Asia-Pacific Journal of Science and Technology: Volume: 24. Issue: 04. Article ID.: APST-24-04-07. Research Article

APST

Asia-Pacific Journal of Science and Technology

https://www.tci-thaijo.org/index.php/APST/index

Published by the Research and Technology Transfer Affairs Division,
Khon Kaen University, Thailand

Effects of type and ratio of carrier on physicochemical properties of microcapsules containing Gac fruit aril

Piyawan Youdee[1] and Varipat Areekul[1,*]

[1]Faculty of Agro-Industry, King Mongkutt's Institute of Technology Ladkrabang, Bangkok, Thailand
[*]Corresponding author: varipat.ar@kmitl.ac.th

Received 13 May 2019
Revised 14 August 2019
Accepted 21 October 2019

Abstract

Gac fruit aril contains high levels of carotenoids, lycopene, and β-carotene. Unfortunately, when exposed to environmental conditions, these bioactive compounds rapidly degrade. Therefore, microencapsulation must be applied in order to reduce the degradation. This study was carried out to determine the effects of the concentration of Gac aril on the efficiency of the process and on the physicochemical properties of the microcapsules. The Gac aril concentrations were varied over a range from 11.1 to 33.3% and over a range of carrier ratios from 1:3 to 1:7. The Gac aril microcapsules were analysed for efficiency and their physiochemical properties. The result indicated that using 20% Gac aril in 3% β-cyclodextrins had significantly produced a higher process yield, total carotenoid contents, encapsulation efficiency, lycopene, and β-carotene contents in the microcapsules than maltodextrin or gelatin ($p < 0.05$). The lowest Moisture content (MC) and Water Activity (A_w) were found at the Gac aril concentration of 25% Gac aril, which had been encapsulated with 20% maltodextrin. The optimum concentration of the carriers was 16.7 % Gac aril with maltodextrin and 20% Gac aril for both gelatin and β-cyclodextrins.

Keywords: Gac aril, Core material concentration, Microencapsulation, β-carotene, Lycopene

1. Introduction

Gac fruits (*Momordica cochinchinensis* Spreng.), botanically classified as family Cucurbitaceae, are traditional fruits grown in Southeast Asia, containing high levels of carotenoids (both β-carotene and lycopene) in the aril, which is the brightly colored flesh covering the seeds [1-2]. Consumption of Gac fruit can increase plasma β-carotene and retinol levels and has been linked with a lower risk of prostate cancer [3-4] and coronary heart disease [5-6]. However, their molecular structures make them very susceptible to degradation when exposed to oxygen, light, moisture, and heat. Carotenoid breakdown and isomerism can occur at high temperatures [7]. Furthermore, they can degrade via the oxidation pathway. Therefore, process conditions should be considered, which can prevent carotenoids from undergoing oxidation and isomerization and which can also improve their solubility to achieve favorable bioavailability.

Encapsulation has been widely used to entrap functional components in a carrier protecting them against deterioration. The mechanism of this protection is to form a carrier-entrapping droplets inside the core of each capsule. In the food industry, microencapsulation, such as solvent dispersion, co-crystallization, interfacial polymerization, and spray drying, are the most potential techniques to protect food ingredients. In addition, as compared to other encapsulation methods, spray-drying is the most common encapsulated method because of its shorter process and lower costs. Encapsulation of carotenoids by spray-drying has been reported as follows: paprika oleoresin in 15-dextrose equivalent (DE) maltodextrin, arabic gum, and sodium caseinate [8]; carrot carotenes in maltodextrin of different DE [9]; pure beta carotene in 25DE maltodextrin [10]; and Gac fruit

