

# Microstructural Characterization of the Garcinia mangostana Fruit at Different Age Level

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**Submission date:** 15-Jun-2021 07:49PM (UTC+0700)

**Submission ID:** 1606904632

**File name:** 20936-FirstProof.pdf (2.79M)

**Word count:** 3300

**Character count:** 17111



# Microstructural Characterization of the *Garcinia mangostana* Fruit at Different Age Level

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## Abstract

Mangosteen (*Garcinia mangostana*) is one of the most popular fruits which has been widely used medicinally. The major constituents are mostly found in the pericarp particularly, xanthones, which are tricyclic isoprenylated polyphenols. Xanthones have been reported to for its anti-oxidant, anti-inflammatory, anti-bacterial, anti-proliferative, proapoptotic and anti-carcinogenic activities. In this study, the mangosteen pericarps in various age levels were investigated with electron microscopy and X-ray spectroscopy. The results revealed the mangosteen at age level 3 showed the highest concentration of xanthone while those younger and older mangosteens exhibited lower xanthone content. The mangosteen pericarp with highest xanthone concentration showed crystalline structure and contained small amount of silver and gold particles which was analyzed with EDX measurement and confirmed by the XRD spectrum.

**Keywords:** Mangosteen, Particle Size, SEM, Xanthone, X-ray Diffraction, XRD

## 1. Introduction

Plants and their fruits have been used since ancient time as supplement and medicine to treat diseases. Approximately two-third of world population uses plant based products as their main medicine<sup>1</sup>. *Garcinia mangostana* L. (Clusiaceae), commonly known as mangosteen, have been used as a traditional medicine in Southeastern Asia for the treatment of diarrhea, dysentery, inflammation, ulcers and wound healing<sup>2,3</sup>. Mangosteen is cultivated mainly in Indonesia, Malaysia, Philipina and Thailand<sup>4-6</sup>. Medicinal properties of *G. mangostana* is associated with its bioactive compounds, xanthone. Xanthone is a tricyclic isoprenylated polyphenol. Previous investigation shows







that xanthone is antioxidant<sup>5,7</sup>, anti-bacterial, anti-proliferative, proapoptotic, anti-carcinogen<sup>8-10</sup> and anti-inflammatory<sup>11</sup>.

Xanthone can be found in every part of mangosteen, mainly in fruit skin. Mangosteen contains 17% of pericarp<sup>12</sup>, which contains 62.05% of water, 0.63% of fat, 0.71% of protein, 1.17% of total sugar and 35.61% of carbohydrate<sup>13</sup>. Xanthone content in the mangosteen pericarp is about 108 to 124 mg per 100 g<sup>14,15</sup>. The ripeness level significantly affects the xanthone content of mangosteen<sup>16</sup>. In this study, we have investigated the physical and chemical properties of mangosteen pericarps depending on the age level in accordance to Palapol, et al. (2009)<sup>17</sup> (Table 1), using various characterization techniques such as PSA (Particle Size Analyzer), Scanning Electron

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Microscope (SEM), X-ray Diffraction (XRD), and X-ray Fluorescence (XRF). The study aims to get insight into the microstructure of mangosteen pericarp in accordance to their age level and xanthone content. Furthermore, it can give insight to maximize the yield of xanthones.

**Table 1.** Age level classification of mangosteen

Age Level	Properties
 Level 1	Green yellowish, high mucus content. The inner part of fruit cannot be separated from the flesh.
 Level 2	Yellow reddish with distribution of red colored fleck. Lower mucus content. The inner part of the fruit still cannot be separated from flesh.
 Level 3	Red brownish. Mucus still exists. The inner part of fruit can be separated from the flesh.
 Level 4	The color becomes red purple. The mucus is lesser. The inner part of fruit can be separated from the flesh and the fruit can be consumed.
 Level 5	Purple reddish. Fruit starts to ripe. The mucus disappears, and the fruit can be easily separated.
 Level 6	Purple blackish. The fruit is riped and can be eaten immediately.

## 2. Methods

### 2.1 Extraction

The extraction of mangosteen was performed following standard procedure. Mangosteen pericarp was obtained from Bandung, West Java. Mangosteen was air dried until the water content was about 5% prior to crushing in an electrical blender. 700 g of mangosteen pericarp mass was macerated using 96% ethanol (1 : 5)

for 5 days and stirred every 3 hours. The extract was dried by evaporation and freeze dried to obtain the powder. The powder was stored in desiccators until used.

