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FULL PAPER

The influence of Gum Arabic on the physicochemical and antimicrobial activity of the microencapsulated Mahkota Dewa (*Phaleria macrocarpa*) leaves

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Abstract

Mahkota Dewa (*Phaleria macrocarpa*) also known as God's Crown has been historically used as an indispensable alternative herbal medicine. Microencapsulation is a process whereby tiny particles or droplets are engulfed or enclosed in a coating matrix to produce small capsules. Generally, without microencapsulation, powders are fragile materials that could easily interfere with other components that are difficult to dissolve in water, lose their beneficial properties and decrease shelf life. It is hoped that the microencapsulation would increase the consistency of the powder during storage and maintain its beneficial properties. The goal of this research is to investigate the physicochemical and antimicrobial activity of Mahkota Dewa leaves encapsulated in different concentrations of gum Arabic (GA) and to determine the form of antioxidant and their role and properties. Mahkota Dewa leaves powders were microencapsulated in 0%, 2%, 4%, 6%, 8% and 10% gum Arabic using an ultrasonic spray dryer at 90°C. The microencapsulated Mahkota Dewa leaves (MMDL) samples were subjected to physicochemical and antimicrobial activity. The results showed that the 6% GA MMDL exhibited the highest yield (3.91%) while 0% GA was the lowest yield (1.64%). The highest total phenolic and flavonoid content was exhibited by 2% GA. The highest DPPH inhibition was depicted in 0% GA which indicates the highest antioxidant activity (54.9±0.01%) and is significantly ($p<0.05$) different from other samples. The highest inhibition was exhibited in 0% GA in the TBA method and FTC analysis. The encapsulated powders were identified to have weak antimicrobial activity against *Bacillus cereus, Escherichia coli, Staphylococcus aureus, Salmonella* and *Listeria monocytogenes*. The powders produced have an irregularly spherical structure and smooth surface with some dented spots on the surface. The different concentration of gum Arabic resulted in different antioxidant activity, flavonoid content and antimicrobial activity of MMDL.

1. Introduction

Mahkota Dewa (*Phaleria macrocarpa* Scheff. Boerl) is also known as God's Crown, or Pau has mostly been traditionally used as an indispensable medicinal plant in Malaysia and Indonesia (Azmir *et al.,* 2014). Despite widespread use as alternative herbal medicine, few attempts have been made to examine bioactive compounds such as flavonoids and their biological activity, such as antioxidant activity and antimicrobial properties in the leaves of Mahkota Dewa. These bioactive compounds are usually associated with volatile losses and degradation (Akdaş and Başlar, 2015). Bioactive compounds are susceptible to deterioration due to the pH, light, oxygen, temperature, and enzymatic activities (Roselló-Soto *et al*., 2019; Ng *et al*., 2020). Furthermore, without microencapsulation or non-coating, powders are fragile, easy to bind to other components and become sticky, difficult to dissolve in water and even decrease the shelf life (Wróblewska-Krepsztul *et al*., 2019).

Microencapsulation by spray drying is the most popular and effective technique used in the food industry (Mohammed *et al*., 2020) due to its availability of equipment and low cost (Costa *et al*., 2015). Moreover,

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microencapsulation by spray drying also helps envelop sensitive ingredients from nutritional deterioration, volatile loss and adverse reaction in a coating material (Wu *et al*., 2014). The choice of material for microencapsulation is important for efficiency and microcapsule stability (da Silva *et al*., 2014). Microencapsulating agents should be water-soluble at an acceptable level (Shukla *et al*., 2016), possess good properties of emulsification, film-forming, and low viscosity (Jafari *et al*., 2008). Therefore, the utilization of gums, maltodextrins, pectins, vegetable fibres and starches as microencapsulating agents in spray drying helps to improve the stability of powders during storage (Bhandari and Howes, 2005). Gum Arabic stands out as a good microencapsulating agent for its excellent emulsification properties and is most widely used (Zainol *et al*., 2019).

Gum Arabic is a product obtained from the dried discharges of sticky stems and branches of *Acacia senegal* which consists of a water-soluble dietary fibre digestible only in the intestines less (FAO, 1999). Gum Arabic is used as encapsulating agents due to its film forming and emulsion stabilization properties. Gum Arabic can be simply mixed, cross-linked, or transformed into semi-interpenetrating networks with other polymers, widely used in the development of microcapsules, nanoparticles, and emulsions for drug-loading applications (Mariod, 2018).

The demand for organic and natural foods or dietary supplements is increasing as consumers are looking for functional foods that come from plant sources that have medicinal properties. In order to address these lapses, enhance and preserve the antioxidant properties of the extract, microencapsulation using gum Arabic may prevent the extracted product from unwanted degradation of bioactive compounds and could provide protection against harsh environmental conditions. Thus, this study is intended to investigate the ramification of various concentrations of Arabic gum against the physicochemical and antimicrobial activity of microencapsulated Mahkota Dewa leaves (MMDL) powder prepared using ultrasonic spray drying technique.

