

MICROENCAPSULATION OF EXTRACTED *GARCINIA MANGOSTANA* (GM) PERICARP

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Abstract—*Garcinia Mangostana* (GM) fruit is one of the most popular fruit and known as a tropical fruit that has high nutrition value and widely cultivated in the tropical rainforest of some Southeast Asian nations. The abundance of mangosteen pericarps during fruit season have been collected and reviewed the contents of pericarp in order to investigate the beneficial antioxidants of mangosteen pericarp fruit by using different methods. The main objectives for this research are to identify the composition of extracted xanthenes in encapsulated *Garcinia Mangostana* (GM) pericarp and to determine the antioxidants activity, total phenolic content, moisture content and antimicrobial activity of encapsulated *Garcinia Mangostana* (GM) pericarp. The scope of research focuses on discovering and identifying the properties of xanthenes from *Garcinia Mangostana* (GM) pericarp which can produce microcapsule. The main method that involves in this research is solvent extraction by using soxhlet extraction and microencapsulation by using freeze drying. DPPH antioxidant activity, total phenolic content (TPC), moisture content and antimicrobial activity are the basic analysis that have been done to determine the properties of extracted xanthenes from *Garcinia Mangostana* (GM) pericarp. The amount of extracted xanthenes in form of liquid from 20 gram of *Garcinia Mangostana* (GM) pericarp is only 22 ml whereas all samples have been detected in the range of 515.0 to 4000 cm⁻¹ through Fourier transforms infrared spectroscopy (FTIR). The highest value of radical scavenging activity before the samples has been freeze dried is 82.69% whereas 88.01% for samples of powder. Furthermore, the total phenolic content (TPC) of xanthone is 5.240 mg GAE/g before freeze drying and 6.359 mg GAE/g after freeze drying. The highest moisture content for the powder of extracted xanthone from *Garcinia Mangostana* (GM) is achieved 7.02 %M. For the antimicrobial activity, most of samples only have single colony of microbes on agar plate after 72 hours incubated in 37 °C. In conclusion, extracted xanthenes from *Garcinia Mangostana* (GM) pericarp has higher content of antioxidant which functions as inhibitor to the free radical activities and benefit to the manufacturing food, cosmetic and also pharmaceutical.

Keywords— *Garcinia Mangostana* (GM), xanthenes, antioxidant, total phenolic content, moisture content, microencapsulation.

I. INTRODUCTION

The mangosteen fruit is a standout amongst the most famous fruit and known as a tropical natural fruit that has high nourishment value. *Garcinia Mangostana* (GM) is a scientific name gives to the mangosteen natural fruit. *Garcinia Mangostana* (GM) belongs to the family of Guttiferae and is named “the queen of fruits” which is broadly planted in the tropical rainforest of some

Southeast Asian nations like Indonesia, Malaysia, Sri Lanka, Philippines, and Thailand (J. Pedraza-Chaverri et al., 2008).

The abundance of *Garcinia Mangostana* (GM) pericarps during fruit season have been collected and reviewed the contents of pericarp in order to create awareness amongst the peoples. It is important to investigate the beneficial antioxidants of *Garcinia Mangostana* (GM) pericarp by using different methods which is 2, 2-diphenyl-1-picrylhydrazyl (DPPH) assay, Folin-Ciocalteu method and Fourier Transform Infrared Spectroscopy (FTIR) analysis. The studied of *Garcinia Mangostana* (GM) pericarps have found that pericarps consist of xanthone which is one types of antioxidants. The xanthone is also known as phenolic compound actually can extract from *Garcinia Mangostana* (GM) pericarp by using solvent extraction. The extracted xanthone can be formulate and produce high quality of antioxidant which is edible and can be supplement into food. It has been proofed that α -mangostin, a xanthone derivative of the fruit hull of *Garcinia Mangostana* (GM), possess protective effects in models where increased oxidative stress and antioxidant deficit are major players (J. Pedra-Chaverri et al., 2008).

Microencapsulation is a research of identifying the potential of extracted *Garcinia Mangostana* (GM) pericarp to be antioxidant. The main objectives for this research are; to identify the amount of extracted xanthone in encapsulated *Garcinia Mangostana* (GM) pericarp and to determine the antioxidants activity, total phenolic content, antimicrobial activity and moisture content of encapsulated *Garcinia Mangostana* (GM) powder. The scope of research focuses on discovering and identifying the amount of xanthone from *Garcinia Mangostana* (GM) pericarp which can produce microcapsule. The main raw material used to conduct the research is *Garcinia mangostana* (GM) pericarp. The abundance of *Garcinia Mangostana* pericarp will collect and the content of xanthone inside the pericarp being extract by using the solvent extraction. The main type of solvent that has been used in this extraction process is Methanol.

