

## Physical properties and prebiotic activity of white dragon fruit (*Hylocereus undatus*) powders produced using different wall materials

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### Abstract

The aim of the present work was to investigate the spray-dried characteristics such as physical properties, morphologies, glass transition temperatures ( $T_g$ ), and prebiotic activity of white dragon fruit (WDF) powders produced using different wall materials, namely resistant maltodextrin (RMD) and maltodextrin (MD), at optimum spray drying conditions. Results showed that RMD decreased water activity and moisture content, and increased bulk density and true density of powder more than MD. In addition, the particle size of RMD-coated powder (WRMD) was smaller than that of MD-coated powder (WMD), and the morphology of the WRMD powder showed that it had a smooth surface as compared to WMD powder, where shrinkage and dent surfaces were observed. The  $T_g$  of WMD powder had higher value, but both types of powders were not significantly ( $p > 0.05$ ) different. Then, both powders were further investigated for their ability to support the growth of *Bifidobacterium longum* BB536 and *Lactobacillus casei* Shirota. The growth of the anaerobic bacteria was determined every 6 h for 24 h at 37°C in six modified MRS media containing glucose, RMD, MD, WRMD powder, WMD powder, and fructooligosaccharides (FOS) as the substrates. Results indicated that all substrates significantly ( $p < 0.05$ ) increased the growth of the probiotic bacteria, with WRMD powder yielding the highest bacterial count. Based on the findings, WRMD powder can be considerably used as a new prebiotic source for the functional food industry.

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### Keywords

powder properties,  
resistant maltodextrin,  
prebiotic,  
morphology,  
glass transition  
temperature

### Introduction

Nowadays, the demand for healthy foods is increasing, and many studies have proven that prebiotic is one of the dietary supplements that can positively affect host health. Guimarães *et al.* (2020) reported that prebiotics are polysaccharides, *e.g.*, inulin and oligosaccharides, fructooligosaccharides (FOS), galactooligosaccharides (GOS), and lactulose that can withstand digestion in the small intestine, but can selectively be fermented by predominant probiotic bacteria in the large intestine, namely bifidobacteria and lactobacilli (Di Criscio *et al.*, 2010; Norhayati *et al.*, 2016; Rovinaru and Pasarin, 2020). The bacterial fermentation produces hydrogen, carbon dioxide, methane, and short-chain fatty acids (SCFA) such as acetic, propionic, and butyric. The production of SCFA results in the decrease in pH, thus leading to the possible reduction

in the numbers of pathogenic bacteria such as *Clostridium perfringens*, *E. coli*, and *Salmonella* spp. (Gibson *et al.*, 1995). Prebiotics can be found naturally in fruits and vegetables. Extensive studies have proven that many oligosaccharides extracted from fruits such as orange peel, dragon fruit flesh, and guava fruit flesh can support the growth of probiotic bacteria (Gómez *et al.*, 2014; Mohd Adzim Khalili *et al.*, 2014; Wichienchot *et al.*, 2010; 2011; Thuaytong and Anprung, 2011). Besides the prebiotic effect, dragon fruit (*Hylocereus undatus*) has also shown other functional properties such as reduced caloric intake and insulinaemia (Mohd Adzim Khalili *et al.*, 2009; Song *et al.*, 2016). This indicates the potential of dragon fruit as a functional ingredient in a variety of food products. However, the deterioration of physical appearance and decomposition by disease of dragon fruit could reduce its value (Mohd *et al.*, 2014).

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Fruit juices from orange, mango, cactus pear, pomegranate, pineapple, açai, and blackberry concentrates have been widely produced into fruit juice powder (Abadio *et al.*, 2004; Tonon *et al.*, 2008; Youssefi *et al.*, 2009; Goula and Adamopoulos, 2010; Ferrari *et al.*, 2012; Zotarelli *et al.*, 2017) due to the perishable nature and limited shelf life of these fruit juices. Fruit powders are convenient, easy to handle, have longer shelf life, suitable for storage and transportation, and can be added into several products such as snacks, beverages, and bakery goods (Etzbach *et al.*, 2020). Therefore, dragon fruit juice can also be powdered to improve its shelf life and product stability.

There are many drying techniques that can be used to produce powders such as freeze, drum, and spray drying; but among these techniques, spray drying has been widely used in many fruits such as sugarcane, tomato pulp, watermelon, dragon fruit, and jussara (Quek *et al.*, 2007; Goula and Adamopoulos, 2008; Nishad *et al.*, 2017; Santana *et al.*, 2018; Yusof *et al.*, 2020) due to its effectiveness under optimum conditions and lower cost, which is 30 to 50 times cheaper than freeze drying (Desobry *et al.*, 1997). However, fruit juice powders produced by spray drying have been reported to have problems in packaging and utilisation due to their stickiness, hygroscopicity, and low solubility (Cano-Chauca *et al.*, 2005). This occurred due to the high content of low molecular weight sugars namely fructose, sucrose, and glucose in fruit juices which exhibited sticky behaviour, thus very prone to caking during storage (Ferrari *et al.*, 2012). One approach to successfully spray dry such products is by adding wall materials which are mostly used to increase the glass transition temperature ( $T_g$ ) of the fruit juice.