2. Materials and methods

2.1 Raw materials

The Mahkota Dewa leaves (*P. macrocarpa*) have been collected from Kuala Nerus, Terengganu. The leaves were washed under running water and mixed into a solution using a food processor, filtered with a muslin cloth and placed in a 4°C refrigerator prior to further processing.

2.2 Microencapsulation by ultrasonic spray dryer

Microencapsulation by ultrasonic spray drying of Mahkota Dewa leaves samples were carried out based on the method by (Zainol *et al*., 2017), with a slight modification. Mahkota Dewa leaves (150 g) were blended in 1500 ml of distilled water, filtered using a muslin cloth and mixed with varying amounts of Arabic gum (2%, 4%, 6%, 8% and 10%). The resulting solution was spray-dried using an atomizer nozzle-equipped ultrasonic-spray dryer (YKNTECH, Kulim, Malaysia). The ultrasonic spray dryer outlet temperature was set at 80°C while the feed flow rate was kept at 8 mL/min. The micro-encapsulated plant powder was collected and stored in an opaque container at 4°C prior to further analysis.

2.3 Determination of total antioxidant activity assay

2.3.1 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity

Approximately 0.1 mM of DPPH working solution was prepared by dissolving 1.9 mg of DPPH in 100 mL of methanol. An aliquot of 4 mL of this solution was applied to 10 mL of diluted extracts (50 mg sample in 100 mL of distilled water), 10 mL of distilled water (control) and 10 mL of regular ascorbic acid, α-tocopherol and butylated hydroxyltoluene (BHT). All the mixture was then left to be incubated in the dark at room temperature for 60 min. The results of the reaction were then measured at 517 nm using the UV-Vis spectrophotometer (Malik *et al*. 2017).

2.3.2 Ferric thiocyanate (FTC) method

A mixture of 4.0 mg of sample in 4 mL of absolute ethanol (99.5%), 4.1 mL of 2.52% of linoleic acid in 99.5% absolute ethanol, 8.0 mL of 0.05 M of phosphate buffer (pH 7.0) and 3.9 mL of water in a screw-cap vial was placed in an oven in a dark or amber bottle at 40°C. Approximately 9.7mL of 75% (v/v) ethanol and 0.1 mL of 30% ammonium thiocyanate have been added to 0.1 mL of this mixture. Precisely 3 mins after 0.1 mL of 0.02 M ferrous chloride was applied to the reaction mixture in 3.5 percent hydrochloric acid, the absorbance was measured at 500 nm at every 24 hrs interval until 1 day after the control absorbance reached its maximum level. Butylated hydroxytoluene (BHT) and α-tocopherol were used as positive controls in this assay, while mixture without sample or blank was used as the negative control (Zainol *et al*., 2018).

2.3.3 Thiobarbituric acid (TBA) test

The same samples prepared for the FTC method were used in this research. In the sample solution, 2.0

mL of the sample solution was added with 0.1 mL of 20% aqueous trichloroacetic acid (TCA) and 2.0 mL of aqueous thiobarbituric acid (TBA) solution (Zainol *et al*., 2018). The mixture was placed in a boiling water bath for 10 min. After cooling to room temperature, it was centrifuged at 3000 rpm for 20 min. The absorbance of the resulting supernatant was measured at 532 nm (Chong *et al*., 2018).

2.3.4 Determination of total phenolic compound (TPC)

A sample of 1 mg/mL of methanol extract was added to 4.5 mL of deionized distilled water and 0.5 mL of Folic-Ciocalteu reagent was applied to the solution being prepared. These samples were stored for 5 min at room temperature, followed by the addition of 5 mL of 7% sodium carbonate and 2 mL of deionised distilled water. The samples were then incubated with intermittent shaking at 40°C for 90 mins and the absorbance was measured at 750 nm using a spectrophotometer. The overall phenolic content was expressed as mg of gallic acid equivalents (GAEs) per gram of sample (Ng *et al*. 2020).

2.3.5 Total flavonoid content (TFC)

An aliquot of 1 mL sample solution was mixed with 4 mL of distilled water in a tube. Then, 0.3 mL of 5% sodium nitrite ($NaNO_2$) was added to the mixture and allowed to react for 5 mins. The mixture was then added to 0.3 mL of 10% aluminium chloride ($AlCl_3$) and allowed to stand for another 5 mins before adding 2 mL of 1 M sodium hydroxide (NaOH) and 2.4 mL of distilled water. The absorbance was then measured at 510 nm against a blank of distilled water. Quercetin was used as a standard compound for the quantification of total flavonoids. All values were expressed as mg quercetin equivalents (QE) per gram of sample (mg QE/g sample) (Looi *et al*., 2020).