II. METHODOLOGY

A. Xanthone Extraction and Quantification

The analysis for phenolic compound called xanthone that was extracted from *Garcinia Mangostana* (GM) pericarp was conducted in order to ensure and identify the content of antioxidant. This analysis is the final section of extraction process to determine the functional group of the phenolic content and its composition. Analysis section on detection of xanthone and determination of the amount of xanthone that was extracted from the *Garcinia Mangostana* (GM) pericarp can be done using Fourier transform infrared spectroscopy (FTIR).

B. 2, 2-Diphenyl-1-Picrylhydrazyl 9 (DPPH) Free Radical Scavenging Assay

Free radical scavenger is utilizing the technique of 2-Diphenyl-

1-Picrylhydrazyl 9 (DPPH) with some adjustment. Exactly, 10 mg of 2-Diphenyl-1-Picrylhydrazyl 9 (DPPH) was weighed and then dissolved it in 100 ml of 80% methanol (0.1 mg/ml). Next, 1 ml of extracted sample was mixed with 4 ml of 80% methanol and 1 ml of 2-Diphenyl-1-Picrylhydrazyl 9 (DPPH) in bottle. The bottle of mixture was closed properly while it was shaken vigorously and kept in dark for 30 minutes at room temperature. The colour of 2-Diphenyl-1-Picrylhydrazyl 9 (DPPH) had been changed from purple to yellow when it was get reduced by the present of antioxidants. Then, the absorbance reading of mixture without any sample was measured as control which consist of 1 ml DPPH and 4 ml 80% methanol. This mixture also was kept in dark for 30 minutes at room temperature. The absorbance of 2-Diphenyl-1-Picrylhydrazyl 9 (DPPH) was determined at 517 nm against blank whereas 80% methanol is use as the main sample of blank in using UV-Visible Spectrophotometer (UV-Vis). A percentage of inhibitor was determined by applying the formula given as follows.

$$\% \text{ inhibition} = [(A_{\text{control}} - A_{\text{sample}})/A_{\text{control}}] \times 100\%$$

A = Absorbance

A_{control} = Absorbance of control without extract

A_{sample} = Absorbance of sample with extract

C. Total Phenolic Content (TPC)

Total phenolic content (TPC) was determined by using the Folin–Ciocalteu method. The assay had been carried out in 1 ml of the extract be transferred to the bottle or test tube to quantify the total phenolic concentration. Next, 500 μ l of Folin-Ciocalteu solution, 1 ml of the sodium carbonate solution and 10 ml distilled water are added. The absorbance was measured at 765 nm against blank (80% methanol) after standing at room temperature (30 °C) for 30 min by using ultraviolet-visible spectrophotometry (UV-Vis). Gallic acid was used as a standard with concentrations ranging from 0.0025 to 0.6 mg/mL. The total phenolic content is expressed as milligram Gallic acid equivalents per gram of pericarp powder. Total phenolic content was calculated as gallic acid equivalent (GAE) by following equation:

$$T = C \times V/M$$

T = total phenolic content (TPC)

C = concentration of gallic acid from the calibration curve (mg/ml)

V = volume of extract solution (ml)

M = weight of extract (g)

D. Moisture Content

The sample of *Garcinia Mangostana* (GM) pericarp which is in powder form had been placed on the pan so that heat was applied evenly to the sample during measurement. A sample pan that had been used in this analysis must be kept at room temperature and proceed to close the heater cover. The measurement of moisture analyzer was started automatically since heater cover was closed. Wait the moisture analyzer to complete the analysis moisture content about 10 minutes at 110 °C. The result of measurement had been checked when there is a short beep sounds indicated that means the measurement was completed. The measurement result had been showed in the display panel and the measurement completed indicator.

E. Antimicrobial Activity

The technique of streaking had been used in this experiment since it is only to observe the appearance of antimicrobial and identify the antimicrobial activity as well. The inoculation loop has picked up the samples which spread out over the surface of the agar and then the agar plate was sealed properly with parafilm. Incubate the agar plate that contained the samples for 72 hours at temperature 37 °C. The agar plate of sample is taken out from incubator and the number of colony that form on agar plate being

observed by using colony counter. Finally, the number of colony that formed on the agar plate was recorded.