The selection of wall materials is very important as they must have high solubility, low viscosity, good emulsifying properties, and bland taste, as well as being non-hygroscopic and low cost (Murúa-Pagola *et al.*, 2009). Previous studies have reported that maltodextrin (MD) and gum Arabic (GA) are the most suitable wall materials that can be used (Gharsallaoui *et al.*, 2007); but GA is more expensive and limited in supply, thus, MD has now become the most commonly and widely used material for spray drying fruit juices (Bhandari *et al.*, 2013). MD with various dextrose equivalents (DE) has been used to encapsulate a variety of fruit juices such as elderberry, red dragon fruit, and raspberry (Bhandari *et al.*, 1993; Murugesan and Orsat, 2011; Yee *et al.*, 2017). Besides MD, resistant maltodextrin (RMD) is also one of the starch-derived products, and its bland taste, soluble fibre, low viscosity, and

non-hygroscopic characteristics are also suitable to be used as wall material to produce high fibre fruit powders. However, this has not yet been proven in any study.

The use of different wall materials and spray drying conditions will produce different physical properties of powders, and the most important characteristics of powder that have been reported are moisture content, solubility, bulk density, hygroscopicity, particle size, and morphology (Bhandari *et al.*, 1997; Grabowski *et al.*, 2006). Therefore, the present work was conducted to investigate the effects of different wall materials, namely RMD and MD, on the aforementioned physical properties of white dragon fruit (WDF) powder produced at optimum conditions. The present work also evaluated their prebiotic activity in order to be applied in the food industry as a new functional ingredient.

## Materials and methods

### *Preparation of dragon fruit juice*

Fresh white dragon fruits of commercial maturity were purchased from a local supplier in Selangor, Malaysia. The fruits were washed, drained, and peeled manually. The pulps were sliced into small cubes, and the juice was extracted using a juice extractor (Santos, France). The seeds were separated and removed through the sieve in the extractor. The juice, which was of 12 °Brix and pH 5.07, was then immediately used for spray drying.

### *Powder production by spray drying*

Spray drying was performed with a pilot-scale spray drier (Model Niro A/S, Gea, Germany) under the following operational conditions: 900 m<sup>3</sup>/min air flow rate; 15,000 rpm rotary atomiser speed; and the feed speed of peristaltic pump (Model BT300-2J) was adjusted based on the outlet temperature. The feed was given at room temperature (25 ± 2°C). Distilled water was fed in the beginning, and used to clean the pipe and atomiser. WDF powders encapsulated with MD and RMD were produced at optimum conditions by setting the inlet temperature at 150 and 153°C, outlet temperature at 75 and 82°C, and wall material concentration at 18 and 20%, respectively. These conditions were chosen following preliminary studies. The feed mixtures containing RMD and MD were homogenised constantly during spray drying to ensure their homogeneity.

### *Physical properties of white dragon fruit powders*

#### *Determination of moisture content*

The moisture content of the powders was determined gravimetrically by oven drying at 105°C until a constant weight was achieved (Kha *et al.*, 2010).

#### *Determination of water activity*

Water activity was determined using a pre-calibrated water activity meter (AQUALAB Series 3 TE, USA) following the manufacturer's instruction manual.

#### *Determination of solubility*

Solubility was determined following the method described by Cano-Chauca *et al.* (2005) with some modifications. Briefly, 100 mL of water and 1 g of WDF powder were homogenised using a Waring blender for 5 min at high speed. The solution was then centrifuged (Heraeus Multifuge 3L, Thermo, Germany) at 3,000 g for 5 min. Next, 25 mL of the supernatant was transferred to a pre-weighed Petri dish, and dried in an oven at 105°C for 5 h. The powder solubility (%) was calculated based on the supernatant dry weight, and compared with its expected dry matter.

#### *Determination of hygroscopicity*

Hygroscopicity was determined following Niro analytical method no. A-14-a prescribed by GEA Group (2005), and a modified version of that as described by Tonon *et al.* (2008). Briefly, 1 g of powder was placed in an airtight desiccator containing NaCl-saturated solution at 25°C (75.29% relative humidity). The powder was collected after 1 w, and weighed. The hygroscopicity was calculated using Eq.1:

$$[\% \text{ Hygroscopicity} = (\% \text{ WI} + \% \text{ MC}) \times 100 / (100 + \% \text{ WI})] \quad (\text{Eq.1})$$

where, MC = moisture content of the powder, and % WI = (sample weight after equilibrium – sample weight) / sample weight × 100.