carotenoids [11]. Different carrier types and encapsulation techniques can affect the degree of protection of the core particles from the environment. The carrier to be used should also meet other criteria, such as high solubility in water, good film forming properties, good emulsifying properties, and low costs [12]. Examples of encapsulation materials used include natural gums (e.g., gum arabic, alginates, and carageenans), proteins (e.g., dairy proteins, soy proteins, and gelatin) and carbohydrates (e.g., dextrins and derivatives). The main advantages of maltodextrins are that they can serve as an efficient barrier against oxidation, they have a neutral taste, and their costs are low. Furthermore, they are highly soluble in water, and even at high concentrations, they form low viscosity solutions. However, maltodextrins have a lack of emulsifying capacity, which means that they should be used in combination with surface active carriers [13]. Cyclodextrins are cyclic molecules derived from starch, which have the ability to encapsulate active components within their ring structures. β-cyclodextrins have both hydrophilic and hydrophobic properties, which enable non-covalent interactions with suitable core materials to form stable complexes. Gelatin is a soluble protein compound obtained by the partial hydrolysis of collagen and is a suitable carrier due to its properties of emulsification, film-formation, water-solubility, edibility, and biodegradation [14-15]. The properties of the carrier and core materials, as well as the drying parameters, are the major factors that affect the efficacy of the process [16]. Youdee and Areekul [17] investigated microencapsulated Gac aril carotenoids with β-cyclodextrin as a carrier and found that the optimum conditions were 3% β-cyclodextrin with an inlet drying air temperature of 135 ˚C. Under these conditions, the Gac aril microcapsules showed the highest values of total carotenoid contents, encapsulation efficiency, and process yield. The ratio between core and carrier is another important factor that affects the encapsulation efficiency. Shu [18] was able to microencapsulate lycopene through a process of spray drying using a wall system consisting of gelatin and sucrose with an optimum ratio of 3:7 and an inlet drying temperature of 190 ˚C. Recently, Nguyen [19] reported that in order to successfully protect carotenoids in Gac powder between the drying process, the optimal ratio of mixed carrier (maltodextrin: gelatin 0.5:0.5) to Gac aril was 1:1.

The chemical concentrations in both the core and wall are important to the process efficacy. The ratio of core to wall directly affects the emulsion stability and encapsulation efficiency. Rocha [20] evaluated the microencapsulation of lycopene in modified starch. The concentration of lycopene was varied from 5 to 15% in a solution containing 30% solids. The results showed that the encapsulation efficiency had increased as the concentration of lycopene decreased. Therefore, the objective of this research was to evaluate the effects of Gac aril concentrations and the carrier or wall ratio on the process efficacy and on the physicochemical properties of Gac aril microcapsules.

2. Materials and methods

2.1 Sample preparation

Mature fresh Gac fruits were purchased from Kanchanaburi Province. The red aril was separated from the Gac fruit. Then it was placed in a double-layered cotton cloth and squeezed to separate the seeds from the aril. The combination of 500 g of the seedless aril was homogenized to obtain a uniform single composite. After that, it was placed into plastic bottles and kept at -20 ˚C for further studies.

2.2 Emulsion preparation

In our previous studies of Gac aril encapsulation [17], the most suitable carriers for the process have been maltodextrin DE12 (Siamwhey, Thailand), gelatin (Cartino Gelatine, Thailand), and β-cyclodextrins (Wagner, Germany). Hot distilled water was added to the mixtures of each carrier, and then 3 different concentrations of Gac aril were added, based on the total solids in the emulsion at 10-30% (Table 1). Then it was immediately homogenized at 11,000 rpm for 15 min in a Wisetis® HG-15D homogenizer (Daihan Scientific, Korea).

Table 1 Gac aril concentrations in the three carriers.

Carriers	Core to wall ratios (c/w)	Gac concentrations (%)	Total solids (˚Brix)
Maltodextrin	7:1	12.5	21
	5:1	16.7	24
	3:1	25.0	27
Gelatin	4:1	11.1	12
	2:1	20.0	21
	1:1	33.3	26
β-cyclodextrin	4:1	11.1	20
	2:1	20.0	23
	1:1	33.3	27

2.3 Spray drying conditions

The samples were spray-dried with a Model Eyela SD-1000 Spray Dryer (Tokyo Rikakikai Co., LTD, Japan), which had been equipped with a spray-drying chamber (150 cm height and 80 cm diameter), a two-way nozzle atomizer, and a cyclone separator, plus a hot air blower and an exhaust blower. The emulsion was fed into the chamber at a rate of 12-15 ml/min and was atomized by the hot air from the blower in a co-current flow mode at the inlet drying air temperature of 125 to 135 °C. The spray-dried microcapsules were collected in the cyclone separator driven by the exhaust blower. The Gac aril microcapsules were then collected and placed in aluminum foil bags and kept at 8 °C for further analysis.