F. Freeze Drying

The sample of xanthone that stored in freezer was depending on the purity of xanthone and the amount of xanthone to be dried by direct submersion in a low temperature bath, or by shell freezing at -20 °C. Immediately, the vacuum will be supply against the container of xanthone and the evaporative cooling that rely by laboratory assistant in order to maintain the lower temperature of xanthone. Basically, this technique can be apply when it only requires small volumes and xanthenes at high eutectic and collapse temperature. Since the vessels were attached to the manifold individually, each vial or flask has a direct path to the collector. Each flask or vial has a direct path to the collector and the competition for molecular space was created in system had been removed. The distance from the collector had been most ideally realized in a cylindrical drying chamber to xanthone vessel was same. Finally, the content of water molecules had been left the amount of xanthone in available vessels farthest from the collector. The process of drying for extracted xanthone which in liquid form using freeze dryer had been completed in 48 hours.

III. RESULTS AND DISCUSSION

A. Sample extraction using soxhlet extraction

Garcinia Mangostana (GM) pericarp has been grinded into small particles initially. About 30 g of grinded *Garcinia Mangostana* (GM) pericarp has been filled into filter membrane and extracted by using soxhlet extractor. About 250 ml of methanol has been used as a solvent. The extraction of *Garcinia Mangostana* (GM) pericarp has been conducted in different parameters such as different temperature and time. The extraction of xanthone has been proceeding into bath evaporation to remove the solvent from extracted xanthone which is oily form. All samples have been obtained about 20 ml each.

B. Fourier transform infrared spectroscopy (FTIR)

The FTIR analysis is the further analysis of extracted xanthone to detect and determine the functional group of the phenolic content and its composition.

Table 1: FTIR result of sample before freeze dry.

SAMPLE T (°C), T (Hour)	IR Spectrum Range	Absorbance Band	Peak
75.86 °C 5 Hours	3275.24 Alcohol, Phenols Broad Appearance	1645.72 C=C (Both sp ²) Medium Appearance	1014.19 Fluoroalkenes Ordinary Bond
80 °C 4 Hours	3291.5 Alcohol, Phenols Broad Appearance	1646.75 C=C (Both sp ²) Medium Appearance	1013.60 Fluoroalkenes Ordinary Bond
80 °C 6 Hours	3276.50 Alcohol, Phenols Broad Appearance	1646.83 C=C (Both sp ²) Medium Appearance	1013.66 Fluoroalkenes Ordinary Bond
90 °C 3.59 Hours	3275.73 Alcohol, Phenols Broad Appearance	1646.96 C=C (Both sp ²) Medium Appearance	1013.60 Fluoroalkenes Ordinary Bond
90 °C 5 Hours	3275.16 Alcohol, Phenols	1646.35 C=C (Both sp ²)	1013.48 Fluoroalkenes Ordinary Bond