#### *Determination of bulk density (BD), true density (TD), and intergranular porosity*

The bulk densities were determined following the method described by Cai and Corke (2000). Briefly, 3 g of sample ( $m_0$ ) was poured through a funnel into a 10-mL graduated cylinder, and then tapped 10 times onto a rubber mat from a height of 10 cm. The volume ( $V_0$ ) was read directly from the cylinder, and was used to calculate the bulk density ( $\rho_B = m_0/V_0$ ). True density (TD) was

determined by helium pycnometry in an AccuPyc II 1340 Automatic Gas Pycnometer (Micromeritics, Norcross, USA). Intergranular porosity ( $\epsilon$ ) was calculated using Eq. 2:

$$\epsilon = 1 - (\text{BD} / \text{TD}) \quad (\text{Eq. 2})$$

#### *Determination of particle size distribution*

Particle size distributions of the spray-dried powders were determined using a laser scattering particle size analyser, Mastersizer 2000 (Malvern Instruments Co., Malvern, UK) which was connected to a dry particle feeder to compress air and vacuum. The particle size distribution was monitored during each measurement until constant successive readings were obtained. The particle size was expressed as D (v, 0.5), which was the volume-weighted median diameter.

#### *Scanning electron microscopy (SEM)*

The shape and surface morphology of the spray-dried WDF powders were evaluated using a scanning electron microscope (LEO 1455 VPSEM) attached to an energy dispersive X-ray spectrometer (Oxford INCA Energy 300 EDX, Germany). Briefly, WDF powder was mounted on a specimen stub with double-sided adhesive carbon tape. The specimen was coated with gold in a vacuumed evaporator, and examined at 20 kV with a magnification of 500×.

#### *Determination of glass transition temperature ( $T_g$ )*

The WDF powders were analysed by differential scanning calorimetry (DSC) on an indium-calibrated Mettler Toledo DSC 822e (Mettler Toledo, OH, USA). Thermograms were recorded by placing 10 to 12 mg of each sample in a 40- $\mu$ L aluminium pan, which was sealed and pierced. An empty sealed aluminium pan was used as a reference. The samples were heated from 25 to 100°C at a heating rate of 10°C/min. The average of onset and end set temperatures was reported as the  $T_g$  of each sample, and the experiments were done in triplicate for each sample.

#### *Determination of prebiotic activity of white dragon fruit powder*

##### *Bacterial strains*

Commercial probiotic bacterial strains, *Lactobacillus casei* Shirota (Yakult, Japan) and *Bifidobacterium longum* BB536 (Morinaga Milk Industry, Tokyo, Japan) were obtained from the stock culture of the Laboratory of Microbiology, Faculty of Biotechnology and Biomolecular Sciences, Universiti Putra Malaysia.

### Modified de Man, Rogosa, and Sharpe (MRS) media preparation

The MRS media was modified following the procedure proposed by Dubey and Mistry (1996), wherein 0.5 g of L-cysteine was added to 1 L of MRS solid and liquid to obtain 0.05% L-cysteine MRS media. The prepared media was then autoclaved at 121°C for 15 min. Prior to autoclaving, the liquid medium was distributed among 50 mL Universal bottles to be used in batch culture fermentation. For solid medium, the autoclaved media was poured onto sterile Petri dishes, and left to solidify at room temperature. The liquid and solid media were refrigerated at 2 to 8°C until further use.

### Bacterial counting

Bacterial counting was conducted through colony forming unit (CFU) analysis on MRS agar plates. Samples (0.1 mL) were taken and serially diluted ten times in Ringer solution (Merck, Germany), and then spread evenly on MRS agar plates. The samples (0.1 mL) were taken from each Universal bottle at an initial time ( $T_0$ ) and at 6 h intervals during an incubation period of 24 h. Duplicated MRS agar plates were used for each serial dilution, and incubated at 37°C for 48 h in an anaerobic jar, after which the total colony count was recorded. Each CFU represented a bacterium existing in the diluted sample. The number of CFUs was divided by the product of the dilution factor and the volume of the plated diluted suspension to determine the number of bacteria per mL in the original solution.

### Sample media and culture conditions

One millilitre of overnight culture (*B. longum* BB536 and *L. casei* Shirota) was inoculated into 9 mL of MRS basal broth with the samples (Table 1). The media were then incubated at 37°C for 24 h in anaerobic conditions. The samples were taken for serial dilution and colony counts every 6 h for 24 h.

### Statistical analysis

All analyses were performed in triplicate. The experimental data were analysed statistically by analysis of variance (ANOVA) and paired sample *t*-test. The differences were significant at  $p < 0.05$ .

