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Microencapsulation of orange-fleshed sweet potato (*Ipomoea batatas*) carotenoid extract by spray-drying with maltodextrin and whey protein concentrates

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KEYWORDS

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ABSTRACT

Orange-fleshed sweet potato (*Ipomoea batatas*) carotenoids were encapsulated in maltodextrin and whey protein concentrates by spray-drying to promote dispersibility in water and looked for the best encapsulant concentration in both encapsulants. The moisture content, wettability, hygroscopicity, color characterization, cold water solubility, and encapsulation efficiency were analyzed for encapsulant concentration of 10%, 20%, and 30% (w/v). The encapsulant concentration of 20% showed the best result with 6.09% moisture content, 11.07 hygroscopicity, 51 s wetting time, 94.50% cold water solubility, and 81.52% encapsulation efficiency for maltodextrin encapsulant. While whey protein concentrates encapsulant gave the result of 6.35% moisture content, 12.44% hygroscopicity, 148.8 s wetting time, 93.13% cold water solubility, and 82.02% encapsulation efficiency. The diffractogram from XRD showed that microcapsule had the amorphous phase dominant and indicated high solubility in water matrix. The microcapsule using maltodextrin encapsulant has smoother and more spherical morphology than microcapsule using whey protein concentrate encapsulant. The color characterization of 30% was lighter, less red, and less yellow than the others based on L^*, a^*, and b^* value respectively. The carotenoid with 20% encapsulant concentration was well encapsulated enough and the addition of more than that did not produce significantly better results.

Introduction

One of the food quality parameters which should be first concerned and be the basic of consumer acceptance is the color. The producers need to look for appropriate color of their product and ensure the stability of the color (Akhavan and Jafari, 2017).

Food dyes are generally categorized into natural dyes and synthetic dyes. Synthetic food dyes had been widely used in the past, but consumer interest is shifting to the natural one because of the toxicity. It was reported that synthetic dyes are carcinogenic when it is consumed in high frequency and more than acceptable daily intake index (Caro et al., 2012). Otherwise, natural dyes become food additive in great demand because of its health potential and consumer concern about the toxicity of synthetic dyes (Rodriguez-Amaya, 2016).

Carotenoid is one of natural food dyes which can give red-orange color. It is isoprenoid pigment and bioactive compound which is widely found in fruits and vegetables. Beside coloring the food, carotenoid also plays good role in health and can prevent some chronic diseases (Khalid et al., 2019).

Orange-fleshed sweet potato (*Ipomoea batatas*) has appealed much attention because of its high carotenoid content and being a good provitamin A resource in developing country, including Indonesia. This crop has been an important industrial crop and food resource that contains beta carotene, starch, minerals, food fibers, and vitamins (Kourouma et al., 2019). Takahata et al. (1993) reported that beta carotene content from 22 orange-fleshed sweet potato planted in varies condition is about 1.1 – 26.5 mg/g.

Orange-fleshed sweet potato also good to be developed as natural dyes because of its high productivity. Based on BPS data, the national productivity of sweet potato continues to increase and reach the value of 160 quintal/hectare in 2015 (www.bps.go.id).

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Carotenoid like another natural pigment is unstable and easy to be degraded by light, oxygen, and heat. Besides, almost all the natural dyes have limitation in food application due to their hydrophobic characteristic (Assadpour and Mahdi Jafari, 2019).

Microencapsulation is a technique used in food industry to increase stability and dispersity of bioactive compound (core material) in water matrix by covering it with encapsulant (Paulo and Santos, 2017). This small droplet size can preserve more surface area for material to interact with water molecule. The higher interaction can increase the dispersity in water. Moreover, encapsulant also protects the core material from light, oxygen, and heat.