2.4 Analysis of Gac aril microcapsules

2.4.1 Moisture content

The moisture content of each sample was determined by drying the samples at 105 °C in the hot air oven until a constant weight was obtained [21].

2.4.2 Water activity

A water activity meter (A_W Sprint TH 500, Novasina, Switzerland) was used to measure water activity at 25 °C.

2.4.3 Process yield

The process yield (%) was calculated as the ratio of the mass of microcapsules obtained at the end of the process to the mass of initial substances added into the emulsion [22].

2.4.4 Microcapsules morphology by SEM

The samples were attached to the stub using carbon tape. The surface was thoroughly coated with gold to a thickness of 10-20 nanometers, and then each sample was scanned using scanning electron microscope (Hitachi S-2500, Japan).

2.4.5 Determination of Lycopene and β-carotene by High performance liquid chromatography (HPLC)

According to the method by Nhung [23], the samples were extracted in a mixture of 2 ml dichloromethane: methanol (6:4). Analyses for lycopene and β-carotene were carried out using HPLC with an Inertsil ODS-3 reverse phase C18 column (150 × 4.6 mm ID). A high-performance liquid chromatograph Shimadzu HPLC system (Kyoto, Japan), 2 LC-10AD pumps, and a Rheodyne injector with the SPD-M10AV photodiode array detector at 460 and 475 nm were used. A gradient mobile phase of dichloromethane [acetronitrile (6:4, v/v)] was used as Solvent A, while methanol was used as Solvent B at a flow rate of 0.9 ml/min. The initial mobile phase composition was maintained at 70% of Solvent A for 5 min, was changed to 80% (5 min), and was then returned to the initial conditions within a period of 5 min. Carotenoid compounds were identified by comparing the peak retention times with standards.

2.4.6 Carotenoids retention

In order to extract the carotenoid content from the Gac samples, an extraction method, which was described by Tran [24] and to which some modifications had been made, was employed. The sample was extracted with a mixture of n-hexane: acetone (v/v 3:2) and was further extracted four times (with 5 ml of the solvent each time) until it was colorless. Total carotenoid content of the Gac aril microcapsules was spectrophotometrically measured at 473 nm and was expressed based on beta carotene equivalents (mg/g powder). The retention percentages were calculated from the ratio between the final total carotenoid contents in the microcapsules and the initial total carotenoids during preparation. Total carotenoid contents were obtained from the summation of lycopene and β-carotene as described above.

2.4.7 Encapsulation efficiency (EE)

The method described by Shu [18] was employed to calculate the encapsulation efficiency. The EE (%) was expressed as the ratio of the total carotenoids after the process of spray drying to the total carotenoids before the spray drying.

2.4.8 Statistical analysis

The experiments were carried out using CRD in duplicate, and all results were presented as mean values with standard deviations. Different mean values were analysed by using the analysis of variance (ANOVA), and when appropriate, Duncan's Multiple Range Test was applied at a confidence level of 95% ($p < 0.05$).

3. Results and discussion

3.1 The effects of Gac aril concentrations on the encapsulation yield and physicochemical properties of the microcapsules

The moisture content and water activity of the Gac aril microcapsules (Table 2) showed that when a higher ratio of Gac aril was added, it had resulted in decreases in moisture content and water activity. The significantly lowest moisture content ($p < 0.05$) and water activity were found at medium to high levels of Gac aril, which had been encapsulated with maltodextrin. As a consequence of adding increased amounts of solid to the emulsion, lower amounts of evaporated water were found.

The encapsulation yields were in the range of 51.69-78.38%, and the ratio of the Gac aril in the emulsion had resulted in higher encapsulation yield (Table 2). According to Beristain [25], the calculation of encapsulation yield is affected by the content of solids before and after spray drying. Therefore, a higher Gac aril content will result in a higher encapsulation yield. However, when excessive amounts of Gac aril were added, the emulsion had become unstable, and the layer of film needed to wrap the core material could not be created, which gave a low encapsulation yield as previously observed by Shu [18].

Table 2 The process yield and physical properties of the microcapsules.