### 2.2 Characterization

The mangosteen extract in various age levels were subjected to microstructure characterization using PSA, SEM, XRD and XRF. For SEM analysis, the mangosteen extract was poured homogenously onto the carbon double tip on the SEM sample holder stub. For the elemental composition in SEM, an energy dispersive X-ray (EDX) detector attached on SEM (SEM JEOL JSEM2650LV) was used. For XRD analysis Imperian with Cu source was used. The XRD results were analyzed using Ritveld Refinement to derive information about the phase and crystals may occur.

## 3. Results

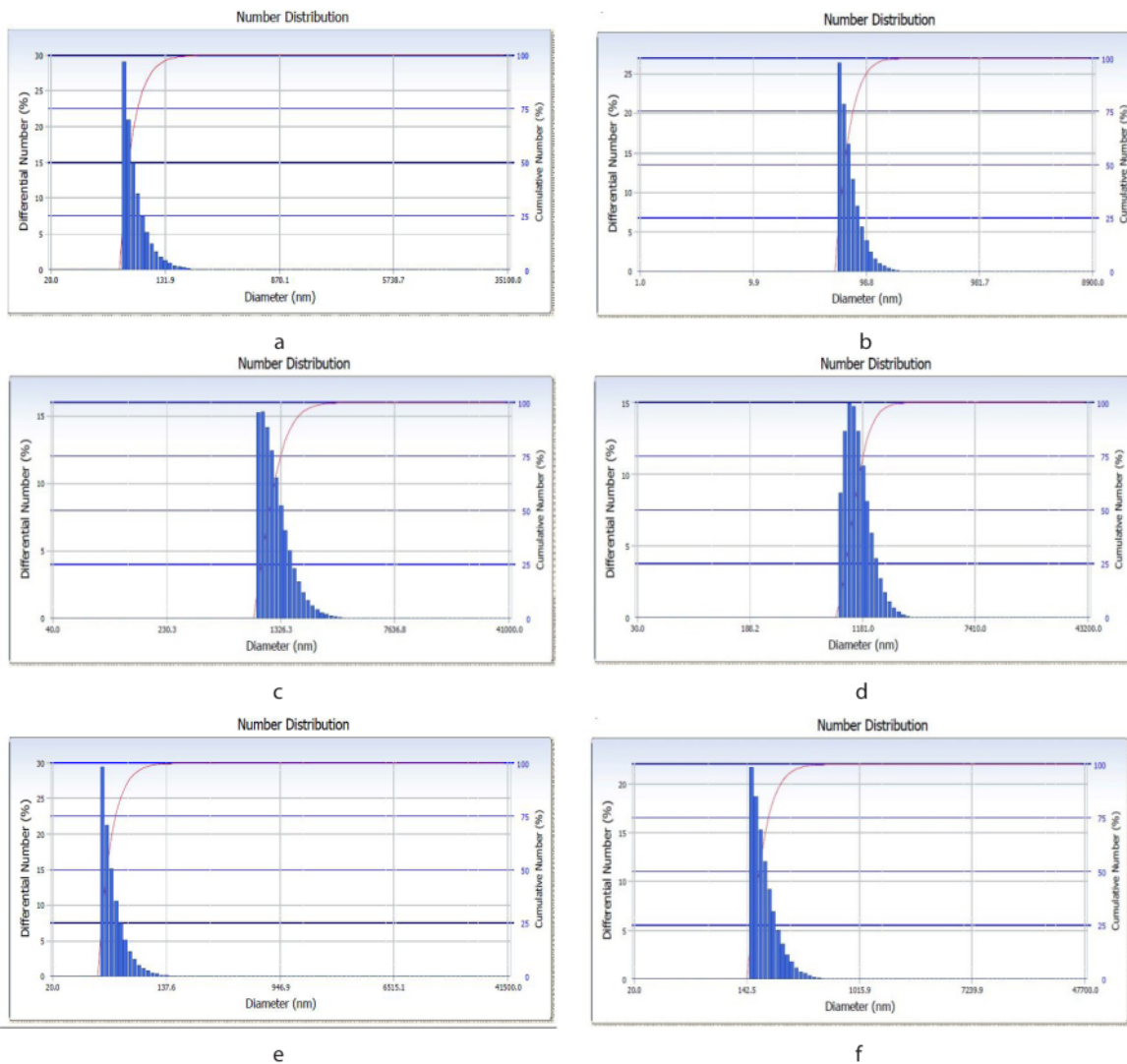
PSA measurement of the mangosteen extract in various age levels is presented in Figure 1 and Table 2. Particle of mangosteen powder at level 5 exhibited the smallest size, whereas those at level 3 exhibited the highest size.

