

## Potential Of *Garcia Mangostana* (Mangosteen) Leaves As An Antibacterial Agent

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### ABSTRACT

This study was to detect the heavy metal and xanthenes contain in mangosteen leaves. The heavy metal that being detect were lead (Pb), nickel (Ni), iron (Fe) and copper (Cu). The leaves were taken from top, middle and bottom of the tree. For xanthenes, the leaves was extracted using methanol in soxhlet extraction process. For detection of heavy metal, Atomic Absorption Spectroscopy (AAS). Fe was the highest concentration which are 1.153 mg·L<sup>-1</sup> at the top, 1.493 mg·L<sup>-1</sup> at the middle and 1.763 mg·L<sup>-1</sup> at the bottom because Fe was the most abundant metal in earth's crust and the most important heavy metal in plant growth. Pb shows the lowest concentration which are 0.028 mg·L<sup>-1</sup> at the top, 0.040 mg·L<sup>-1</sup> at the middle and 0.078 mg·L<sup>-1</sup> at the bottom which showed that the soil is not contaminated by Pb. For the detection of concentration of xanthenes, Ultraviolet-Visible spectroscopy (UV-Vis) was used. From the UV-Vis results, there were xanthenes peak observed in the range of 306 nm to 310 nm and 229 nm to 240 nm. This confirmed that mangosteen leaves contain xanthenes. The concentration of xanthenes were 0.2267 mol·L<sup>-1</sup>, 0.2274 mol·L<sup>-1</sup> and 0.2296 mol·L<sup>-1</sup>. The average zones of antibacterial activities for mangosteen leaves extracted were 0.944 mm for bacteria *E. coli*, *B. subtilis*, *S. aureus* and *P. aeruginosa*. From the analysis result showed that *Garcia Mangostana* leaves have a potential act as an anti-bacterial agent.

KEYWORDS: *Garcia Mangostana* leaves, heavy metal, xanthenes, anti-bacterial

## 1 INTRODUCTION

Heavy metal is a group of inorganic chemical, such as Pb, Cr, As, Zn, Cd, Cu, Hg, Fe and Ni. These heavy metal are contain in the soil [1]. The presence of heavy metal in the soil can severely inhibit the biodegradation of organic contamination. Heavy metal may give a risk and hazards to human and other ecosystem. The ecosystem can make a contact with heavy metal through drinking water, direct contact with contaminate soil and ground water. Heavy metal also gave an effect on plant growth, ground cover and have a negative impact on soil. It can be only physically removed or be transformed into nontoxic compounds. Some of these metals are necessary for plant growth, such as Zn, Cu, Mn, Ni, and Co, while others have unknown biological function, such as Cd, Pb, and Hg[2].

Xanthenes is believed to have anti-cancer effects, anti-inflammatory, anti-viral and cardiovascular protection with the antioxidant effects [3]. Xanthenes could be found on the skin of fruit, fruit, bark and leaves of mangosteen. Xanthenes have a unique chemical structure composed of a tricyclic aromatic system (C<sub>6</sub>-C<sub>3</sub>-C<sub>6</sub>). Isoprene, methoxyl and hydroxyl groups located at various locations on the A and B rings, resulting in a diverse array of xanthenes compounds. Xanthone most researched are alpha, beta and gamma mangostin, garcinone E, 8-deoksigartanin and gartanin. Fig. 1 shows the most abundant structure of α mangostin, β mangostin and γ mangostin in mangosteen leaves [4].

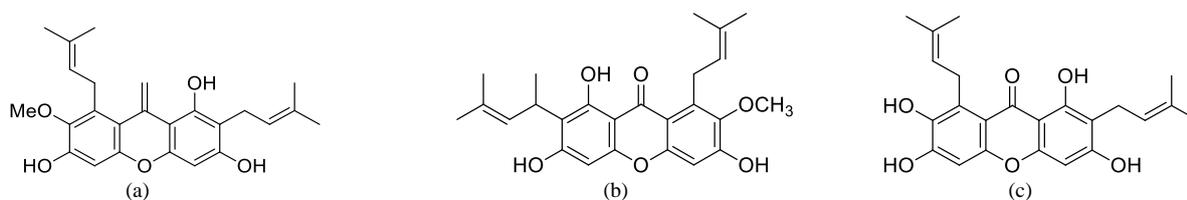


Fig. 1 Chemical structure of (a)  $\alpha$ -mangostin; (b)  $\beta$ -mangostin and (c)  $\gamma$ -mangostin

Previous studies reported  $\alpha$ -mangostin isolated from the extract of dried *G. mangostana* rind showed antioxidant, anticancer, and cytotoxicity activities [5].  $\alpha$ -mangostin is one of the earliest naturally occurring xanthone isolated from mangosteen.  $\alpha$ -mangostin also has potential for anti-obesity and treatment for Alzheimer's disease.  $\alpha$ -mangostin, 1-isomangostin and mangostin triacetate exhibited anti-inflammatory activity in a rat model. Besides, the  $\gamma$ -mangostin is widely studied in pharmaceutical property [6].