	Broad Appearance	Medium Appearance			Multiple Broad Peak	(usually 3 or 4)	Alkenes
90 °C 5 Hours	3327.50 Alcohol, Phenols Broad Appearance	1634.75 C=C (Both sp ²) Medium Appearance	1014.35 Fluoroalkenes Ordinary Bond	90 °C 5 Hours	2362.90 N-H Bond Ammonium Ions Multiple Broad Peak	1758.18 Aldehyde Cyclic 5-Membered	667.94 Vinyl Cis-disubstituted Alkenes
90 °C 5 Hours	3310.97 Alcohol, Phenols Broad Appearance	1642.34 C=C (Both sp ²) Medium Appearance	1013.55 Fluoroalkenes Ordinary Bond	90 °C 5 Hours	2025.87 C-N R-N=C=S	1492.17 Aromatic C=C Weak to Strong (usually 3 or 4)	667.91 Vinyl Cis-disubstituted Alkenes
90 °C 5 Hours	3351.98 Alcohol, Phenols Broad Appearance	1642.34 C=C (Both sp ²) Medium Appearance	1014.38 Fluoroalkenes Ordinary Bond	90 °C 5 Hours	1920.25 C-N R-N=C=S	1551.01 Aromatic C=C Weak to Strong (usually 3 or 4)	667.84 Vinyl Cis-disubstituted Alkenes
90 °C 5 Hours	3349.57 Alcohol, Phenols Broad Appearance	1640.04 C=C (Both sp ²) Medium Appearance	1014.02 Fluoroalkenes Ordinary Bond	90 °C 5 Hours	2375.32 N-H Bond Ammonium Ions Multiple Broad Peak	1582.11 Aromatic C=C Weak to Strong (usually 3 or 4)	667.80 Vinyl Cis-disubstituted Alkenes
90 °C 6.41 Hours	3275.95 Alcohol, Phenols Broad Appearance	1646.48 C=C (Both sp ²) Medium Appearance	1013.59 Fluoroalkenes Ordinary Bond	90 °C 5 Hours	2355.72 N-H Bond Ammonium Ions Multiple Broad Peak	1602.15 Aromatic C=C Weak to Strong (usually 3 or 4)	662.87 Vinyl Cis-disubstituted Alkenes
100 °C 4 Hours	33293.87 Alcohol, Phenols Broad Appearance	1646.34 C=C (Both sp ²) Medium Appearance	1014.64 Fluoroalkenes Ordinary Bond	90 °C 6.41 Hours	2377.09 N-H Bond Ammonium Ions Multiple Broad Peak	1758.10 Aldehyde Cyclic 5-Membered	667.71 Vinyl Cis-disubstituted Alkenes
100 °C 6 Hours	3327.87 Alcohol, Phenols Broad Appearance	1646.61 C=C (Both sp ²) Medium Appearance	1017.59 Fluoroalkenes Ordinary Bond	100 °C 4 Hours	2353.00 N-H Bond Ammonium Ions Multiple Broad Peak	1540.97 Aromatic C=C Weak to Strong (usually 3 or 4)	667.95 Vinyl Cis-disubstituted Alkenes
104.14 °C 5 Hours	3323.53 Alcohol, Phenols Broad Appearance	1612.30 C=C (Both sp ²) Medium Appearance	1011.32 Fluoroalkenes Ordinary Bond	100 °C 6 Hours	2352.29 N-H Bond Ammonium Ions Multiple Broad Peak	1758.27 Aldehyde Cyclic 5-Membered	667.92 Vinyl Cis-disubstituted Alkenes
Table 2: FTIR result of sample after freeze dry.				104.14 °C 5 Hours	2352.51 N-H Bond Ammonium Ions Multiple Broad Peak	1756.99 Aldehyde Cyclic 5-Membered	667.90 Vinyl Cis-disubstituted Alkenes
SAMPLE T (° C), T (Hour)	IR Spectrum Range	Absorbance Band	Peak				
75.86 °C 5 Hours	2375.35 N-H Bond Ammonium Ions Multiple Broad Peak	1757.93 Aldehyde Cyclic 5-Membered	667.8 Vinyl Cis-disubstituted Alkenes				
80 °C 4 Hours	2353.10 N-H Bond Ammonium Ions Multiple Broad Peak	1757.03 Aldehyde Cyclic 5-Membered	667.86 Vinyl Cis-disubstituted Alkenes				
80 °C 6 Hours	2376.10 N-H Bond Ammonium Ions Multiple Broad Peak	1757.00 Aldehyde Cyclic 5-Membered	667.94 Vinyl Cis-disubstituted Alkenes				
90 °C 3.59 Hours	2364.08 N-H Bond Ammonium Ions	1540.66 Aromatic C=C Weak to Strong	668.09 Vinyl Cis-disubstituted				

According to the results that detected by the Fourier Transform Infrared Spectroscopy (FTIR), all the samples that has been analyzed and detected in the range of 515.0 to 4000 cm⁻¹. Based on the results of FTIR for samples that obtained before freeze drying, the composition that detected in all samples was alcohol and phenol broad appearance. This is because the samples still consists of methanol which not fully removes from the samples through bath evaporator. Furthermore, all samples were detected in C=C (Both sp²) medium appearance and fluoroalkene ordinary bond. Most of samples was achieved the transmittance at range between 96.1 %T to 97.6 %T which highly corresponding to the wavelength.

According to the results of FTIR for samples that obtained after freeze drying, most of samples were detected consist of N-H Bond and ammonium ions with multiple broad peak. In additional, most of samples were detected consist of aldehyde with cyclic 5-membered vinyl with cis-disubstituted alkenes. Most of samples was achieved the transmittance at range between 130 %T to 150 %T which is highly corresponding to the wavelength.

