methanol. The concentration of the stock solution was 1000 µg/ml. Different dilutions of extract were made from the stock solution such as 50, 100 and 200 µg/ml. The same procedures as the standard rutin were followed by the extracts. The graph for standard rutin and the extracts were plotted. Total flavonoid content was expressed by using the following formula:⁷

Total Flavonoid Content, $C = A/B$

A- Expressed as mg RE/g dry weight of extract

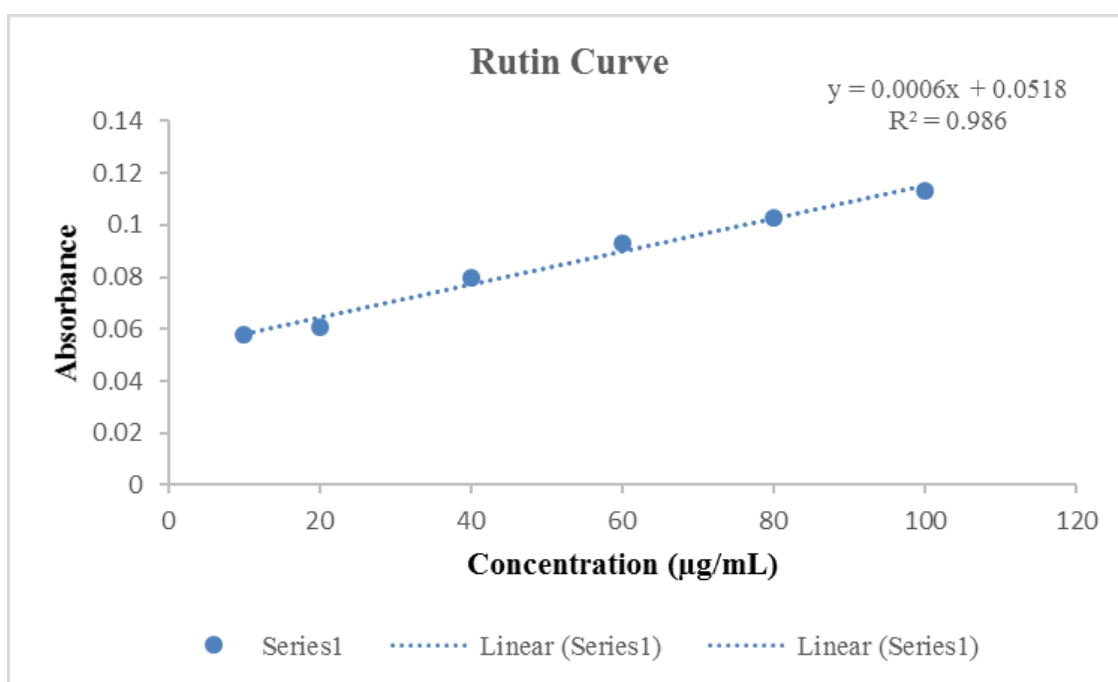
A- Equivalent concentration of rutin established from calibration curve

B- Dry weight of extract

RESULTS

Table 1: Concentration and corresponding absorbance of rutin

Concentration (µg/mL)	Absorbance
10	0.058
20	0.061
40	0.080
60	0.093
80	0.103
100	0.113