Carriers	Samples	Gac concentrations (% w/v)	Moisture contents (%)	A_w	Process yields (%)
Maltodextrin	M1	12.5	4.42 ± 0.84^{b}	0.31 ± 0.01^{ab}	61.72 ± 2.16^{ab}
	M2	16.7	3.92 ± 1.12^{a}	0.30 ± 0.01^{ab}	65.33 ± 1.63^{b}
	M3	25.0	3.24 ± 1.13^{a}	0.27 ± 0.01^{a}	69.44 ± 1.48^{b}
Gelatin	G1	11.1	5.98 ± 1.46^{c}	0.40 ± 0.01^{c}	51.69 ± 3.52^{a}
	G2	20.0	5.53 ± 1.08^{c}	0.34 ± 0.01^{b}	53.30 ± 2.59^{a}
	G3	33.3	4.80 ± 1.29^{c}	0.30 ± 0.01^{ab}	54.19 ± 1.07^{a}
β-cyclodextrin	B1	11.1	5.96 ± 1.29^{c}	0.40 ± 0.01^{c}	68.28 ± 0.17^{b}
	B2	20.0	5.78 ± 1.66^{c}	0.36 ± 0.01^{b}	78.38 ± 1.87^{c}
	B3	33.3	5.10 ± 1.02^{bc}	0.31 ± 0.02^{ab}	75.30 ± 2.38^{c}

Values are mean ± SD The values in the same column followed by different superscripts were found to be significantly different ($p < 0.05$).

3.2 The Effects of Gac aril concentrations on the morphology of the microcapsules

Figure 1 shows the micrograph from SEM of the microcapsules enclosed with each carrier. After spray drying under selected conditions, the microcapsules had a spherical shape with a notch and no cracks. This represented a good core encapsulation. The notches found in the microcapsules covered with maltodextrin (M) and gelatin (G) might have occurred during the spray drying process, causing the powder particles to shrink during the drying and cooling processes as reported by Sheu [26] and Pedroza-Islas [27]. In addition, free carotenoids were partially found on the surface of the microcapsules.

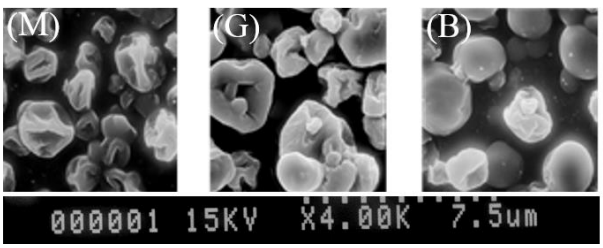

Figure 1 Micrograph of the microcapsules with different Gac aril concentrations, in which M = maltodextrin, G = gelatin, and B = β-carotene.

3.3 The effects of Gac aril concentrations on the retention percentage of carotenoids and EE of the microcapsules

The retention of the carotenoids in the microcapsules is shown in Figure 2. When the Gac aril concentrations with maltodextrin were increased, the carrier had tended to retain more carotenoids. However, Shahidi [28] found problems in microencapsulation associated with the use of hydrolyzed starches, like maltodextrin, such as their lack of emulsification properties and poor flavor retention. With gelatin and β-cyclodextrin, there was an exact ratio between the core and the carriers, which maximized the retention of carotenoids. When the Gac aril content was excessive, the excess carotenoids remained on the surface of the microcapsules and as a consequence, caused oxidation. In addition, Beristain [25] stated that when the ratio of the core material was increased too much, it had taken a long time to form the film encasing large core particles. Thus, there can be an increased loss of carotenoids, which can occur during the drying process. Additionally, carrier properties are also important for the retention of carotenoids. Figure 1 shows that among all carriers used in this experiment, β-cyclodextrin had significantly shown a higher retention percentage ($p < 0.05$). β-cyclodextrin has an ability to encapsulate active material within their ring structures. Moreover, being both hydrophilic and hydrophobic in nature, it leads to non-covalent interactions with carotenoids to form stable complexes [29].