Orange-fleshed sweet potato potential as the natural food colorants has been reported by Ginting (2013). Carotenoid microencapsulation has also been extensively studied, both from standard chemical compounds and extracts from natural ingredients. However, microencapsulation of carotenoid extracts from orange-fleshed sweet potato has not been studied to the best of our knowledge. The objective of this study was to extract orange-fleshed sweet potato carotenoids as natural colorants to be microencapsulated by spray drying technique using maltodextrin and whey protein concentrate encapsulant to promote dispersibility in water and expand its application in the food sector.

Research Methods

Materials

Orange-fleshed Sweet Potato (*Ipomoea batatas*) was obtained from Magetan, Indonesia. Maltodextrin DE 10 – 12 was purchased from Lihua Starch Co., Ltd. (Qinghuadao, China). Isopro Whey Protein was purchased from PT KSM Mirota (Yogyakarta, Indonesia). β-carotene standard (C9750) and tween 20 surfactant were obtained from Sigma-Aldrich.

Carotenoid Extraction

Carotenoid extraction was done based on Machmudah and Goto (2013) with modifications. Orange-fleshed sweet potato was peeled and thinly sliced. The sliced potato was dried in a cabinet dryer at 50^{o}C for 18 hours. The dried potato was ground using HR-2115 dry mill blender (Philips, Netherlands) and sieved using 40-mesh-sized standard sieve. Sweet potato powder was extracted with 96% ethanol (1:3 w/v) with mixing until the color change of solvent was saturated. Ethanolic extract was partitioned using n-hexane (1:1 v/v). Solven evaporation was done using RV-10 Rotary Evaporator (IKA, Germany) at 40 ^{o}C. Carotenoid extract was solubilized in 10 mL refined palm oil by stirring for 30 min at room temperature. Solubilized carotenoid extract in oil was cooled under light protection at 4 ^{o}C and prepared to be oil phase in emulsification.

Emulsification

The emulsion was formed based on Medeiros et al. (2019). Tween 20 (1.5% w/v) was solubilized in 90 ml distilled water by stirring for 30 min (AF1) and was solubilized in 100 mL distilled water with encapsulant addition (AF2) by stirring for 1 h at ambient temperature. The emulsion was formed by homogenizing aqua phase 1 (AF1) with oil phase (OP) at 17000 rpm for 10 min using T18 basic Ultra-Turrax (IKA, Germany). Obtained emulsion was then added by aqua phase 2 (AF2) using the same condition to form the final emulsion. The encapsulants used in this study were maltodextrin and whey protein concentrate with the concentration of 10%, 20%, and 30% (w/v) for each of them.

Spray-Drying

The final emulsion was immediately fed to the Mini Spray-Dryer B290 (Buchi, Switzerland). The condition was set on inlet temperature of 170^{o}C, aspirator setting of 70%, airflow of 536 L/h (45 on the nozzle rotameter scale), and feed flow of 4.5 ml/min (pump setting of 15%).

Moisture Content

The moisture content was determined thermogravimetrically using a MB120 Moisture Analyzer (Ohaus, China) by drying one gram of the spray-dried carotenoid extract.

Hygroscopicity

Hygroscopicity was calculated as the moisture absorption was determined based on Etzbach et al. (2020) method with modifications by exposing the powders to humid air with 81% relative humidity. A 0.5 g of powder was weighted in aluminum dish and placed in a desiccator containing 200 mL of a saturated solution of Na_2SO_4 at 25^{o}C.

Wettability

Wettability was determined using the method described by Erbay and Koca (2015) with modifications. A 0.075 g of microcapsule was sprinkled over the surface of 100 mL distilled water in 250 mL beaker glass at ambient temperature without agitation. The time taken for

the powder particles to sediment or sink or submerse below and disappear from the surface of water was measured and used for a relative comparison of the extent of wettability between the samples.