Graph 1: Graph of standard rutin

Table 2: Concentration and corresponding absorbance of the leaves extract

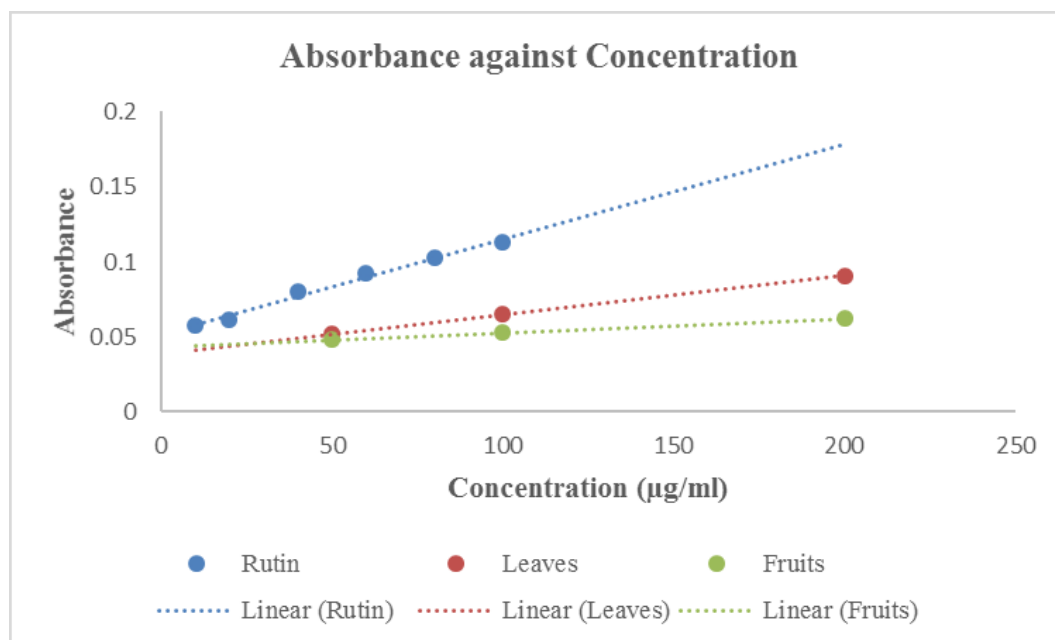
Concentration (µg/mL)	Absorbance
50	0.052
100	0.065
200	0.091

Table 3: Concentration and corresponding absorbance of the fruits extract

Concentration (µg/mL)	Absorbance
50	0.048
100	0.065
200	0.062

Total flavonoid content, $C = A/B$
 C-expressed as mg RE/g dry weight of extract
 A-Equivalent concentration of rutin established from calibration curve (mg)
 B-Dry weight of extract
 $y = 0.0006x + 0.0518$ (y = leaves extract absorbance reading)
 $0.091 = 0.0006x + 0.0518$
 $x = 65.33 \mu\text{g/mL}$
 $= 0.0653 \text{ mg/mL}$
 $A = 0.0653 \text{ mg/mL RE}$
 $B = 0.1 \text{ mg of the extract (concentration: } 0.1 \text{ mg/mL)}$
 $C = A/B$
 $= 0.0653 \text{ mg/mL RE} / (0.1/1000)$
 $= 653.3 \text{ RE/g}$

Total flavonoid content, $C = A/B$
 C-expressed as mg RE/g dry weight of extract
 A-Equivalent concentration of rutin established from calibration curve (mg)
 B-Dry weight of extract
 $y = 0.0006x + 0.0518$ (y = fruits extract absorbance reading)
 $0.065 = 0.0006x + 0.0518$
 $x = 22 \mu\text{g/mL}$
 $= 0.022 \text{ mg/mL of flavonoid content which is equivalent to the rutin in the extract}$
 $A = 0.022 \text{ mg/mL RE}$
 $B = 0.1 \text{ mg of the extract (concentration: } 0.1 \text{ mg/mL)}$
 $C = A/B$
 $= 0.022 \text{ mg/mL RE} / (0.1/1000)$
 $= 220 \text{ RE/g}$



Graph 2: Graph of absorbance against concentration for leaves and fruits extract of *Lagerstroemia floribunda*

DISCUSSION

In total flavonoid content determination, the absorbance of sample and rutin reagent mixture was observed. The standard compound was rutin, and total flavonoid content was expressed as µg/mL. Rutin equivalent is obtained by using the standard curve equation: $y = 0.0006x + 0.0518$, $R^2 = 0.986$, where y is the absorbance at 430nm, and x is the total flavonoid content expressed in µg/mL. The flavonoid content in methanolic leaves and fruits extract were found as 653.3 RE/g and 220 RE/g respectively.

CONCLUSION

Lagerstroemia floribunda plant was selected for this study. The method used for extraction of *Lagerstroemia floribunda* was Soxhlet extraction method. All the standardization parameters for the leaves and fruits were according to pharmacopoeia standards. The flavonoid content in the plant gives the antioxidant activity features, acting as free radical scavengers, hydrogen donating sources, singlet oxygen quenchers, or metal ion chelators. *Lagerstroemia floribunda* leaves extract showed higher existence of flavonoid constituents than fruits.

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