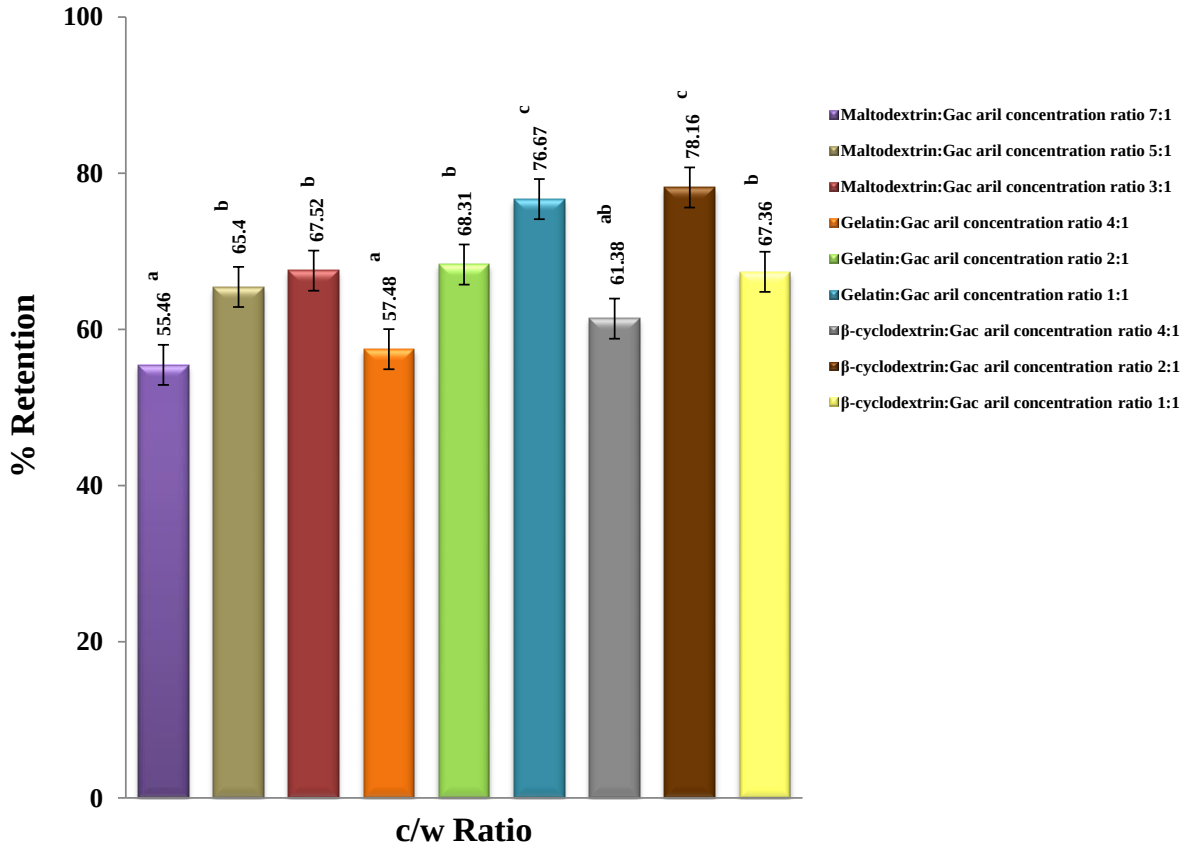

Figure 2 The percentages of carotenoid retention of the microcapsules with different c/w ratios of each carrier. The values in the same column followed by different superscripts were found to be significantly different ($p < 0.05$).

Increasing the amount of Gac aril had resulted in higher EE in the samples, which corresponded to the retention percentage (Figure 3). β-cyclodextrin showed a significantly ($p < 0.05$) greater EE compared to the other two carriers. However, it was noted that when the concentration of the aril extract was increased from medium to high, the EE was not found to increase with all the carriers. This could have been due to an overload of carotenoids, which would have meant that the carrier would not have been able to retain all the core materials. Moreover, this would have resulted in a lower encapsulation efficiency and in excess carotenoids remaining on the surface of microcapsules in large quantities as reported by Tan [30]. Beristain [24] also reported that an unsuitable core and carrier ratio can cause incomplete entrapment, resulting in low encapsulation efficiency. Moreover, carrier properties also play a major role in encapsulation efficiency. The lack of emulsification of maltodextrin and a higher viscosity of gelatin led to emulsion instability and thereby, resulted in lowering the encapsulation efficiency.