## Results and discussion

### Moisture content, water activity, hygroscopicity, and solubility of white dragon fruit powders

Table 2 shows the physical properties of the spray-dried WDF powders with different carriers (MD and RMD). The MC and  $a_w$  of the two powders showed significant differences ( $p < 0.05$ ). Nevertheless, they remained within the recommended values, thereby assuring the microbiological stability ( $a_w < 0.60$  and  $MC < 5\%$ ). WRMD powder had lower MC and  $a_w$  than that of MD. Therefore, the powder with the presence of RMD exhibited higher hygroscopicity than that with WMD. The lower the MC and  $a_w$  values, the greater the capacity of the powder particles to adsorb ambient moisture. This finding is consistent with those published by Santana *et al.* (2013) who worked on the spray drying of pequi pulp extract. They reported that the moisture content was inversely proportional to the hygroscopicity value.

Table 2 also shows no significant difference ( $p > 0.05$ ) in solubility (*i.e.*,  $> 90\%$ ) that was found between the two powders produced. This indicated that WDF powders produced could be dissolved easily in water at room temperature. This finding could be attributed to the fact that both wall materials, MD and RMD, are easily dissolved in water (Takeiti *et al.*, 2010). As a result, these two carriers enhanced the dissolution ability of the powders. In addition, solubility is also influenced by hygroscopicity, where solubility increases with increasing hygroscopicity.

### Bulk density, true density, and porosity

The bulk density of WRMD powder was significantly higher than the WMD powder ( $p < 0.05$ ). Bulk density is an important characteristic for the packaging design and transportation volume. It

Table 1. Samples used as substrates in fermentation.

| Sample      | Glu                            | RMD                            | MD                            | WRMD  | WMD  | FOS                            |
|-------------|--------------------------------|--------------------------------|-------------------------------|---|--|--------------------------------|
| Composition | MRS basal<br>broth + 5%<br>Glu | MRS basal<br>broth + 5%<br>RMD | MRS basal<br>broth + 5%<br>MD | MRS basal<br>broth + 5%<br>RMD-coated<br>powder | MRS basal<br>broth + 5%<br>MD-coated<br>powder | MRS basal<br>broth + 5%<br>FOS |

Glu: glucose; RMD: resistant maltodextrin; MD: maltodextrin; MRS: de Man, Rogosa, and Sharpe agar; and FOS: fructooligosaccharides.

Table 2. Physical properties of spray-dried dragon fruit powders produced using resistant maltodextrin (WRMD powder) and maltodextrin (WMD powder).

| Physical property                            | WRMD                       | WMD                        |
|--|----------------------------|----------------------------|
| Moisture content (%)                         | 2.33 ± 0.08 <sup>a</sup>   | 3.86 ± 0.01 <sup>b</sup>   |
| Water activity (a <sub>w</sub> )             | 0.18 ± 0.00 <sup>a</sup>   | 0.27 ± 0.01 <sup>b</sup>   |
| Hygroscopicity (%)                           | 23.43 ± 0.44 <sup>a</sup>  | 15.20 ± 0.22 <sup>b</sup>  |
| Solubility (%)                               | 96.50 ± 1.16 <sup>a</sup>  | 95.30 ± 0.21 <sup>a</sup>  |
| Bulk density (kg/m <sup>3</sup> )            | 728.6 ± 0.03 <sup>a</sup>  | 609.9 ± 0.04 <sup>b</sup>  |
| True density (kg/m <sup>3</sup> )            | 153.65 ± 0.03 <sup>a</sup> | 150.67 ± 0.00 <sup>a</sup> |
| Intergranular Porosity, ε (%)                | 52.00 ± 0.83 <sup>a</sup>  | 59.43 ± 0.12 <sup>b</sup>  |
| Particle size (μm)                           |                            |                            |
| D <sub>10</sub>                              | 13.89 <sup>a</sup>         | 28.08 <sup>b</sup>         |
| D <sub>50</sub>                              | 25.34 <sup>a</sup>         | 49.20 <sup>b</sup>         |
| D <sub>90</sub>                              | 45.39 <sup>a</sup>         | 81.9 <sup>b</sup>          |
| Glass transition temperature, T <sub>g</sub> | 62.56 ± 0.19 <sup>a</sup>  | 63.10 ± 0.59 <sup>a</sup>  |

Values in the same row with different lowercase superscripts are significantly different ( $p < 0.05$ ).

can be expressed as a function of solid density (true density) and porosity, which measures the average density of a large volume of the powder in specific medium. Bulk density is affected by factors such as particle size and shape. Zotarelli *et al.* (2017) reported that the bulk density and porosity of spray-dried mango powder are inversely proportional to its particle size. The interstitial air content between particles increased with increasing size of WMD powder (Table 2). Therefore, a higher volume was occupied, and a lower bulk density was obtained (Goula and Adamopoulos, 2010).