**Table 2.** Particle size of mangosteen pericarp extract as measured by PSA

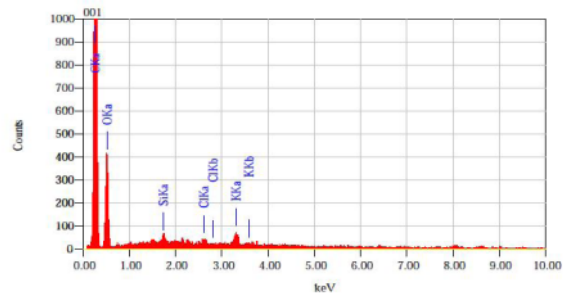
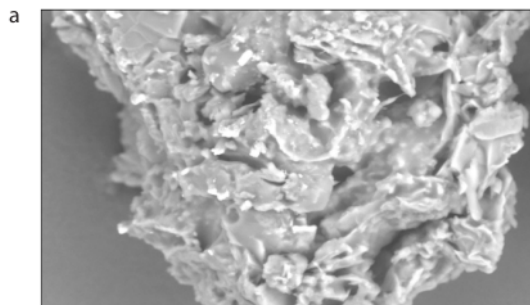
Sample	Size (nm)
1	81.8 ± 21.1
2	72.6 ± 20.4
3	1244.1 ± 341.0
4	1112.2 ± 256.5
5	57.0 ± 14.5
6	199.2 ± 58.9

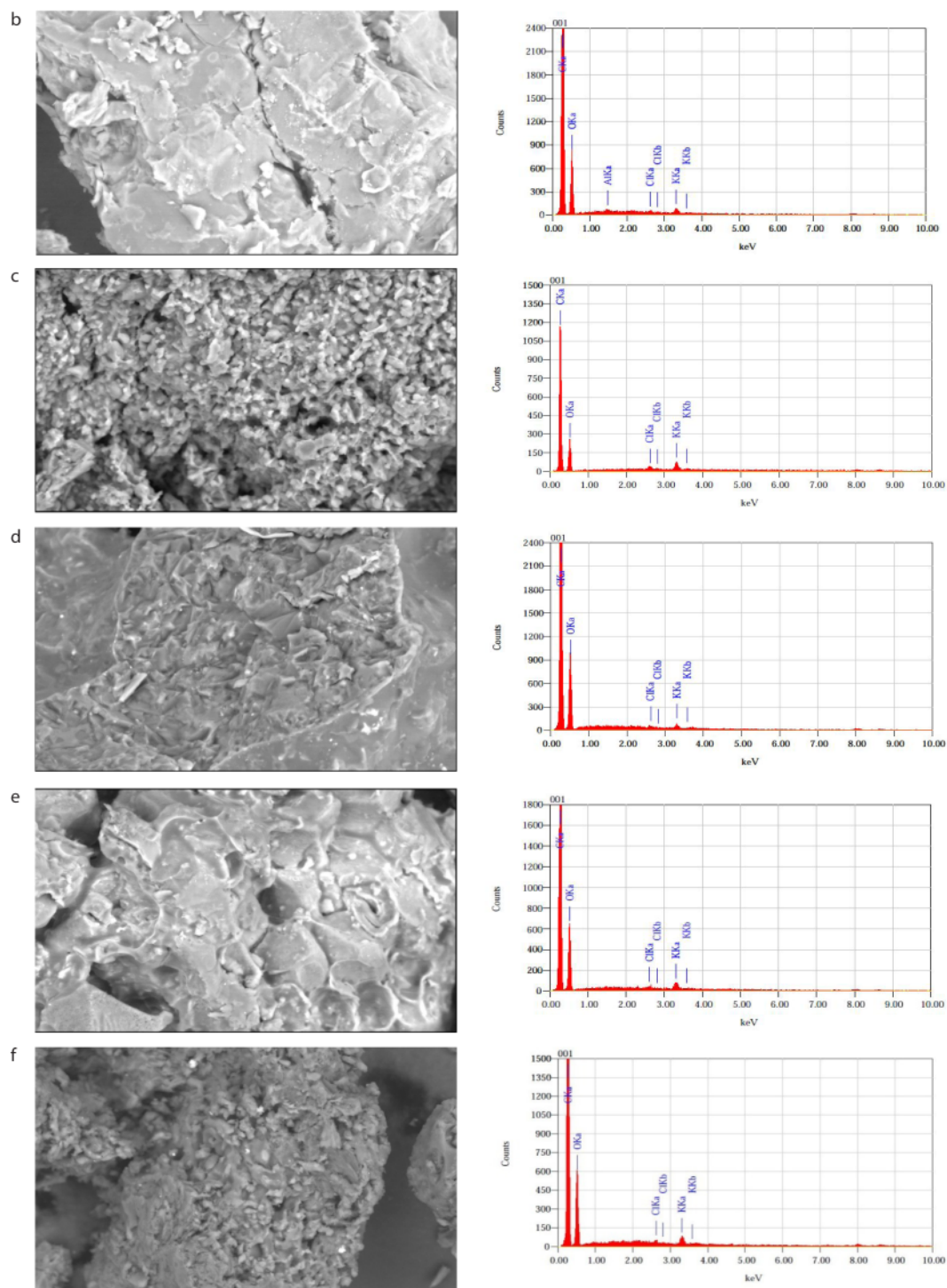
Data are presented in Mean ± Standard Deviation