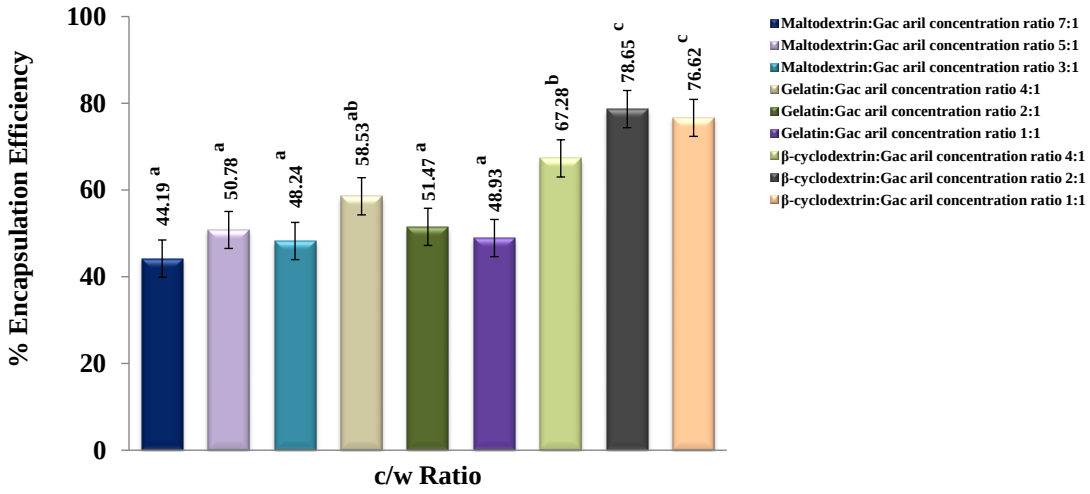

Figure 3 The encapsulation efficiency of the microcapsules with different c/w ratios of each carrier. The values in the same column followed by different superscripts were found to be significantly different ($p < 0.05$).

3.4 The effects of Gac aril concentrations on the lycopene and β-carotene contents of the microcapsules

The lycopene and β-carotene contents are shown in Figure 4. The average amount of lycopene in the microcapsules was in the range of 57.54-64.87 µg/g, and for β-carotene, it was 11.05-18.14 µg/g. For maltodextrin with 16.7 % Gac aril, the highest amounts of lycopene and β-carotene were 60.00 and 15.39 µg/g, respectively. Meanwhile, the highest lycopene and β-carotene contents for 20.0% Gac aril with gelatin and β-cyclodextrin were found to be significantly higher at 58.54 and 13.05 µg/g and 64.87 and 18.12 µg/g, respectively.