True density corresponds to the solid particles that comprise the powder. Both samples exhibited similar true density values (Table 2). The true density of WRMD powder was slightly higher than WMD powder. Meanwhile, porosity is the percentage of spaces between particles. Results showed that the porosity of WRMD powder was lower than that of WMD powder. Tonon *et al.* (2010) revealed that the larger number of spaces between the particles could be attributed to the higher quantity of available oxygen. As a result, the compound being protected may be rapidly lost due to the degradation reactions. The porosity of açai juice powder produced with MD DE10 (Tonon *et al.*,

2010) was found to be significantly higher than WRMD and WMD powders produced in the present work. This indicates that RMD is a better wall material than MD for microencapsulation. Cai and Corke (2000) reported that hygroscopicity increases with decreasing T<sub>g</sub> (Table 2), because lower molecular weight implies shorter chains and more hydrophilic groups. The results of the present work are in agreement with their report.

#### *Particle size distribution of white dragon fruit powder*

The diameters of WDF particles coating with MD and RMD ranged from approximately 17.13 to 120.7 μm and 8.2 to 83.7 μm, respectively. Table 2 shows that the median D<sub>50</sub> of WRMD powder was 25.34 μm. The median D<sub>50</sub> of WMD powder was higher than WRMD powder due to the molecular size of the wall material (Tonon *et al.*, 2010), and higher use of RMD concentration. Janiszewska and Witrowa-Rajchert (2009) revealed that the diameter of spray-dried materials is affected by atomisation, material properties, raw material concentration and viscosity, and drying conditions. They also reported that the particle size of the powder with 25% MD was higher than the powder

with 30% gum Arabic. In addition, Crosby and Marshall (1958) reported that small particles have thicker shells than large particles as characterised by greater bulk density. This observation was confirmed in the present work. As shown in Table 2, the particles and bulk density of WRMD powder were smaller and larger, respectively, than those of WMD powder.

#### Glass transition temperature ( $T_g$ )

The  $T_g$  values of WDF powders were determined to investigate the critical conditions for powder storage, namely, the temperature at which the powders are not susceptible to deteriorative changes such as stickiness, caking, and collapse. As shown in Table 2 and Figures 1a and 1b, WMD powder showed a higher  $T_g$  value than that of WRMD powder. However, no significant difference was found between the  $T_g$  values of both powders.

In addition, the  $T_g$  values of both powders were still above the ambient temperature (Reid and Levine, 1991). The higher the  $T_g$  value, the more stable the powder is. The reason is that temperatures above  $T_g$  change the physical properties of the powders, such as an increase in the free molecular volume. Free volume is defined as a small amount of unfilled volume that is available for free movements of the solid particles (Windle, 2001).

The lower molecular weight (Kishimoto *et al.*, 2009) of the compounds in the powder mixture might decrease the  $T_g$  value (Roos *et al.*, 1996). This result is in agreement with the findings of Tonon *et al.* (2009) who reported that açai juice produced with MD DE10 showed higher  $T_g$  than that produced with MD DE20. In addition, the  $T_g$  values of WDF powders were also affected by the solubility of MD and RMD, in which the  $T_g$  value of the powder increased with increasing carrier solubility.