C. 2, 2-Diphenyl-1-Picrylhydrazyl 9 (DPPH) antioxidant activity

The technique of 2-Diphenyl-1-Picrylhydrazyl 9 (DPPH) had been used to identify the radical scavenging activity in each of the samples of xanthone. Physically, the purple colour of 2-Diphenyl-1-Picrylhydrazyl 9 (DPPH) solutions was changed to yellow colour as the reaction occurred due to the samples consist of antioxidant.

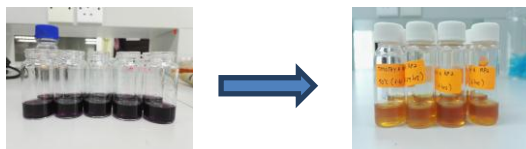


Fig. 1: DPPH solution changes from purple to yellow.

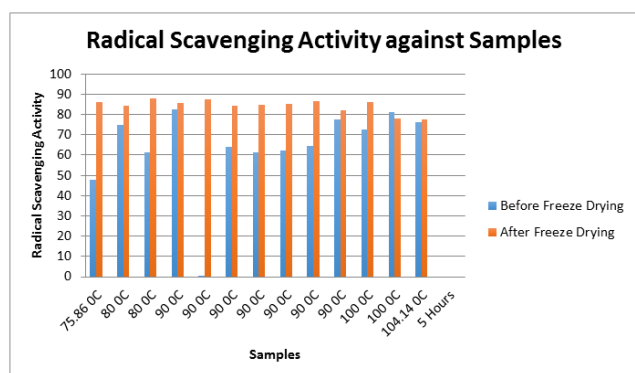


Fig. 2: Results of radical scavenging activity.

According to the result of experiment before the sample had been freeze dry to produce powder, the values of radical scavenging activity that obtained in range 47.93% to 82.69%. Whereas, the reading of absorbance that obtained for every samples with different parameters is between ranges of 0.351 nm to 0.788 nm. The highest value of radical scavenging activity is 82.69% at temperature 90 °C in 3.59 hours of extraction process whereas the reading of absorbance is only 0.351. For the result of experiment after the sample had been freeze dry to produce powder, the values of radical scavenging activity that obtained in range 77.47% to 88.01%. Whereas, the reading of absorbance that obtained for every samples with different parameters is between ranges of 0.257 nm to 0.483nm. The highest value of radical scavenging activity is 88.01% at temperature 80 °C in 6 hours of extraction process whereas the reading of absorbance is only 0.257nm.

This condition means that the lower reading of absorbance can gave the highest value of radical scavenging activity as it consists of high antioxidant which can inhibit the free radicle activity in human body. Thus, the sample that obtained from freeze drying has high antioxidant as the value of radical scavenging activity is higher than the sample obtained before freeze drying.

D. Total phenolic content (TPC)

Gallic acid is used as a standard with concentrations ranging from 0.0025 to 0.6 mg/mL. The measurement was carried out in quadruplicate to perform standard calibration curve.

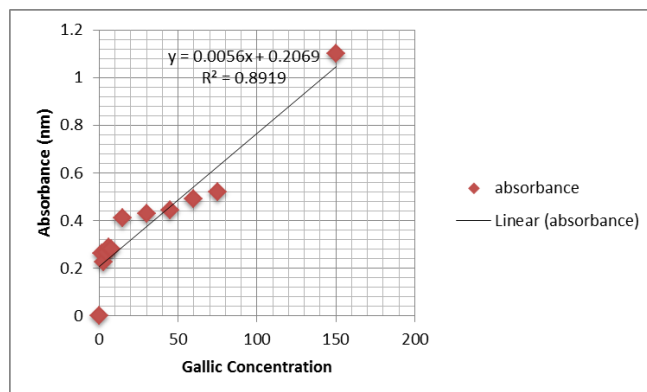


Fig. 3: Standard curve of total phenolic content (TPC).

The total phenolic content is expressed as milligram gallic acid equivalents per gram of pericarp powder (mg GAE/g). Physically, the yellow colour of Folin-Ciocalteu solutions was changed to purple colour as the reaction occurred due to the samples consists of antioxidant.



Fig. 4: Folin-Ciocalteu solution changes from yellow to purple.