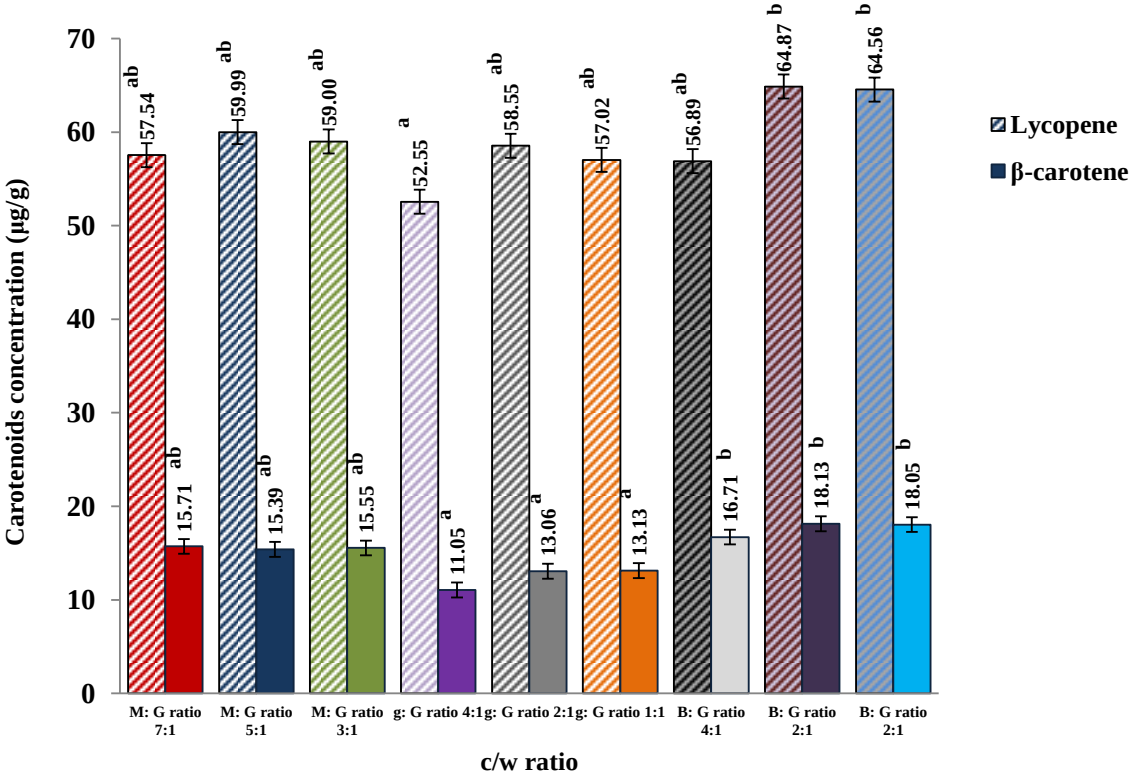

Figure 4 Lycopene and β-carotene contents of the microcapsules with different Gac aril concentrations. The values in the same column followed by different superscripts were found to be significantly different ($p < 0.05$).

4. Conclusion

The type and ratio of carrier were found to have exerted significant effects upon the efficacy of microencapsulation and its physicochemical properties ($p < 0.05$). Our results indicated that the most suitable condition was using 20.0% Gac aril encapsulated with 3% β-cyclodextrin because it gave the highest efficiency and quality. Not only did the β-cyclodextrin effectively retain the Gac carotenoids within their microcapsules, but among the encapsulation efficiency carriers tested, it also gave the best encapsulation efficiency. In comparison, β-cyclodextrin, maltodextrin, and gelatin retained lower amounts of carotenoids and demonstrated a lower encapsulation efficiency.

5. Acknowledgements

We are grateful to the Faculty of Agro-Industry of King Mongkutt's Institute of Technology Ladkrabang for providing the enabling facilities for this study. Special appreciation goes to Prof. Dr. Keith Thompson for kindly making English suggestions.