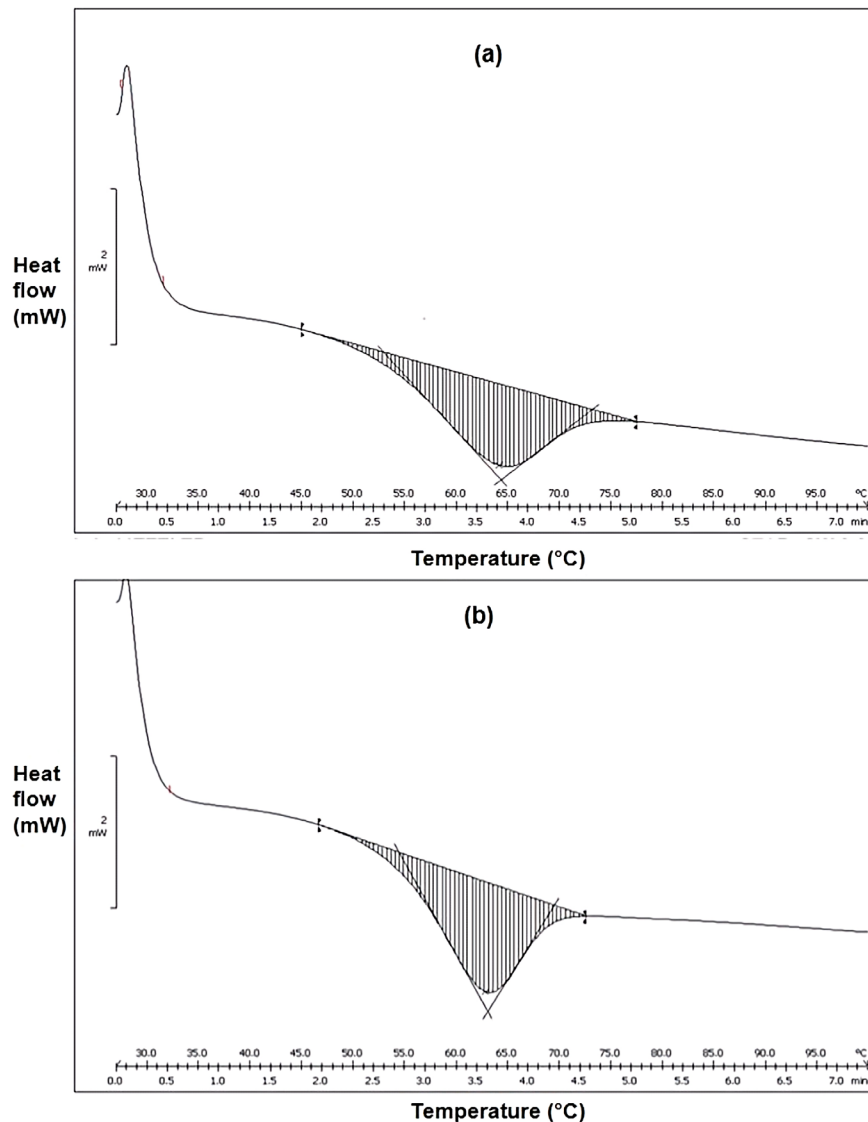


Figure 1. Thermogram of spray-dried dragon fruit powders produced using (a) maltodextrin, and (b) resistant maltodextrin.

### Morphology of white dragon fruit (WDF) powders

The scanning electron microscopy (SEM) micrographs (Figures 2a and 2b) showed that WDF powders coated with RMD and MD were significantly different in terms of size and shape. Structural analysis revealed that WMD powder was spherical in shape with many surface indentations and cracks, whereas WRMD powders were spherical in shape, smooth surfaces, with less shrinkage and dents. Amin (2009) produced red dragon fruit powders coated with RMD and MD as wall materials, and observed similar shapes and morphology. The concavities and cracks in WMD powder can be associated with the rapid evaporation of the liquid droplets during the spray drying process (Rosenberg *et al.*, 1985), or the effects of the wall material that inhibit the formation of a supporting structure during water evaporation (Favaro-Trindade *et al.*, 2010).

As mentioned earlier, WRMD powder showed smooth surfaces with less dents. This may be attributed to the structure of the wall material that may increase the plasticising properties of RMD for shrinkage prevention and material protection. As shown in Figures 2a and 2b, the small particles strongly adhered to the surface of the large particles, thus resulting in the formation of many agglomerates. This may be attributed to the static

electrical effects and van der Waals forces. Walton and Mumford (1999) demonstrated that particles with an agglomerate structure are composed of individual grains of materials bound together by sub-micron dust of the same material, and that these agglomerated structures may bind to each other as well. Zhang *et al.* (2000) reported that plasticisers play important roles in the formation of spherical microcapsules with smooth surfaces. These results imply that RMD encapsulated the core material better than MD.

### Fermentation of substrates by *L. casei* Shirota and *B. longum* BB536 in pure culture

The present work investigated the ability of WRMD powder to support the growth of *L. casei* Shirota and *B. longum* BB536 (Figures 3a and 3b). Of the six substrates tested (glucose, RMD, MD, WMD, FOS, and WRMD), MRS medium supplemented with 5% WRMD provided the highest bacterial counts for both types of probiotic bacteria. The maximum value indicated for *L. casei* Shirota increased from  $6.05 \pm 0.11$  to  $8.75 \pm 0.00 \log_{10}$  CFU/mL after 18 h of fermentation. For *B. longum* BB536, the maximum value increased from  $6.04 \pm 0.26$  to  $8.56 \pm 0.10 \log_{10}$  CFU/mL after 12 h of fermentation. Results showed a significant difference ( $p < 0.05$ ) from the medium supplemented