## 2 OBJECTIVE

The purpose of this study is to detect heavy metal and xanthenes in *Garcinia Mangostana* leaves. The objectives of this study are; to extract *Garcinia Mangostana* leaves using soxlet extraction; to determine the amount of heavy metal in *Garcinia Mangostana* leaves using atomic absorption spectroscopy; to determine the concentration of xanthenes in mangosteen leaves using UV-Visible spectroscopy; to study the antibacterial activities agent of the xanthenes toward bacteria.

## 3 SIGNIFICANCE (S)

In mangosteen leaves there is a presence of heavy metals. The heavy metal that consists in the mangosteen are Pb, Cr, As, Zn, Cd, Cu, Hg, Fe and Ni [7]. Atomic Absorption Spectroscopy (AAS) was used to detect the amount of heavy metal. Therefore, AAS lamp of Fe, Cu, Ni, Pb was used to analyse the heavy metals such as Fe, Cu, Ni, Pb in mangosteen leaves. Xanthenes were found in mangosteen leaves [3] and has a properties of atomic electron transition where it can be detect by UV-Vis. Xanthenes will show two peak at the range of 223 nm to 244 nm and 280 nm to 316 nm [8]. By using Beer's Law formula  $A = \epsilon bc$ , the concentration of the xanthenes was determined.

## 4 METHODOLOGY/ TECHNIQUE (S)

### 4.1 Raw materials, Chemicals and Instruments

Mangosteen leaves was taken from Bandar Seri Jempol, Jempol, Negeri Sembilan. Hydrochloric acid 98% and metanol 80% were used. Instruments used were PG instrument limited, T80/80+ double beam Ultraviolet-Visible (UV-Vis) spectrophotometer and Perkin Elmer A Analyst 700 Flame Atomic Absorption Spectroscopy (FAAS) with WinLab 32 AA software.

### 4.2 Method

**Sample preparation:** The mangosteen leaves was cleaned and washed from any residual compost. It was cut into a small pieces ( $0.5 \times 1.0 \text{ cm}^2$ ) and dried overnight in a tray dryer at  $45^\circ \text{C}$ . After that, the leaves was grinded using grinder (around 18 meshes) [9]. 15 g of the samples was put into the soxhlet extractor. 150 ml of 98% (v/v) methanol was added into the round bottom flask and then it was heated for 7 hours. The extract is stored in desiccator for 24 hours [10].

**Determination of Heavy Metal in leaves by using AAS:** 98% of hydrochloric acid was diluted to 10% (v/v) and 15% (v/v). 8 g of the samples was put in a crucible and partially cover then put in the furnace at  $450^\circ \text{C}$  for 12 hours. The sample was dissolved in 10% (v/v) hydrochloric acid and then put 1% (v/v) hydrochloric acid. Analysis of heavy metal such as Fe, Cu, Pb and Ni was done by using AAS [11].

**Detection of xanthenes in the mangosteen leaves using UV-Vis:** 80% methanol was added to the sample. A solvent mixture-to crude extracted mass ratio (10, 30 and 50) and methanol mass ratio was used. The absorbance of the sample was explored using UV-Vis spectroscopy [10].

### Antibacterial activity agent of xanthenes

**Preparation of nutrient agar (NA):** 23 g of NA powder was dissolved in 1 L of distilled water. The mixture was heated and stirred until it was boil and the NA powder was completely dissolved. Then the mixture was autoclave for 15 minutes at  $121^\circ \text{C}$ . The mixture was cooled in a laminar flow and then poured into sterile petri dishes. The

sample crude was dissolved with dimethyl sulfoxide (DMSO). The concentration of the sample used was 100 mg·L<sup>-1</sup>.

**Disc diffusion method:** The sub cultured were spread into NA petri dishes by using sterile cotton swab. The sub cultured that was used were *E. coli*, *B. subtilis*, *A. aureus* and *P. aeruginosa*. 6 mm diameter of Whatmann filter paper was soaked into the sample. The filter paper was then placed on the agar surface [12]. Sample was added into 1 petri dishes. The petri dishes were put into an incubator for 24 hours at 37 °C. The clear inhibition zone around the filter paper was the presence of the antibacterial activity. The zone of the antibacterial activities was measured. The activity were performed in triplicate [12].