2.4 Physical analysis

2.4.1 Determination of functional groups

Functional groups present in MMDL were measured using a Fourier Transform Infrared Spectroscopy (FTIR). FTIR using an attenuated total reflection (ATR) technique was used in this experiment to investigate the structural changes of cellulose paper by obtaining its infrared spectra. Two milligrams of MMDL powder were added with 200 mg KBr. After homogenising, the powder was pressed into pellets (1-2 mm thick films) with a 15-ton hydraulic press. FTIR spectra were obtained of wavenumbers from 400 to 4000 cm^{-1} during 64 scans, with 2 cm^{-1} resolution (Hau *et al*., 2018).

2.4.2 Morphological characteristic of MMDL

Morphology of MMDL was measured using scanning electron microscopy (SEM) where the sample was stubbed with double-sided adhesive tape and coated with gold. The dried samples were mounted on conductive double-sided tape on a specimen stub. The specimen stub was then attached to the height adjustment screw. The height of the holder was adjusted so that clearance between the topmost point of the specimen and the lowest face of the gauge becomes 1 mm (Noorfarahzilah *et al*., 2020).

2.4.3 Colour profile

The encapsulated powder was evaluated using a colorimeter (Minolta Chromater CR-300, Japan). The Minolta Chromater CR-300 is a compact tristimulus colour analyser for measuring the reflective colours of surfaces. Colour, as perceived, has three dimensions which are L*, a* and b*. Each sample was measured in triplicate individually (Chew *et al*., 2020).

2.5 Microbiological analysis

2.5.1 Antimicrobial analysis

The antimicrobial activity of MMDL powder extract was performed using a modified well diffusion technique as described by Liew *et al*. (2020). The microorganism tested, *Bacillus cereus, Listeria monocytogenes, Salmonella, Escherichia coli, Klebsiella pneumonia, Pseudomonas aeruginosa,* and *Staphylococcus aureus* was incubated at 37°C for 24 hrs in nutrient broth. One colony of cultivated bacteria was selected, mixed with saline water and stored in a freezer at -40°C. Approximately 100 µL of the bacteria suspension was then swabbed over Mueller Hinton Agar (MHA) plates. The small wells were cut in agar using sterile micropipette tips. The stab wire was used to remove the excess agar. One drop of molten agar was loaded into the wells followed by 50 µL of extracts samples. The plates were incubated at 37°C for 24 hrs and the inhibition zones around the wells were recorded as the final results.

2.6 Statistical analysis

All analysis was conducted in triplicates and the statistical analysis was obtained using the MINITAB 14 statistical software. The significance difference from the triplicate analysis was performed by analysis of variance (Mohd Zin *et al*., 2017).

3. Results and discussion

3.1 Yield of MMDL Powder

Table 1 indicates an increase in the output of the product as the concentration of Arabic gum increases.

The 10% microencapsulated Mahkota Dewa leaves (MMDL) sample exhibited the highest yield followed by 8%, 6%, 4%, and 2% of gum Arabic microencapsulation. The data are in concert with Adejoro *et al*. (2019), who quoted that the maltodextrin/gum Arabic microparticles were smaller and more homogenous than those of native starch even at higher loading concentration. The overall yield of encapsulation was also correlated with overall emulsion stability and minimum droplet size (Carneiro *et al*., 2013). The study also indicated that using a higher homogenization pressure and/or longer homogenization time while preparing sample emulsion could result in a smaller particle size (Jafari *et al*., 2008). Spray drying processes can produce sticky products, particularly when feed materials are rich in sugars and acids. This may result in the adhesion of the particles to the internal wall of the drying chamber, resulting in agglomeration of the particles and lower encapsulation output (De Melo Ramos *et al*., 2019). Thus, the addition of additives such as gums, maltodextrins, pectins, vegetable fibres and starches as encapsulation agents in spray drying helps to improve the stability of the powders during storage.

3.2 Chemical analysis

3.2.1 Total phenolic content (TPC)

Table 1 also shows the 6% MMDL sample exhibited a higher total phenolic content (TPC) than that of other samples, while the 2% MMDL sample had a lower overall phenolic content than the rest of the samples. The TPC of the samples was found to be not significantly different ($p<0.05$) within the samples, indicating that the GA concentration did not cause a change in the total phenolic content of the samples at the same temperature in the ultrasonic spray dryer. High temperatures can cause plant phenols to degrade at the time of extraction and at high temperatures (Che Sulaiman *et al*., 2017). The results may be due to a microencapsulation ultrasonic spray drying technique that involves a defensive mechanism consisting of the creation of a wall membrane that covers the particles of the encapsulated material. This means that at these two stages, there is no leakage of active ingredients, phenolic compounds from wall materials (Mozafari *et al*., 2008).