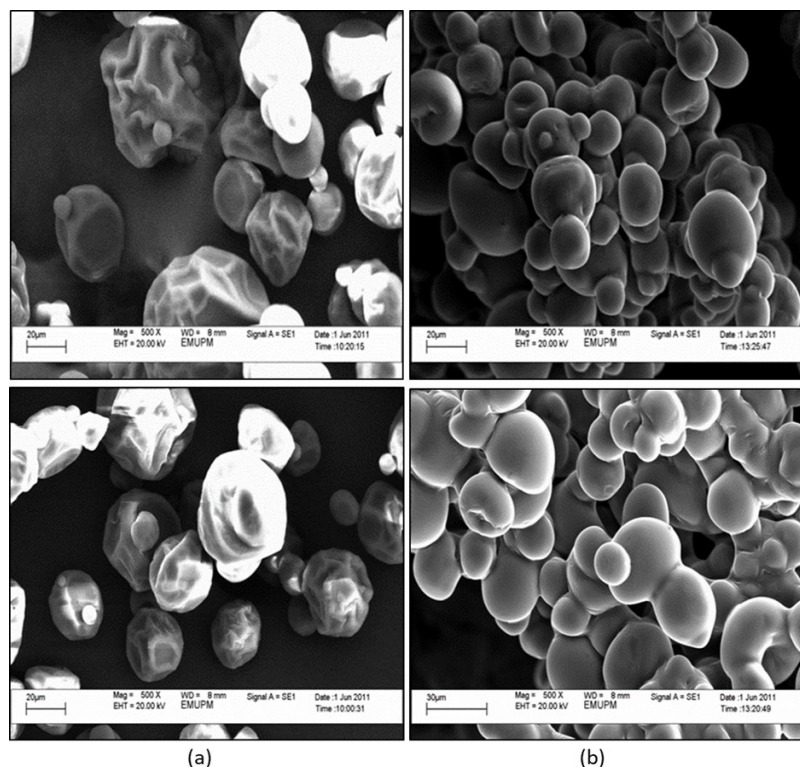


Figure 2. Scanning electron micrographs (500×) of microcapsules of spray-dried dragon fruit powders produced using (a) maltodextrin, and (b) resistant maltodextrin.

with 5% glucose, which was used as a control sample. *L. casei* Shirota and *B. longum* BB536 increased in number to  $8.21 \pm 0.08$  and  $8.07 \pm 0.07$   $\log_{10}$  CFU/mL, respectively, after 18 h of fermentation with glucose sample. Beyond 24 h fermentation with glucose, the number of *L. casei* Shirota still increased to  $8.54 \pm 0.18$   $\log_{10}$  CFU/mL, but for *B. longum* BB536, the number decreased to  $7.85 \pm 0.18$   $\log_{10}$  CFU/mL.

In the present work, both types of probiotic bacteria followed a similar pattern of bacterial growth (Figures 3a and 3b), in which the number of cells doubled during the exponential phase from 0 to 6 h, after which, the growth rate was maintained or decreased during the stationary phase, before finally reaching the dead phase after 18 to 24 h. A probable reason for this pattern is the depletion of nutrients

after a certain period of fermentation. The results agree with Droop (1975), who observed that the bacterial growth rate decreases once the nutrients needed for cell metabolism are exhausted.

When RMD and MD were used as substrates, the growth of both tested bacteria showed no significant difference ( $p > 0.05$ ) from 0 to 18 h, but beyond 18 h fermentation, the cell growth of *L. casei* Shirota in RMD decreased to  $7.99 \pm 0.11$   $\log_{10}$  CFU/mL. However, the maximum cell growth of *B. longum* BB536 in both media containing RMD and MD was shown at 24 h fermentation. Figures 3a and 3b indicated that the cell growths of both bacteria cultivated with WMD and WRMD at 6 to 18 h were significantly ( $p < 0.05$ ) higher than RMD and MD media. The significant difference was probably due to the oligosaccharides of white dragon fruit juice