Cold Water Solubility

Cold water solubility was determined according to Eastman and Moore (1984). One gram powder was suspended in 100 mL distilled water by stirring at 300 rpm for 30 min. The suspension was centrifuged with a DM0636 centrifuge (DLAB Scientific, USA). A 25 mL of aliquot was taken from supernatant and placed in ceramic evaporating dish to be evaporated using water bath evaporator. The precipitate was dried overnight at 110 ^{o}C to a constant weight. Cold water solubility was calculated according to the following equation:

$$CWS\ (\%) = \frac{4\ x\ weight\ of\ solid\ in\ 25\ mL\ of\ supernatant}{weight\ of\ powder\ sample} x100 \quad \ldots(1)$$

Color Analysis

The color parameter *L**, *a**, and *b** were measured with a Chroma Meter CR-400 (Konika Minolta, Japan) by placing the microcapsule powders in petri dish until the surface of petri dish was covered by a powder layer of 100 mm thickness.

X-Ray Diffraction

The microcapsule and encapsulant powder were analyzed on a MiniFlex benchtop X-Ray Diffractometer (Rigaku, Japan) with a scanning rate of 8^{o}/min. The diffraction spectra were recorded at the diffraction angel of 2θ from 4^{o} to 80^{o} at room temperature. Relative crystallinity (RC) was calculated according to the following equation:

$$RC\ (\%) = \frac{area\ of\ the\ crystalline\ peaks}{total\ area\ of\ crystalline\ and\ amorphous\ peaks} x100 \quad \ldots(2)$$

Scanning Electron Microscopy

To observe the morphology of the microcapsules, all samples were mounted on aluminum stubs before being gold-coated using MC1000 Ion Sputter Coater (Hitachi, Japan) and viewed under SU3500 Scanning Electron Microscopy (Hitachi, Japan) with 5000× magnification.

Encapsulation Efficiency

Encapsulation efficiency was calculated according to the following equation:

$$EE\ (\%) = \frac{total\ carotene - surface\ carotene}{total\ carotene} x100 \quad \ldots(3)$$

Total carotene and surface carotene were measured according to Loksuwan (2007) method. Total carotene was measured by dispersing 50 mg of powder in 25 mL hexane. The solvent was shaken at 500 rpm for 10 min. The hexane phase was measured at 450 nm with Visible Spectrophotometer (Thermo Fisher, USA). As for determining surface carotene, 50 mg of powder was dispersed in 25 mL hexane and shaken at 100 rpm for 15 s. The powder particle was centrifuged at 1000 g for 1 min. The supernatant was measured at 450 nm with visible spectrophotometer (Thermo Fisher, USA). Concentration of beta carotene was used in encapsulation efficiency measurement after plotting absorbance vs concentration as beta carotene standard curve.

Statistical Analysis

Statistical analyses were performed using SPSS software (ver. 22.0 for Windows, SPSS Inc., Chicago, IL). Data were analyzed by ANOVA and Duncan post hoc test. Statistical significance was set at $p < 0.05$.

Results and Discussion

Moisture Content

The moisture content could be shown in the Table 1 and Table 2 ranging from 4.94 to 7.02%. The moisture content of the microcapsules depends on encapsulant type and concentration ($P < 0.05$). Higher encapsulant concentration gave lower moisture content. The higher encapsulant concentration had more solid particle in the spray-dryer feed and less amount of water which would be evaporated. It led to the decrease of moisture content in microcapsules. A similar result was obtained by Kha et al. (2010) who found the lowest moisture content of Gac fruit aril powder was obtained from the highest encapsulant concentration among 10%, 20%, and 30% (w/v). However, Goula et al. (2010) found the inverse that higher encapsulant concentration gave the higher moisture content. The higher concentration of encapsulant produced the viscous mixture which makes the water difficult to diffuse.