3.2.2 Total flavonoid content (TFC)

The amount of flavonoid showed a marginally growing trend from 0% GA to 6% followed by a small decrease in 8% and 10% of the MMDL sample. (Table 1). The highest total flavonoid content was observed in 6% GA and was significantly different ($p < 0.05$) from other MMDL samples. The loss of total flavonoid in uncoated powder of MMDL (0% GA), may be due to the degradation of the phenolic compound. Addai *et al*. (2013) quoted that the degradation of phenolic compounds might be due to the higher rate of respiration which would result in loss of phenolic and flavonoids contents. Some flavonoid compounds were possibly destroyed at high temperatures, resulting in a decrease in total flavonoid content. In addition, the different amount of flavonoid may be due to the transfer or release of glycosides or the binding of phenolic to free phenolic derivatives (Panche *et al*., 2016). Heating can cause the formation of monomers by hydrolysis of C-glycoside, which increases the content of these flavonoids by heating at 120°C for 30 mins since Quercetin conjugates in onion are resistant to thermal degradation (Sharma *et al*., 2015).

3.3 Antioxidant activity in MMDL sample powder

3.3.1 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay

Inhibitions of DPPH radical scavenging activity showed a decreased trend as the concentration of GA increased, accompanied by a small rise in inhibitions at 6% GA (Table 1). All samples tested were significantly ($p<0.05$) different except for 0% GA and 6% GA.

Table 1. Antioxidative activity (DPPH, FTC and TBA value), total phenolic content, total flavonoid contents and yield of MMDL powder encapsulated in different percentages of gum Arabic

Samples	DPPH	FTC (5th day Abs)	TBA (% inhibition)	Total phenolic content (mg GAE/g sample)	Total flavonoid content (mg QE/g sample)	Yield of MMDL powder encapsulated
0% GA	$53.72±4.41^{c}$	$0.84±0.12^{b}$	$79.36±11.44^{a}$	$45.52±3.14^{ab}$	$58.62±2.56^{c}$	$2.00±0.01^{c}$
2% GA	$48.18±5.52^{d}$	$1.13±0.12^{a}$	$80.21±15.52^{a}$	$30.19±3.47^{b}$	$61.28±3.29^{c}$	$2.45±0.02^{bc}$
4% GA	$23.78±7.33^{f}$	$1.22±0.11^{a}$	$66.89±13.75^{b}$	$50.01±2.39^{a}$	$80.78±9.08^{bc}$	$2.66±0.01^{b}$
6% GA	$33.97±5.24^{e}$	$1.15±0.14^{a}$	$84.93±14.23^{a}$	$67.99±3.88^{a}$	$86.66±0.70^{a}$	$2.72±0.01^{b}$
8% GA	$35.72±5.21^{e}$	$1.25±0.08^{a}$	$87.88±15.21^{a}$	$57.08±2.30^{a}$	$74.30±0.78^{c}$	$3.44±0.01^{ab}$
10% GA	$36.55±3.24^{e}$	$1.26±0.07^{a}$	$89.65±13.46^{a}$	$45.63±3.96^{ab}$	$78.74±2.52^{bc}$	$3.59±0.02^{a}$
BHT	$72.09±6.22^{a}$	$1.14±0.43^{a}$	$22.35±3.53^{c}$	$45.52±3.14^{ab}$	$58.62±2.56^{c}$	
α-tocopherol	$66.93±5.75^{b}$	$0.86±0.41^{b}$	$34.91±6.72^{c}$	$30.19±3.47^{b}$	$61.28±3.29^{c}$	

Values are expressed as mean±standard deviation of triplicate testing. Values with different superscript within the column are significantly different ($p < 0.05$).

Despite that, the uncoated powder, (0% GA) showed significantly ($p<0.05$) the highest DPPH value (53.7±0.81%), while, 4% GA sample showed the lowest DPPH inhibition (20.4±1.77%). These findings may be due to a microencapsulation technique with gum Arabic, which covers the extracted compounds and protects them from the environment. Jyothi *et al*. (2010) stated that the microencapsulation helps to minimise the core reactivity of environmental factors and minimise the transmission rate of the core material to the outside environment by creating a physical barrier between the core compound and the other external compound of the product. Moreover, microencapsulation by spray drying also helps envelope sensitive ingredients from nutritional deterioration, volatile loss and adverse reaction in a coating material (Wu *et al*., 2014). This is in agreement with Wu *et al*. (2014) who stated that GA could link to flavonoid in plants when they come into contact with the emulsion. It is also able to retain the flavonoids extracted throughout the spray drying process. This finding is supported by Gharsallaoui *et al*. (2007) which cited that gum Arabic acts as a semi-permeable membrane that encapsulated the samples which produced a limited barrier against oxidation and unstable core material.