Figure 2 shows the SEM image of the microstructure of mangosteen pericarp extract at the age levels 1-6 which generally showed granular and plate like structure composing of large coagulates. Sample of age levels 3 and 6 have shown clearly smaller powder grains than those of the sample of age levels 1, 2, 4 and 5. It was distinguished by the contrast deficiency by the sample with fine grain size. It is almost impossible to measure single grain on the sample 1, 2, 4 and 5 because the sample forms large block of particle agglomerations.



**Figure 1.** Particle size distribution of, a. Sample 1; b. Sample 2; c. Sample 3; d. Sample 4; e. Sample 5; f. Sample 6.



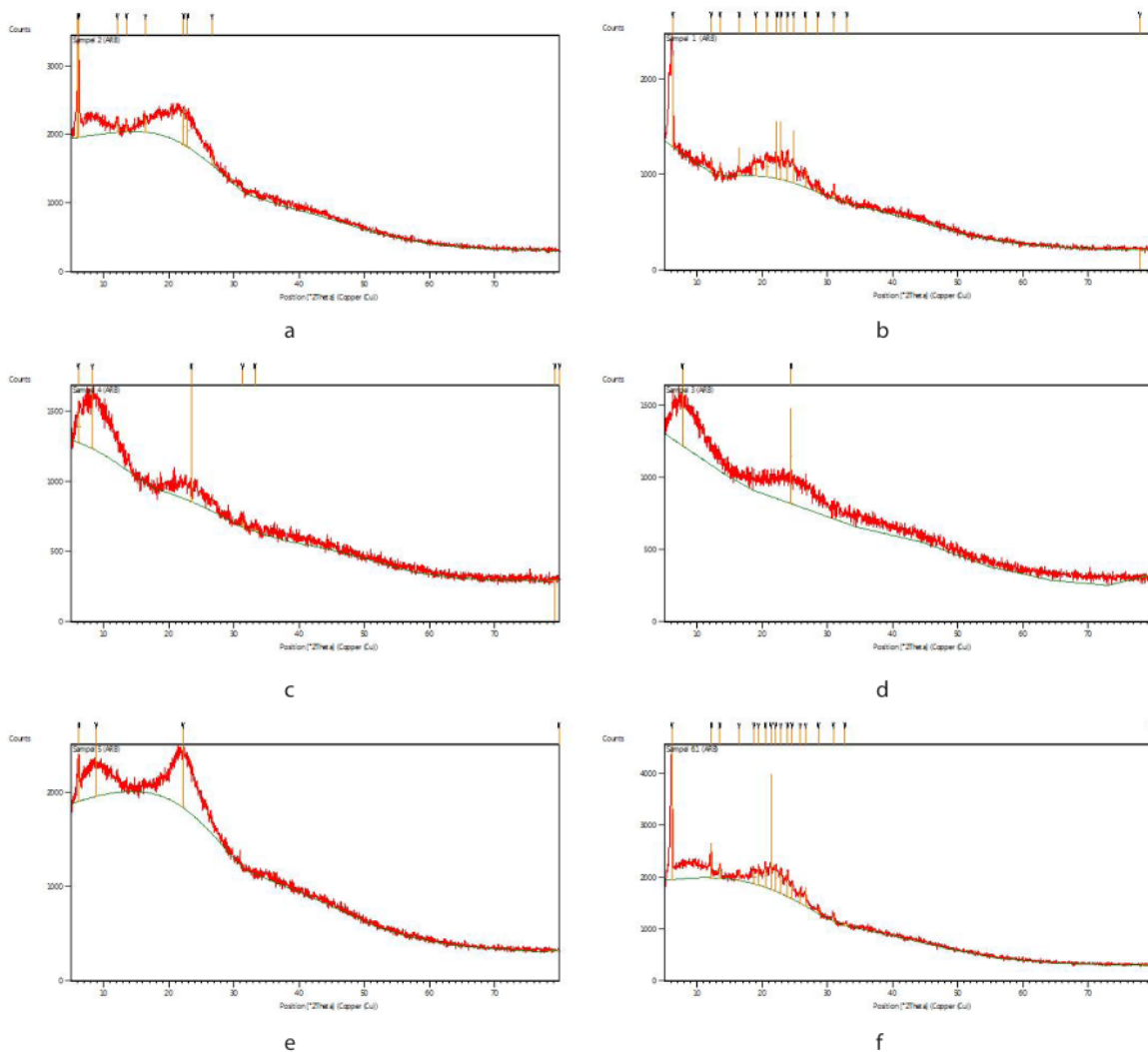


**Figure 2.** SEM SE images of mangosteen pericarp samples at age level and their corresponding EDX spectrum, a. Sample 1; b. Sample 2; c. Sample 3; d. Sample 4; e. Sample 5; f. Sample 6.



The corresponding EDX spectrums are also represented in Figure 2 on the right side revealing the elemental composition of the samples. Mainly the samples contain C, O, K and Cl. Sample 1 revealed the elemental content Si, Cl and K about 0.33%, 0.22% and 0.72% respectively. Sample 2 was crystalline in nature containing Al 0.1%, whilst Si was not detected. Level of Cl and K were significantly reduced. Sample 3 showed globular form, softer than samples 1 and 2 which was about 5  $\mu\text{m}$ , as shown in EDX results. Both Cl and K content was significantly reduced, amorphous and its silicone was reduced as well. Sample 4 was seen as