Table 1. Physical Properties of Microcapsule with Maltodextrin Encapsulant

Samples	Moisture Content (%)	Hygroscopicity (%)	Wettability (s)	Cold Water Solubility (%)	Color Characteristics		
					*L**	*a**	*b**
M10	7.02 ± 0.17[a]	10.33 ± 0.16[a]	58.8 ± 0.17[a]	93.49 ± 0.73[a]	85.71 ± 0.16[a]	-5.80 ± 0.01[a]	40.42 ± 1.09[a*]
M20	6.09 ± 0.13[b]	11.07 ± 0.04[b]	51.0 ± 0.33[b]	94.50 ± 1.76[a]	86.74 ± 0.16[a]	-6.06 ± 0.02[a]	37.70 ± 0.02[a]
M30	4.94 ± 0.05[c]	11.87 ± 0.05[c]	40.5 ± 0.50[c]	94.87 ± 1.44[a]	88.68 ± 0.29[b]	-6.54 ± 0.10[b]	20.95 ± 1.43[b]

*Mean values within a column followed by the same letters are not significantly different at $p < 0.05$ according to Duncan's Multiple Range Test.

Table 2. Physical Properties of Microcapsule with Whey Protein Concentrates Encapsulant

Samples	Moisture Content (%)	Hygroscopicity (%)	Wettability (s)	Cold Water Solubility (%)	Color Characteristics		
					*L**	*a**	*b**
W10	6.99 ± 0.24[a]	11.90 ± 0.03[a]	175.7 ± 1.67[a]	92.30 ± 1.96[a]	86.09 ± 0.45[a]	-5.92 ± 0.03[a]	40.54 ± 1.56[a*]
W20	6.35 ± 0.01[b]	12.44 ± 0.03[b]	148.8 ± 1.83[b]	93.13 ± 1.66[a]	86.84 ± 0.10[a]	-6.06 ± 0.04[a]	37.06 ± 0.12[a]
W30	5.72 ± 0.03[c]	12.84 ± 0.05[c]	118.0 ± 0.33[c]	95.38 ± 1.80a	88.13 ± 0.44[b]	-6.41 ± 0.06[b]	23.26 ± 0.46[b]

*Mean values within a column followed by the same letters are not significantly different at $p < 0.05$ according to Duncan's Multiple Range Test.

Lingua et al. (2020) obtained the explanation that the encapsulant concentration is proportional to the moisture content at low temperature under 140 ^{o}C while at high temperature over 160 ^{o}C, the moisture content decreases as the encapsulant concentration increases due to the more evaporation energy making the powder more dried. It was reported that a 30% concentration of maltodextrin as wall material of blueberries phenolic extract at 160 ^{o}C spray drying inlet temperature had a lower moisture content than the 20% one while an inverse result was found in lower temperature (140 ^{o}C).

The mean value of microcapsule moisture content using maltodextrin and whey protein concentrate respectively were 6.02% and 6.35%. The higher moisture content of whey protein concentrate encapsulant is due to the strong ability of the protein to bind water (Shi et al., 2013).

Hygroscopicity

Hygroscopicity is the ability of the microcapsules to bind water molecule from the surrounding air. The higher encapsulant concentration made the microcapsule more hygroscopic. The higher encapsulant concentration has lower moisture content which makes the bigger water concentration gradient between particle and surrounding air. This gradient was the driving force of moisture absorption of particles. The hygroscopicity calculated by the amount of absorbed water in particle was higher in the higher water concentration gradient. This result was confirmed by the result of Ferrari et al. (2011). The hygroscopicity of whey protein concentrate encapsulated powder was higher than maltodextrin encapsulated powder due to crystallinity relative of whey protein concentrate which was lower than maltodextrins. Material having low crystallinity relative was amorphous phase dominant and would adsorb more moisture from surrounding.

Wettability

Wettability indicates how fast the microcapsule being wet due to the absorption of water molecule by the powder. The time needed to wet the powder decreases as the encapsulant concentration increase in both of maltodextrin and whey protein concentrate encapsulant ($p < 0.05$). It showed that the higher encapsulant concentration led to the higher wettability. Erbay and Koca (2015) said that particle size was one of the important factors affecting wettability. The larger particle size has irregular and agglomerated shape with more space in interstices of particle enhancing the penetration of water (Schuck, 2011). The low wettability of powder using whey protein concentrate encapsulant was agreed with the result of Erbay and Koca (2015).