3.3.2 Ferric thiocyanate (FTC) method

Table 1 also shows the activity of powder encapsulated MMDL with different concentrations of GA on the fifth day of incubation, as indicated by Malik *et al.* (2017) who reported that, in the FTC assay, the blank and sample's absorbance readings increased gradually before a sudden drop on 6th day. The data shows the 8% (1.25±0.08%) and 10% MMDL samples (1.26±0.07%) displayed high antioxidative properties in FTC analysis, whereby 0% MMDL samples (0.84±0.12%) extricate the lowest absorbance FTC values of all samples. The inhibitory effect of ferrous chloride oxidation on ferric particles by antioxidant compounds assessed by the formation of a ferric thiocyanate complex indicates the existence of good antioxidant activity. The outcome demonstrates that the greater part of its specimens showed great impact in repressing linoleic corrosive oxidation similar to the positive controls. All samples with the exception of 0% of MMDL displayed very remarkably ($p<0.05$) distinctive antioxidant activity with α-tocopherol and BHT. Similarly, Zainol *et al*. (2017) reported that, in the light of the findings, the distinctive microencapsulated plant powder suggested a number of cell reinforcement activities in the FTC strategy that could be attributed to the lowering of hydroperoxides, the inactivation of free radicals, and the chelation of metal particles or the mixing of them.

3.3.3 Thiobarbituric acid (TBA) test

The findings of the TBA method are not consistent with the results of the FTC method, where antioxidant activity in TBA was found to be much higher than in FTC. This means that MMDL samples exhibited antioxidant activity stronger at the second stage of lipid peroxidation. The 0% GA MMDL exhibited the highest ($p<0.05$) inhibition (97.1%) while samples 2%, and 4% GA illustrated a much lower inhibition. The results could be due to the slow release of phenolic compounds from the encapsulated samples, which were not able to control the formation of malonaldehyde (Roostaee *et al*., 2017). This phenomenon could also be due to the antioxidant in standards being interrupted during the experiment because the samples used for TBA are the same for FTC. Generally, the antioxidant by TBA method is greater than the FTC method. These findings show that the level of peroxide at the initial stage of lipid oxidation is lower than the level of peroxide at the secondary stage. It may also prove that the secondary product is even more consistent than the primary product.

3.4 Physical analysis

3.4.1 Colour analysis of microencapsulated Mahkota Dewa leaves powder

Table 2 shows an increase in lightness or L* values from low gum Arabic (GA) to high GA, with the MMDL sample encapsulated with 6 percent GA being the brightest powder. GA is a colourless encapsulating agent that does not change the powder's colour. In this analysis, however, higher GA concentration increased powder lightness, which may be attributed to the technique of spray drying at moderate temperature. Sagar and Suresh Kumar, (2010), reported that the increase in temperature during spray drying might have contributed to the reduction of betalains content. In the encapsulated powder, the negative a* value shows the green chlorophyll colour of the MMDL sample. From the findings, it was revealed that the powder exhibits the highest greenish colours with 4% GA 0% GA while 6% GA exhibited the lowest greenish colour, which is according to the lightness of the MMDL samples. The 2% GA MMDL sample showed the nearest green colour to the uncoated sample, which was the best encapsulation colour compound compared to the uncoated samples. The positive b* value showed a yellow colour ranging from (29.66±2.63%) to (25.59±02.015%) which showed no substantial difference ($p < 0.05$) from other samples. The high positive b* value could relate to the high green colour in the 4% GA sample, which could be due to the green colour of the Mahkota Dewa leaves. Colour measurement is important because it determines the quality of the powder produced during the drying process

Table 2. Colour profile of MMDL sample encapsulated in different percentages of gum Arabic

Parameter	A (0% GA)	B (2% GA)	C (4% GA)	D (6% GA)	E (8% GA)	F (10% GA)
L*	55.35±4.48[c]	59.26±7.54[bc]	63.32±2.32[bc]	64.11±6.72[bc]	69.94±7.33[b]	74.45±6.24[a]
a*	-17.11±1.53[a]	-16.88±1.26[a]	-8.60±1.01[c]	-12.21±0.78[b]	-12.64±0.87[b]	-11.23±0.55[b]
b*	27.61±3.13[a]	28.88±2.66[a]	28.64±2.21[a]	27.25±2.28[a]	27.47±1.78[a]	26.65±2.82[a]
Colour	Brownish orange					

Values are expressed as mean±standard deviation of triplicate testing. Values with different superscript within the column are significantly different ($p < 0.05$).

and reflects the sensory quality of the powder (Mohd Nawi *et al*., 2015).