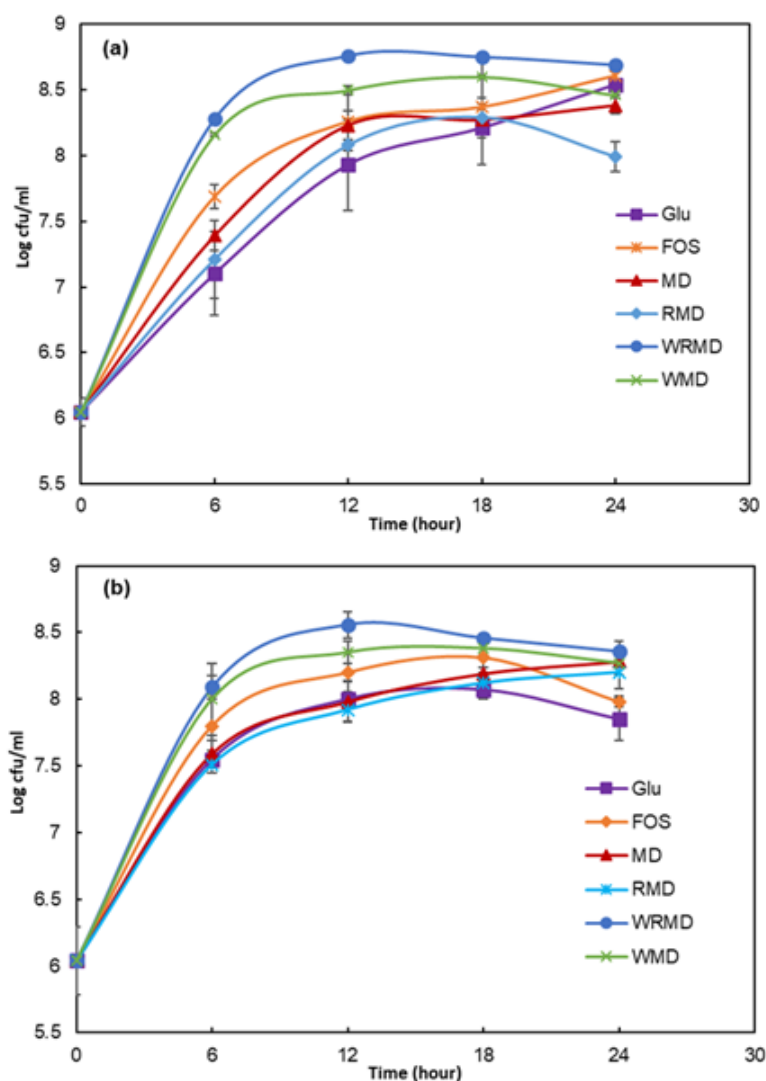


Figure 3. Growth curve of (a) *L. casei* Shirota and (b) *B. longum* BB536 in media containing six different substrates against time. Glu: glucose; FOS: fructooligosaccharides; MD: maltodextrin; RMD: resistant maltodextrin; WMD: white dragon fruit coating with MD; and WRMD: white dragon fruit coating with RMD.



which were reported to have a prebiotic effect (Wichienchot *et al.*, 2011).

However, when both probiotic bacteria cultivated with WRMD was compared with WMD, the cell growth did not differ significantly ( $p > 0.05$ ) at 6 to 18 h fermentation. Even though there was no significant difference, the total number of probiotic bacteria for WMD was lesser than that of WRMD. The smaller difference between the total counts for WMD and WRMD was probably due to differences in the molecular structures of the carriers encapsulating WDF juice, namely MD and RMD, respectively.

MD is a polysaccharide produced from hydrolysed starch, and a mixture of polymers of D-glucose units linked by  $\alpha$  (1-4) or  $\alpha$  (1-6) glycosidic bonds (Wang and Wang, 2000). RMD, an indigestible dextrin produced by treating corn starch with acid, enzymes, and heat, has a similar composition to MD, but also contains  $\alpha$  (1-2) and  $\alpha$  (1-3) linkages and levoglucosan, which aid partial hydrolysis by human digestive enzymes. Reportedly, approximately 90% of RMD reaches the large intestine, where half of this amount is metabolised by intestinal bacteria (Flickinger *et al.*, 2000). Hopkins *et al.* (1998) also reported that RMD can effectively promote the growth of beneficial bacteria.

Figures 3a and 3b also show that the prebiotic nature of FOS was significant ( $p < 0.05$ ), and supported the growth of *B. longum* BB536 and *L. casei* Shirota, where the cell growth increased from  $6.05 \pm 0.11$  to  $8.61 \pm 0.05 \log_{10}$  CFU/mL, and from  $6.04 \pm 0.26$  to  $8.31 \pm 0.11 \log_{10}$  CFU/mL, respectively. This result was consistent with Rycroft *et al.* (2001) who reported that FOS increased the number of lactobacilli. The findings of the present work also agree with Bielecka *et al.* (2002) who reported that a majority of bifidobacterium species (mostly *B. longum* and *B. animalis*) use FOS. Although FOS can stimulate probiotic growth, the present work showed that FOS was less effective than WRMD or WMD. This result is probably due to the composition of WRMD, where WDF itself contained oligosaccharides that have been reported to stimulate the growth of *L. delbruekii* and *B. bifidum* better than inulin (Wichienchot *et al.*, 2011). In addition, WRMD was encapsulated with RMD, which can be partially fermented by colonic bacteria (Nathaniel *et al.*, 2008). It is also possible that the combination of two compounds in WRMD (RMD and WDF juice) can produce a synergistic effect in the human colon.

Results also indicated that the media containing WRMD was better for probiotic bacteria

than WMD. As a dietary fibre, RMD has several beneficial physiological properties. Unlike FOS, RMD is fermented slowly in the large intestine; thus, it does not cause flatulence. Therefore, adding RMD as a wall material in the production of WDF powder may improve intestinal regularity without causing any discomfort. This may be the most attractive quality of RMD because it makes the consumers feel the difference if it is taken as a supplement with their regular meals. Several clinical studies have indicated that if RMD is ingested with meals, it can moderate the postprandial rise in blood glucose levels, and regulate insulin secretion (Unno *et al.*, 2002).

## Conclusion

RMD was found to be an effective carrier for the spray drying of WDF juice to obtain powders with high fibre. RMD decreased water activity and moisture content more than MD. Results showed that the bulk density of WRMD powder was higher, and the particle size was smaller than WMD powder. This finding indicates that WRMD powder needs lesser space to load a specified volume for packaging and transportation. The surface morphology of WRMD powder was also smoother than WMD powder, which exhibited shrinkage and dents. Based on the data obtained through *in vitro* pure culture fermentation, the amount of cell growth for all media samples significantly increased within 24 h of fermentation. The highest bacterial count for *L. casei* Shirota and *B. longum* BB536 was recorded when WRMD was used as a substrate, and significant difference was observed as compared to the commercial prebiotic, FOS. The ability of WRMD to support the growth of probiotic bacteria has proven that WRMD is a potential source of new prebiotic for the food industry.

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