granules and also in combined form. Sample 4 has granule size of 20  $\mu\text{m}$ , crystalline and its level of Si was reduced. The content of Cl was 0.10% and K was 0.28%. Sample 5 showed granules form and also combined form. Its granule were between 5-50  $\mu\text{m}$ , crystalline and Si content was reduced. Sample 5 were mainly composed by C, O, Cl and K with 61,18%, 37,49%; 0,19% and 0,69% respectively. Sample 6 showed granules form and also combined form. Granule was 10  $\mu\text{m}$ , crystalline and Si content was reduced. Main components of sample 5 were C, O, Cl, and K with 62,41%, 36,67%; 0,23% and 0,68%. respectively.



**Figure 3.** XRD analysis of mangosteen peel extracts, a. Sample 1; b. Sample 2; c. Sample 3; d. Sample 4; e. Sample 5; f. Sample 6.

**Table 3.** Elemental concentration of mangosteen peel extract analyzed with X-ray fluorescence

Elements	Samples					
	1	2	3	4	5	6
K	50.96	50.59	53.58	50.21	51.22	51.03
Cl	13.45	11.18	12.83	9.61	11.90	12.20
Ca	4.92	8.10	1.48	8.56	5.90	4.67
Px	1.78	1.68	1.67	1.50	1.39	1.75
Sx	1.01	1.25	2.06	1.25	1.01	2.20
Na	1.37	1.27	1.08	1.67	1.77	1.22
cd	1.50	0.77	1.89	0.91	1.18	1.11
Ag	1.33	2.01	3.20	1.31	1.52	–
Fe	0.924	0.836	0.821	1.05	1.986	–
Zn	0.879	0.386	0.257	0.482	0.712	–
Mo	0.712	–	0.854	0.678	–	0.575
Rb	0.75	0.873	0.88	0.985	1.18	0.523
Mg	0.410	0.425	0.290	0.392	0.315	0.331
Si	0.295	0.294	0.249	0.280	0.569	0.401
Zr	0.239	0.209	0.125	0.219	0.167	0.249
Ar	0.291	0.291 / 0.254	0.339	0.246	0.203	0.203
Mn	0.218	0.218	0.156	0.453	0.195	0.293
Ti	0.115	0.115	–	–	0.089	0.504
Ba	–	–	0.80	–	0.84	–
Au	–	–	0.74	–	–	–

The XRD patterns of mangosteen pericarp extracts are shown in Figure 3. As shown in XRD analysis, samples 1, 2 and 3 were amorphous. Although sample 2 was previously seen as crystalline, it was mainly amorphous based on XRD results. Lowest peak in samples 4, 5 and 6 indicates carbon crystal.