3.3.2 Determination of functional groups

Figure 1 shows the absorption band of the FTIR spectrum at 3385.39 cm^{-1}, indicating the existence of the aromatic OH-group, 2927.17 cm^{-1} indicating the existence of the saturated C-H group, 1617.06 cm^{-1} is associated with the C = O functional group, while 1514.60 cm^{-1} and 1432.56 cm^{-1} are depicted as conjugated C = C aromatic. These data are within the range of the previous analysis of Mahkota Dewa leaves extracted using methanol (Nor Fariza *et al*., 2012). Hartati *et al.* (2005), cited that Mahkota Dewa had an aromatic ring replacement for C = O, an OH-group, a C-H-group, an unsaturated C = O-β and an aromatic ring. In plants, three broad groups of secondary metabolites are nitrogen-containing compounds, terpenoids and phenols. In addition, flavonoids are characterised by the presence of a C_{15}-(C_6-C_3-C_6) flavonoid nucleus based on a heterocyclic ring structure derived from phenylalanine (ring B) and polyketide biosynthesis (ring A) bound by an oxygen-containing pyran or pyron ring (ring C) (Ndhlala *et al*., 2010). In addition, flavonoids, including flavonoids, flavonols (Quercetin and Kaempferol) and condensed tannins have antioxidant activity that depends on the existence of free OH groups, in particular 3-OH groups (Panche *et al*., 2016). The data from the experiments (Table 3) also showed that MMDL contains aromatic compounds, namely the OH group, C = O group and CH group, and the spray drying encapsulation did not affect compounds in the encapsulated samples (Hartati *et al*., 2005).

Table 3. Functional group presence in MMDL powder

Functional group	FTIR band (cm^{-1})	Characteristic
Aliphatic Primary Amines	1617.06	Amines are colourless, aliphatic amines are transparent to UV, stronger bases than ammonia
Alkynes Monosubstituted	2103.13	Strong acidity, high boiling point
Aliphatic Hydrocarbons	2927.17, 1514.60 and 1432.56	Contain C-H stretching.
Primary Aliphatic Alcohols	3385.39	Contain polar OH group, boiling point higher than alkane

3.3.3 Morphological properties of MMDL microcapsules

Table 4 shows micrographs of the microencapsulated Mahkota Dewa leaves powder (MMDL) formed using different concentrations of Arabic gum (GA). The resulting powders had irregularly spherical structure and smooth surface but some dented spots on the surface in the 4% GA and 6% GA samples. Rosenberg *et al*. (1985) quoted that the formation of the dented surfaces could be due to the shrinkage of the particles induced by high temperature during the spray drying process. The greater surface area of the particles of MMDL was provided by the dented surface and spherical shape of the samples. These properties are attributed to high solubility and good bulk density. In comparison to freeze-dried powder of yoghurt, the powders were found to be non-spherical and showed linear structure while the spray-dried yoghurt was found to be in spherical particle and the inside of the particle had a hole in the centre and these properties are the same with spray dried milk powder (Koç *et al*., 2012). Core or wall ratio, wall content concentration, pH value, and also stirring speed on the morphology, particle size distribution output and loading rate are included in the morphology of microcapsules encapsulated by gelatine and gum Arabic using coacervation techniques (Marfil *et al*., 2018). However, it is interesting to note that due to high surface area, the powders are more exposed to the environment compared to other powders thus the rate of oxidation in 4% GA is higher than other powders. Chemical reactions occurred faster in finer powder than coarse powder. The spherical powder reacts with methanol faster to extract the polyphenols in the powder.

3.4.4 Antimicrobial analysis

The results obtained from antimicrobial analysis at a concentration of 0.3 mg/well against 5 types of bacteria (Gram-positive and Gram-negative) which are *B. cereus, S. aureus, Salmonella, E. coli* and *L. monocytogenes* (Figure 2). The MMDL powders exhibited weak to moderate inhibitory activities against all the bacteria