6. References

[1] Aoki H, Kieu NTM, Kuze N, Tomisaka K, Chuyen NV. Carotenoid pigments in Gac fruit (*Momordica cochinchinensis* Spreng.). Biosci Biotechnol Biochem. 2002;66(11):2479-2482.
[2] Vuong LT. Underutilized β-carotene-rich crops of Vietnam. Food and Nutr Bull. 2000;21(2):173-181.
[3] Chan R, Lok K, Woo J. Prostate cancer and vegetable consumption. Mol Nutr Food Res. 2009;53(2): 201-216.
[4] Guns ES, Cowell SP. Drug insight: lycopene in the prevention and treatment of prostate cancer. Nat Clin Pract Urol. 2005;2(1):38-43.
[5] Rao AV, Agarwal S. Role of lycopene as antioxidant carotenoid in the prevention of chronic diseases: a review. Nutr Res. 1999;19:305-323.
[6] Vuong LT, Dueker SR, Murphy SP. Plasma β-carotene and retinol concentrations of children increase after a 30-d supplementation with the fruit *Momordica cochinchinensis* (Gac). Am J Clin Nutr. 2002;75(5): 872-879.
[7] Lee MT, Chen BH. Stability of lycopene during heating and illumination in a model system. Food Chem. 2002;78(4):425-32.
[8] Beatus Y, Raziel A, Rosenberg M, Kopelman IJ. Spray-drying microencapsulation of paprika oleoresin. LWT. 1985;18:28-34.
[9] Wagner L, Warthesen J. Stability of spray-dried encapsulated carrot carotenes. Food Sci. 1995;60:1048-1053.
[10] Desobry S, Nett F, Labuza T. Comparison of spray-drying, drum-drying and freeze-drying for β-carotene encapsulation and preservation. Int J Sci. 1997;62:1158-1162.
[11] Kha TC, Nguyen MH, Roach PD. Effects of spray drying conditions on the physicochemical and antioxidant properties of the Gac (*Momordica cochinchinensis*) fruit aril powder. Food Eng. 2010;98(3):385-392.
[12] Gharsallaoui A, Roudaut G, Chambin O, Voilley A, Saurel R. Applications of spray- drying in microencapsulation of food ingredients: an overview. Food Res Int. 2007;40:1107-1121.
[13] Özbek ZA, Ergönül PG. Influence of wall material composition on microencapsulation efficiency of cold pressed pumpkin seed oil by freeze-drying. Tech Nutr Food Sci. 2018;3:1-4.
[14] Goula AM, Adamopoulos KG. Effect of maltodextrin addition during spray drying of tomato pulp in dehumidified Air: I. drying kinetics and product recovery. Drying Technology. 2008;26(6):714-725.
[15] Bruschi M, Cardoso M, Lucchesi M, Gremiao M. Gelatin microparticles containing propolis obtained by spray-drying technique: preparation and characterization. Int J Pharm. 2003;264(1-2):45-55.
[16] Zhu X, Xu SY, Wang Z. Gelatin as microencapsulating agents for β- carotene. Food Ferment Ind. 1998;24:11-15.
[17] Youdee P, Areekul V. Microencapsulation of Gac aril using β-cyclodextrin as wall material. Proceedings 1st Joint ACS AGFD-ACS ICSCT Symposium Thailand, 2014; Mar 4-5, Bangkok: ACS; 2014.
[18] Shu B, Yu W, Zhao Y, Liu, X. Study on microencapsulation of lycopene by spray-drying. J Food Eng. 2006;76:664-669.
[19] Nguyen PM. Investigation the different ratios of carrier material to protect carotenoids in GAC (*Momordica cochinchinensis* spreng) powder in drying process. Int J Multidiscip Res Dev. 2014;1(2):71-76.
[20] Rocha G, Favaro- Trindade C, Ferreira Grosso C. Microencapsulation of lycopene by spray drying: Characterization, stability and application of microcapsules. Food Bioprod Process. 2012;90:37-42.
[21] AOAC International. Official methods of analysis. 17th ed. AOAC Int.: Gaithersburg, MD; 2000.

[22] Che Man YB, Irwandi J, Abdullah WJW. Effect of different types of maltodextrin and drying methods on physicochemical and sensory properties of encapsulated durian flavor. J Sci Food Agr. 1999;79:1075-1080.
[23] Nhung DTT, Bung PN, Ha NT, Phong TK. Changes in lycopene and beta carotene contents in aril and oil of Gac fruit during storage. Food Chem. 2010;121:326-331.
[24] Tran TH, Nguyen MH, Zabaras D, Vu LTT. Process development of Gac powder by using different enzymes and drying techniques. Food Eng. 2008;85:359-365.
[25] Beristain CI, Garcia HS, Vernon- Carter EJ. Spray- dried encapsulation of Cardamom (*Elettaria cardamomum*) essential oil with Mesquite (*Prosopis juliflora*) Gum. LWT. 2001;34(6):398-401.
[26] Sheu TY, Rosenberg M. Microstructure of microcapsules consisting of whey proteins and carbohydrates. Food Sci. 1998;63:491-494.
[27] Pedroza-Islas R, Vernon-Carter EJ, Dúran-Domínguez C, Trejo-Martinez S. Using biopolymer blends for shrimp feedstuff microencapsulation-I. microcapsule particle size, morphology and microstructure. Food Res. Int. 32:367-374.
[28] Shahidi F, Han X.Q. Encapsulation of food ingredients. Crit Rev Food Sci Nutr. 1993;33(6):501-547.
[29] Özkan G, Bilek SE. Microencapsulation of natural food colorants. Nutr Food Sci. 2014;3(3):145-156.
[30] Tan LH, Chan LW, Heng PWS. Effect of oil loading on microspheres produced by spray drying. J Microencapsul. 2005;22(3):253-259.