XRF is X-ray method similar to EDX, yet it measures smaller element above 11 atomic number. XRF was performed to confirm results obtained from EDX and XRD analysis. Results of XRF are shown in Table 3. Most of elements were in accordance to previous results analyzed with EDX and there were also several more elements found. Elements K, Cl, Ca, Px, Sx, Na, Cd, Rb, Mg, Si, Zr, Ar and Mn were detected in all samples. Meanwhile, Ag, Fe, Zn, Mo and Ti was found only in sample 1. Similarly, Ag, Fe, Zn and Ti were detected in sample 2; Ag, Fe, Zn, Mo, Ba and Au were detected in sample 3; Ag, Fe, Zn and Mo were detected in sample 4; Ag, Fe, Zn, Ti and Ba were detected in sample 5 and Mo and Ti were detected in sample 6.

## 4. Discussion

Dehydration is an efficient alternative for fruit storage in which reduction of water is associated with the decline of chemical and enzymatic reactions that can damage foods. Gentle drying techniques, such as osmodehydration or lyophilisation (freeze drying), have been used as new alternative for the commercialization of tropical fruits to reduce the difficulties for its handling and transport and to acquire added value solids products<sup>18</sup>. In this study, we evaluated microstructure of freeze dried mangosteen pericarp extract at various age levels. The structural studies of the fruit powders were done using PSA (Particle Size Analyzer), Scanning Electron Microscope (SEM), X-ray Diffraction (XRD) and X-ray Fluorescence (XRF).

In the present study, particle size of the mangosteen extract was clearly heterogeneous. Particle of mangosteen powder at level 5 exhibited the smallest size, whereas those at level 3 exhibited the highest size. Particle size distribution does not follow a typical ideal Gauss bell

form. Additionally, it seems that the levels of age were not in correlation with the particle size. The particle size may depend on the processing method. This can be also originated from the coagulation of mangosteen particles.

SEM technique is a powerful tool for determining and observing the caking phenomenon on the powder surface. The corresponding EDX spectrums revealing the elemental composition of the samples, exhibited component C, O, K and Cl in samples. XRF is X-ray method similar to EDX, yet it measures smaller element above 11 atomic numbers. XRF was perform to confirm results obtained from EDX and XRD analysis. Most of elements were in accordance to previous results analyzed with EDX and there were also several more elements found.

Crystallization is essential for the stability of powdered juice and can be investigated through XRD analysis. Amorphous state is indicated with diffused and large peaks in which the molecules are disorderly displayed producing disperse bands, whereas crystalline materials yield sharp and defined peaks since they are presented in a highly ordered state<sup>18</sup>. In this study, samples 1, 2 and 3 were amorphous, whilst samples 4, 5 and 6 indicates carbon crystal.

Xanthones are flavonoid commonly found in mangosteen peel<sup>19</sup>. In the present study, mangosteen at age level 3 showed the highest concentration of xanthone while those younger and older mangosteen exhibited lower xanthone content. Xanthones have been reported to exhibit various medicinal properties including anti-microbial, anti-malarial, anti-oxidant and anti-inflammatory. It has been reported that scavenging activity of mangosteen is higher when the fruit were younger and its activity decrease as the fruit age increase<sup>16</sup>. It is suggested to be associated with decreased synthesis of antioxidants during the fruit ripening process resulting in oxidative stress. Chemical constituents and antioxidant capacity of fruit are affected by fruit growth stage in which immature fruit contains the highest antioxidant capacity<sup>20</sup>. Study done by Huang, et al. also found decreased antioxidant activity in the late phase of fruit enlargement which also explains correlation between increasing oxidative stress and metabolic changes with aging of citrus fruits<sup>21</sup>.

## 5. Conclusion

Mangosteen at age level 3 showed the highest concentration of xanthone while those younger and older mangosteen exhibited lower xanthone content. Mangosteen pericarp with highest xanthone concentration has crystalline structure and contains small amount of silver and gold particles. These findings provide information regarding characteristics of mangosteen peel that can be further used for the optimization of xanthone content.

## 6. Acknowledgment

This study was personally funded by authors. We would also like to thank Indonesian Institute of Life Science (LIPI) for its assistance and supervision.

## 7. Conflict of interest

Authors declare that there is no conflict of interest.

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