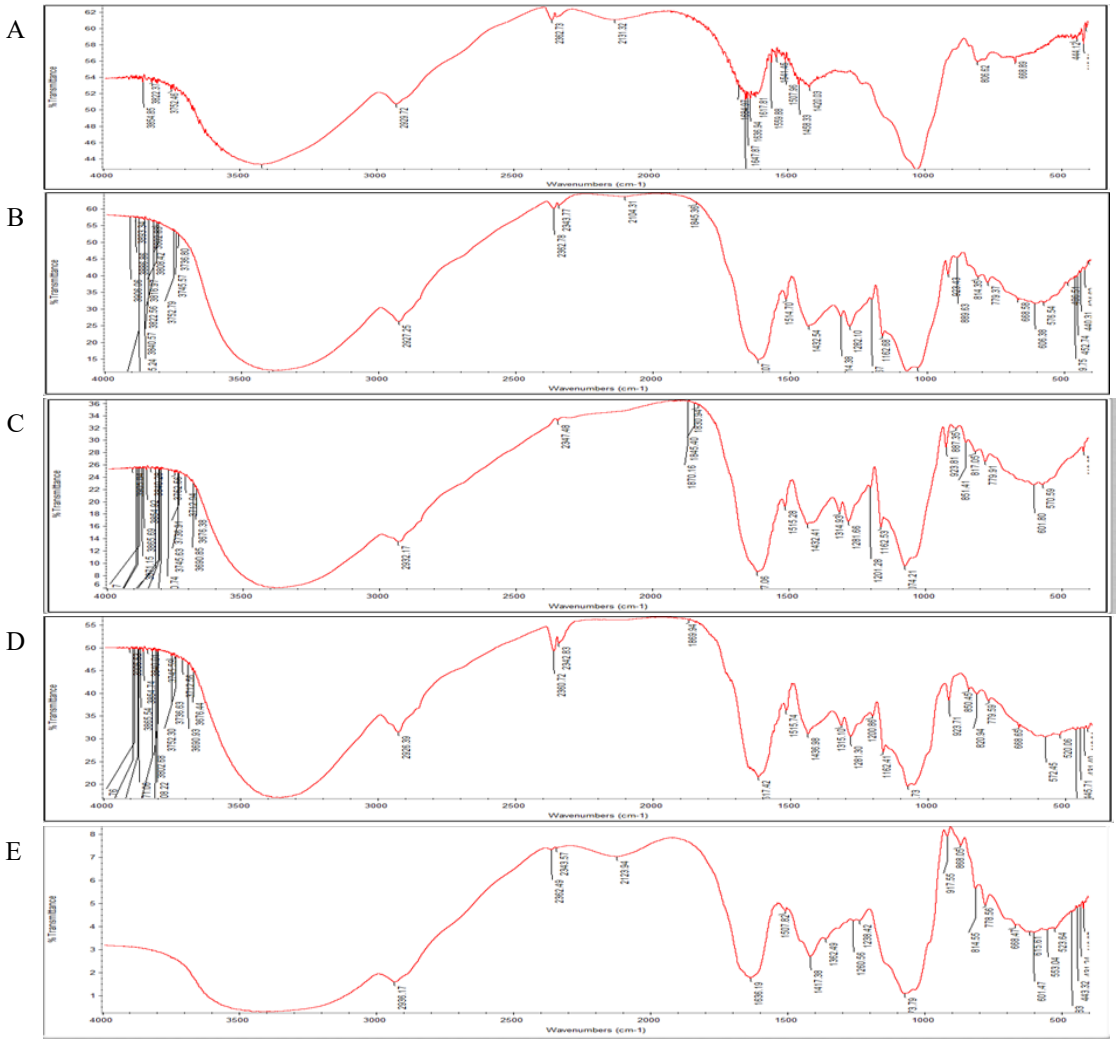
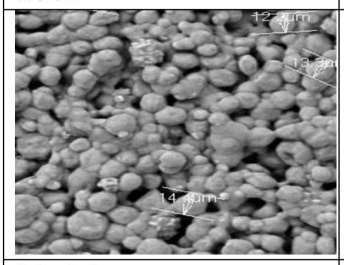
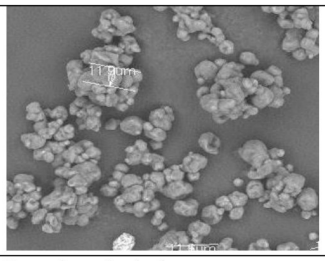
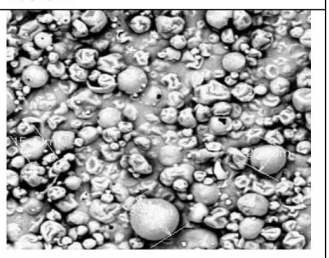
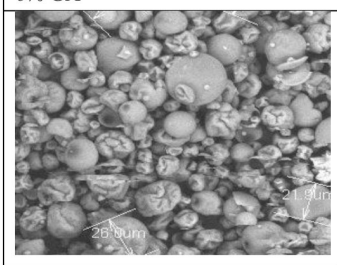
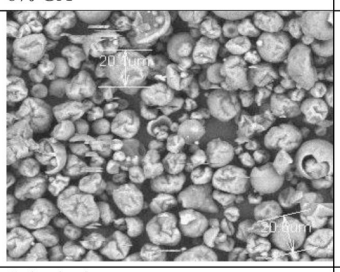
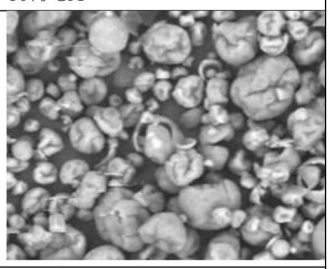