Cold Water Solubility

The cold-water solubility of all treatments showed the value >92%. This indicated that all samples are water soluble due to the binding ability of encapsulant stabilizing the powder dispersion in water. All concentrations in maltodextrin and

whey protein concentrate encapsulants are not significantly different ($p < 0.05$).

Color Characteristic

Among the samples, microcapsule with 30% encapsulant concentration has a significant color difference. The higher *L**, more negative *a**, and lower *b** values showed that the 30% encapsulant concentration is lighter, less red, and less yellow than the others respectively. It was due to the excess of encapsulant which made the carotenoid color inside faded. The physical appearance of 30% encapsulant concentration was white dominant because the yellow color in the core was covered by whiter encapsulant as the wall material.

X-Ray Diffraction

Figure 1 showed diffuse and broad peak of sample diffractograms. This pattern indicated that all samples has more amorphous phase (Caparino et al., 2012). The amorphous structure on the microcapsule powder was produced from the rapid evaporation in the spray drying step (Barclay et al., 2010). The microcapsule has lower relative crystallinity than pure encapsulant due to replacement of interaction between chains of encapsulant by hydrophobic interactions between the encapsulant and carotenoid extract (Medeiros et al., 2019). Carotenoid extract like other herbal substance is more wanted to have an amorphous structure to give greater protection for encapsulated substance as well as more soluble and easy-handling properties (Drusch, 2007).

Scanning Electron Microscopy

According to the Figure 2, higher encapsulant concentration led to the increase of particle size and the decrease of particle homogeneity. At higher concentration, some of fine particles were attached to the bigger ones. The various size of particles caused the attachment force and agglomeration among particles. As seen in the picture, the lower concentration also had smoother surface, while the higher concentration had some cracks and dents on the surface negatively affecting the flow ability and reconstitution properties (Zhang et al, 2018). Particles with smoother surface are likely used in food application. Microcapsule with maltodextrin encapsulant (a-c) showed the smoother and more spherical particle than microcapsule with whey protein concentrate encapsulant (d-e). The shrunken and irregular surface of microcapsule with whey protein concentrate was formed due to the presence of fat milk on WPC (Erbay and Kocha, 2015).