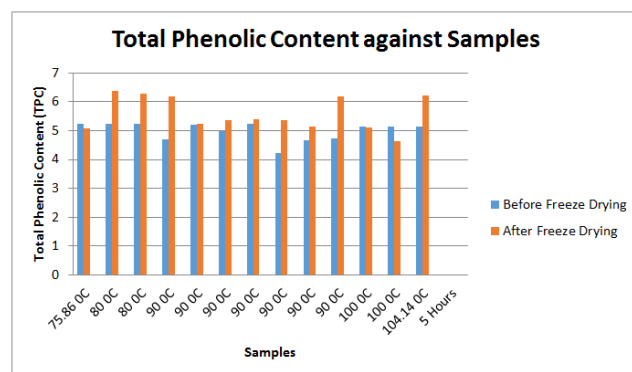


Fig. 5: Result of total phenolic content (TPC).

According to the result of experiment before the sample had been freeze dry to produce powder, the values of total phenolic content (TPC) that obtained in range 4.229 mg GAE/g to 5.240 mg GAE/g. Whereas, the reading of absorbance that obtained for every samples with different parameters is between ranges of 3.049 nm to 3.728 nm. The highest value of total phenolic content (TPC) is 5.240 mg GAE/g at temperature 80 °C in 6 hours of extraction process whereas the reading of absorbance is only 3.728 nm. For to the result of experiment after the sample had been freeze dry to produce powder, the values of total phenolic content (TPC) that obtained in range 4.646 mg GAE/g to 6.359 mg GAE/g. Whereas, the reading of absorbance that obtained for every samples with different parameters is between ranges of 3.613 nm to 4.480 nm. The highest value of total phenolic content (TPC) is 6.359 mg GAE/g at temperature 80 °C in 4 hours of extraction process whereas the reading of absorbance is only 4.480 nm.

This condition means that the higher reading of absorbance can gave the highest value of total phenolic content as it consists of high antioxidant which can inhibit the free radicle activity in human body. Thus, the sample that obtained from freeze drying has high antioxidant as the value of total phenolic content is higher than the sample obtained before freeze drying.

E. Moisture content

All samples have been analyzed by using the moisture analyzer at temperature 110 °C in 10 minutes. The reading of initial and final weight and also moisture content for each samples have been obtained directly from the moisture analyzer.

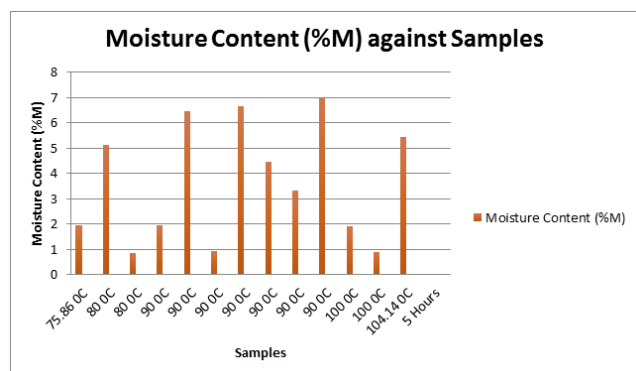


Fig. 6: Moisture content of samples in powder form.

Based on the results that obtained during the experiment, the highest moisture content for the powder of extracted xanthone from *Garcinia Mangostana* (GM) is achieved 7.02 %M which has been extracted at temperature 90 °C in 6.41 hours. The sample at this condition consists of high amount of water since it required more time of extraction process. The longer time taken, the more cycle of solvent extraction occurs so the moisture content of sample also high.

F. Antimicrobial Activity

Most of samples which in liquid form only showed the formation of colony after 72 hour kept in the incubator at temperature of 37 °C. The lowest Minimum inhibitory concentration (MIC) to gram positive bacteria was 0.5 mg/mL.

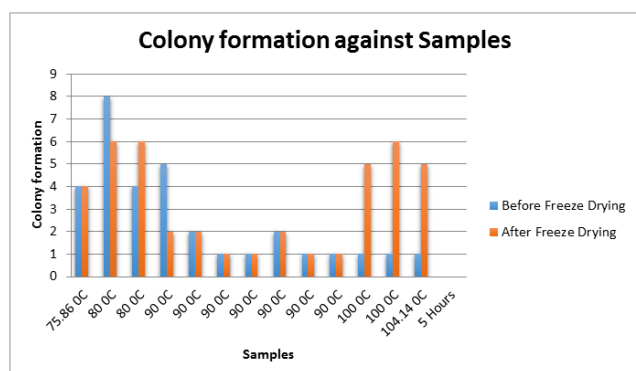


Fig. 7: Colony formation of samples before and after freeze dry.