Figure 1. FTIR transmission spectrum of ultrasonic spray dried microencapsulated Mahkota Dewa leaves powder (MMDL) encapsulated in different percentages of gum Arabic. A = 2% GA; B = 4% GA; C = 6% GA; D = 8% GA and E = 10% GA

Table 4. Morphology and characteristic of microcapsules at 90˚C

0% GA	2% GA	4% GA
		
Spherical structure Smooth surface Non-consistent size No dents Particle size: (13.3 µm ± 1.1%)	Irregular spherical structure Smooth surface Non-consistent size Have dents Particle size: (10.53 µm ± 1.11%)	Irregular spherical structure Smooth surface Non-consistent size Have dents Particle size: (23.2 µm ±3.30%)
6% GA	**8% GA**	**10% GA**
		
Irregular spherical structure Not smooth surface Consistent size Have dents Particle size: (15.13 µm ± 0.23%)	Spherical structure Smooth surface Consistent size Have dents Particle size: (14.1 µm ± 0.85%)	Irregular spherical structure Not smooth surface Consistent size Have dents Particle size: (11.1 µm ± 0.96%)

tested. This is in line with Hendra *et al*. (2011) who cited that different parts of Mahkota Dewa fruits showed a weak to moderate ability with an inhibition range of 0.97 -2.17 cm at a concentration of 0.3 mg/well. Previous studies have proved that leaves of Mahkota Dewa have antimicrobial activities. Due to the presence of flavonoids, saponins, polyphenols and tannins content, Mahkota Dewa have greater inhibition activity against Gram-positive bacteria than Gram-negative bacteria. This phenomenon may be explained by the fact that Gram-negative bacteria are usually more resistant to antimicrobial agents than Gram-positive bacteria. The presence of an outer membrane permeability barrier in Gram-negative bacteria limits the function of antimicrobial agents in bacterial cells (Liew *et al*., 2020).

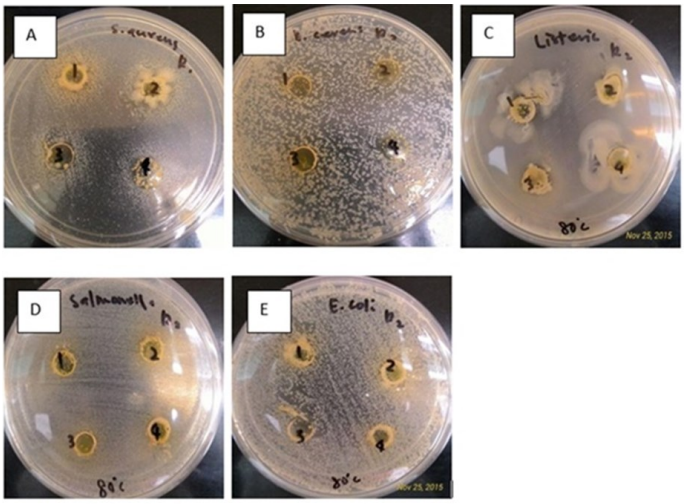


Figure 2. Growth inhibition of some food poisoning bacterial strains caused by MMDLD samples. Note: A = *Staphylococcus aureus*; B = *Bacillus cereus*; C = *Listeria monocytogenes*; D = *Salmonella*; E = *Escherichia coli*, 1 = 0% GA; 2 = 2% GA; 3 = 4% GA; 4 = 6% GA

4. Conclusion

This study found that 6% of GA MMDL produced the highest yield (3.91%) while 0 per cent of GA produced the lowest yield (1.64%), while 2% of GA MMDL produced the highest total phenolic and flavonoid content. In addition, different concentrations of gum Arabic showed some variance in antioxidant activity in the DPPH Radical Scavenging Activity, FTC and the TBA methods. During the initial stage of lipid peroxidation measured using the FTC method, the antioxidant activities showed no significant difference between all samples in both temperatures. However, antioxidant activity in the secondary stage of lipid peroxidation measured using the TBA method is higher than the FTC method. The lightness (L*) of MMDL powder increased as the gum Arabic increases while the highest greenish are exhibited by 4% GA and yet no significant differences were recorded. The encapsulated powders were identified to have weak antimicrobial activity against *B. cereus, E. coli, P. aeruginosa, S. aureus, Salmonella* and *L. monocytogenes*. The morphology of powders produced was identified to have characteristics of irregular spherical and smooth surfaces with some dented spots on the surface. In view of this finding, MMDL could serve as a good source of antioxidants and could be used as a food product to boost its nutritional and antioxidant properties.

Conflict of interest

The authors declare that is no conflict of interest

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