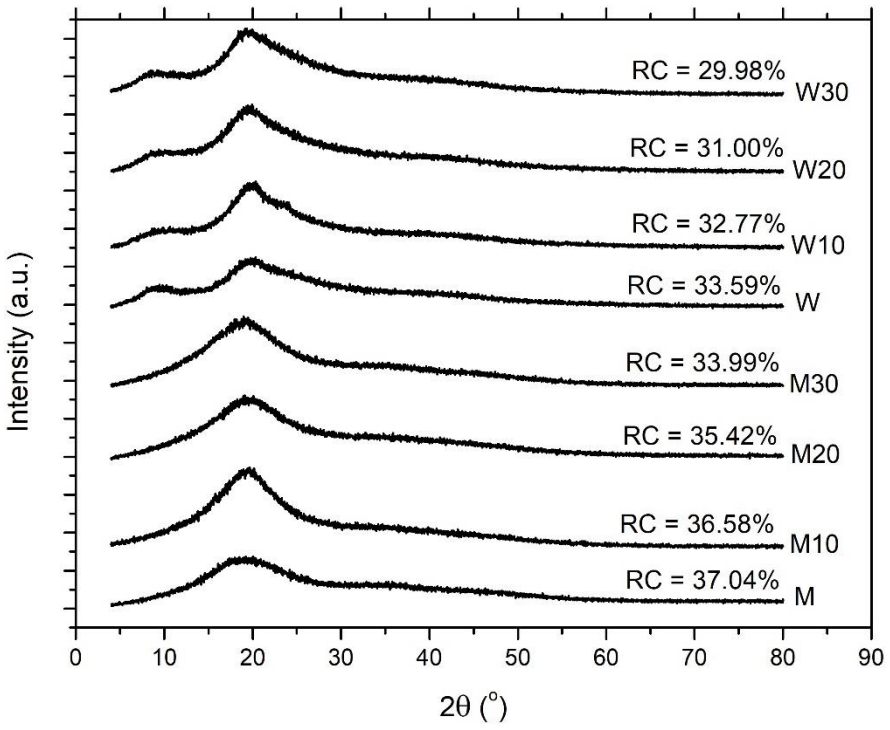

Figure 1. XRD diffractograms of encapsulants and microcapsules

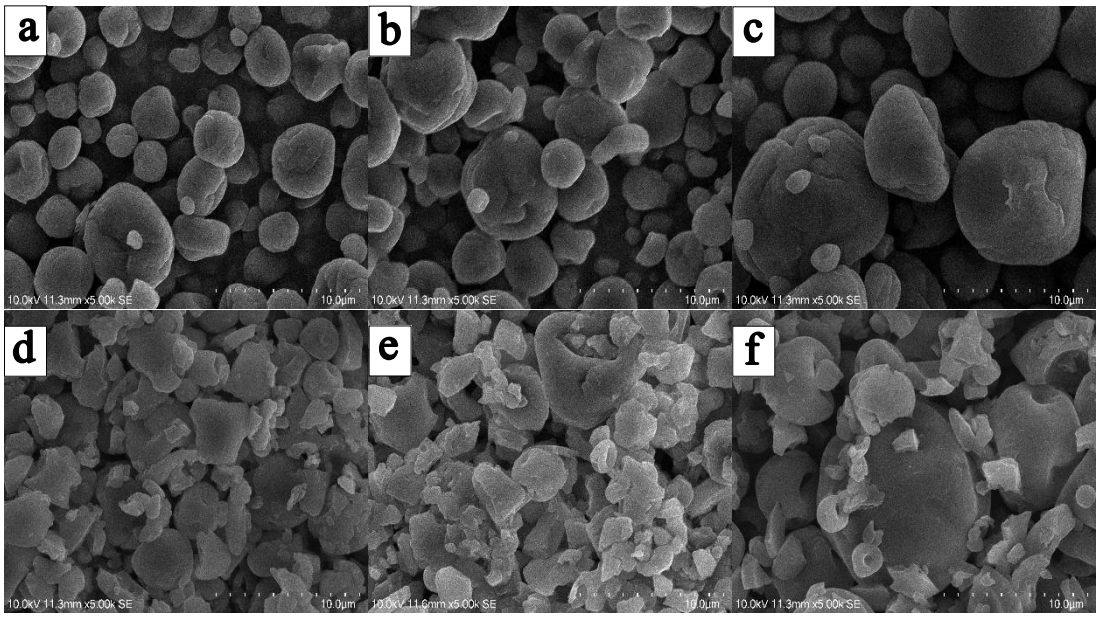

Figure 2. Morphology of microcapsules using encapsulant of maltodextrin with 10% (a), 20% (b), 30% (c) concentration and whey protein concentrates with 10% (d), 20% (e), and 30% (f)