## 5 RESULT

### 5.1 Concentration of heavy metal in mangosteen leaves.

The highest concentration of the heavy metal was for all part of mangosteen tree was Fe as shown in Table 2. The average concentration of Fe is 1.153±0.003 mg·L<sup>-1</sup> at top, at the middle part is 1.493±0.003 mg·L<sup>-1</sup> and for the bottom part is 1.763±0.008 mg·L<sup>-1</sup>. The amount of Fe uptake by the mangosteen is the highest because Fe is the most abundant metal in earth's crust [13] and most important heavy metal in plant growth [14].

The lowest heavy metal content in the mangosteen leaves is Pb which 0.028 mg·L<sup>-1</sup> at top, 0.040 mg·L<sup>-1</sup> at middle and 0.078 mg·L<sup>-1</sup> at the bottom part of the mangosteen tree. This is because, the soil is not contaminated. Amount of Pb increased when the amount of contamination in soil is increase [15]. Traunfeld and Clement [16] stated that industrial sites are high in Pb. The contaminated area contain Pb at least 400 mg·L<sup>-1</sup> in the soil and 22 mg·L<sup>-1</sup> in the plants [17].

**Table 2** The average concentration of heavy metal in the mangosteen

Sample	Heavy metal (mg·L <sup>-1</sup> )			
	Cu	Fe	Pb	Ni
Top	0.166±0.001	1.153±0.003	0.028±0.057	0.132±0.006
Middle	0.217±0.000	1.493±0.003	0.040±0.070	0.160±0.003
Bottom	0.228±0.002	1.763±0.008	0.078±0.032	0.166±0.008

### 5.2 UV-Visible spectroscopy

All the sample have shown the present of two peak at range 306 nm to 310 nm and at range 229 nm to 240 nm. Xanthenes will show two peak at range 223 nm to 244 nm and at range 280 nm to 316 nm [8]. As comparison, the UV-Vis spectrum shows almost same as peak range from others researcher [8]. This was confirmed that mangosteen leaves contain xanthenes.

### 5.3 Concentration of Xanthenes

The concentration of xanthenes can be determine by using Beer's Law formula  $A=\epsilon bc$  where A is absorbance,  $\epsilon$  is molar absorptivity, b is path length and c is concentration. Fig. 4.12 shows the absorbance of xanthenes in mangosteen leaves. The concentration of xanthenes that have been calculated is shown in the Table 3. The molar absorptivity used in the calculation of xanthone was  $13.5 \times 10^3 \text{ L}\cdot\text{mol}^{-1}\cdot\text{cm}^{-1}$  and the path length is 1 cm [18].

**Table 3** The concentration of xanthone from the concentration of mangosteen leaves.

Concentration of mangosteen leaves (mg·L <sup>-1</sup> )	Concentration of xanthone (mol·L <sup>-1</sup> )
10	0.02267
30	0.02274
50	0.02296

### 5.4 Xanthenes as antibacterial agent.

Based on the Table 4, the extracted sample had shown antibacterial activities toward the bacteria of *E. coli*, *B. subtilis*, *P. aeruginosa*, and *S. aureus*. The average inhibition zones of antibacterial activities of sample is 0.944 mm. At molarity of 0.0002548 mol·L<sup>-1</sup> sample shows positive antibacterial activity towards *E. coli*, *B. subtilis*, *S. aureus* and *P. aeruginosa* [19]. Based on this result and the statement, it can be concluded that the concentration of xanthenes in the mangosteen leaves can act as antibacterial for *E. coli*, *B. subtilis*, *S. aureus* and *P. aeruginosa*.

**Table 4** The concentration of xanthone from the concentration of mangosteen leaves.

Type of Bacteria	<i>E. coli</i>	<i>B. subtilis</i>	<i>S. aureus</i>	<i>P. aeruginosa</i>
Zones of xanthenes antibacterial activity (mm)	0.860	1.033	1.033	0.850

## 6 CONCLUSION

The concentration of Fe in the leaves is the highest at the top part of the tree is 1.153 mg·L<sup>-1</sup>, at the middle part is 1.493 mg·L<sup>-1</sup> and bottom part is 1.763 mg·L<sup>-1</sup>. In the UV-Vis spectroscopy, the two peak of xanthone was obtained at the range of 306 nm to 310 nm and at range 229 nm to 240 nm. Xanthenes were obtained almost the same for all concentrations which are 0.02267 mg·L<sup>-1</sup> at concentration 10 mg·L<sup>-1</sup> of sample, 0.02274 mg·L<sup>-1</sup> at concentration 30 mg·L<sup>-1</sup> of sample and lastly 0.02296 mg·L<sup>-1</sup> at the 30 mg·L<sup>-1</sup> of sample. The zones of antibacterial activities for mangosteen leaves extract were 0.860 mm for *E. coli*, 1.033 mm for *B. subtilis*, 1.033 mm for *P. aeruginosa* and 0.850 mm for *S. aureus*. This concludes that the concentration of xanthenes in the mangosteen leaves can act as antibacterial.

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