The results of colony formation that obtained through the colony counter have been shown as below;

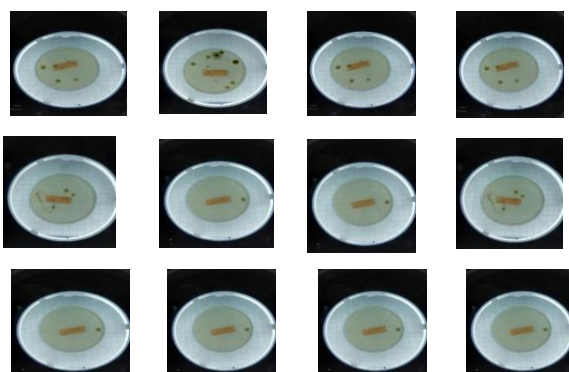


Fig. 8: Colony formation of samples in before freeze drying.

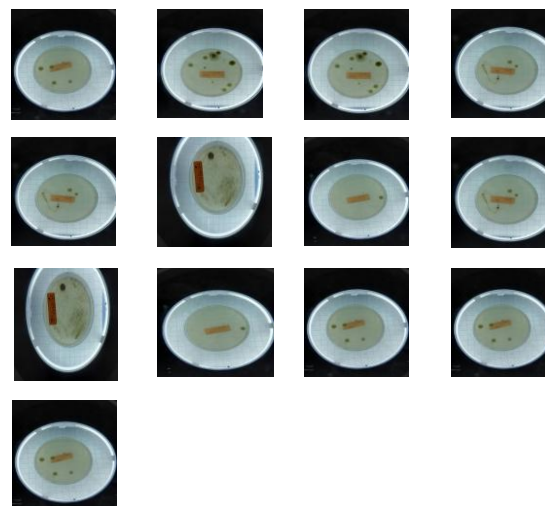


Fig. 9: Colony formation of samples after freeze drying

From the observation, the highest number of colony formation for samples that obtained before freeze drying on the agar plate is 8 colonies at temperature 80 °C in 4 hours of extraction process. This is because the microorganism still not inactivated during extraction process since the temperature of 80 °C cannot inhibit the activity of microorganism. For the samples that obtained after freeze drying in form of powder, the highest number of colony formation on agar plate is 6 colonies at temperature 80 °C in 4 and 6 hours and also 100 °C in 6 hours. The number of colony formation higher at temperature of 100 °C in 6 hours because the way of handling during experiment was exposed to the environment. Besides, the raw materials or *Garcinia Mangostana* (GM) pericarp does not have any pre-treatment and sterilization process before preceded to the preparation of samples. Thus, the presence of microorganisms on the surface of *Garcinia Mangostana* (GM) pericarp still detected and does not inhibited.

G. Freeze drying (microencapsulation)

The extracted xanthone has been dried by using freeze dryer in two days which equal to 48 hours. Freeze drying helps to remove the amount of water and excess of solvent extraction (methanol) that available in the samples to get a powder. From the observation, the amount of extracted xanthone in powder form that obtained is about 10 g for each of samples. Physically, the colour of collected powder for each sample is yellowish.



Fig. 10: Samples in powder form after freeze drying.

IV. CONCLUSION

The extraction of xanthone from *Garcinia Mangostana* (GM) pericarp proves that it consists of high antioxidant. This antioxidant can avoid and prevent the free radical activities in human body. It is proved by the analysis done on xanthone such as chemical, physical and biological such as 2, 2-Diphenyl-1-

Picrylhydrazyl 9 (DPPH) antioxidant activities, total phenolic content (TPC), moisture content and antimicrobial activity. Thus, the sample that obtained from freeze drying in form of powder has high antioxidant as the value of radical scavenging activity and total phenolic content (TPC) is higher than the sample obtained before freeze drying. This condition means that the lower reading of absorbance can give the highest value of radical scavenging activity whereas the higher reading of absorbance can give the highest value of total phenolic content (TPC) as it consists of high antioxidant which can inhibit the free radical activity in human body. The moisture content was achieved 7.02 %M which has been extracted at temperature 90 °C in 6.41 hours as it took long time to be extracted. For antimicrobial activity, the appearance of microbes colony of extracted xanthone on agar plate in average 1 colony for most samples since it does not have any pre-treatment or sterilization process.

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