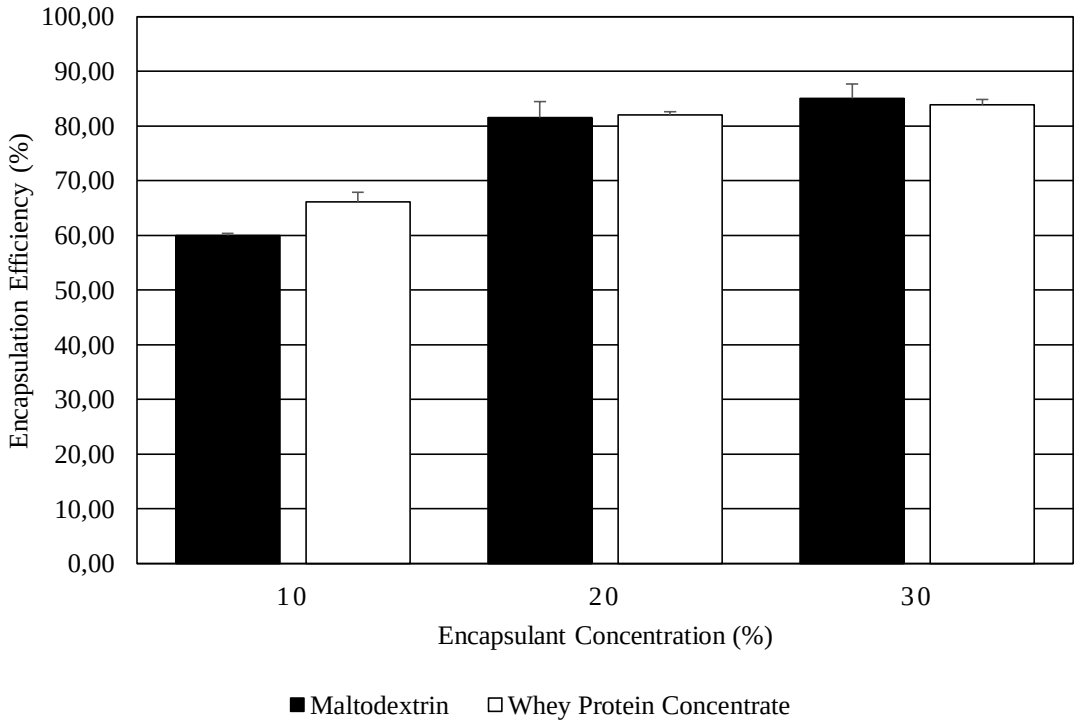

Figure 3. Encapsulation efficiency of microcapsules with maltodextrin and whey protein concentrates encapsulant

Encapsulation Efficiency

As shown in the Figure 3, the encapsulation efficiency was significantly affected by encapsulant concentration ranging from 59.96% to 84.99% ($p<0.05$). Higher encapsulant concentration led to higher encapsulation efficiency. Etzbach et al. (2020) concluded that the particle size of powder affects the encapsulation efficiency. The fine powders had more surface area and made the value of surface carotene higher. It resulted in lower encapsulation efficiency. In this study, the particle size of powder can be confirmed from the SEM micrograph. The encapsulant concentration of 10% showed smaller homogenous particles than

the others which showed the smallest encapsulation efficiency.

The picture showed that at a concentration of 20% encapsulant, the carotenoid was well encapsulated enough. The addition of encapsulant did not increase the encapsulation efficiency significantly. Duncan's Multiple Range Test (DMRT) showed that concentrations of 20% and 30% are not significant difference ($p < 0.05$). The limit might be related to the ability of encapsulant type. Maltodextrin which usually be used as a secondary wall material (a filler) easily becomes crystalline form due to their low glass transition temperature. The compact structure of wall material can be disrupted and produce agglomerated powder which can lower the efficiency (Bae and Lee, 2008).

Etzbach et al. (2020) obtained the encapsulation efficiency of goldenberry's spray-dried powder using maltodextrin, modified starch, inulin, alginate, gum Arabic, and the combination thereof ranging from 16.4% to 77.2%. Medeiros et al. (2019) obtained the efficiency of incorporation of carotenoid extract of Cantaloupe Melon using whey protein concentrate (WPC) was 77%. From the data in the picture 3, encapsulant type did not affect the encapsulation efficiency significantly ($p < 0.05$).

Conclusion

The results of this study indicate that carotenoid extract of orange-fleshed sweet potato can be encapsulated by spray-drying technique using maltodextrin and whey protein concentrates. The microcapsules produced were water soluble and easier to be applied in a food matrix. The 20% encapsulant concentration showed the best results for carotenoid extract encapsulation from orange-fleshed sweet potato. The addition of the encapsulant more than 20% did not significantly increase the encapsulation efficiency.

Declarations

Conflict of interests The authors declare no competing interests.

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