Egypt. J. Chem. **Vol. 67,** No. 6 pp. 87 - 93 (2024)

Egyptian Journal of Chemistry

http://ejchem.journals.ekb.eg/

Influence of selected hydrocolloids and isolated pea protein on drying rate and antioxidant capacity of mulberry filling using hot air drying

Montri Chaisawang[1,2]* and Apinan Sripywan[2]

[1]*Food Processing and Culinary Science, Faculty of Science and Technology, Rajamangala University of Technology Rattanakosin, Nakhon Pathom, Thailand*

[2]*General Education, Faculty of Liberal Arts, Rajamangala University of Technology Rattanakosin, Bangkok, Thailand*

Abstract

Antioxidant-fortified with dried mulberry has become an important choice for resolving nutrition gaps in bakery products. Because of its high phenolic content and antioxidant potential, mulberry fruits have been implemented medicinally. Selected hydrocolloids and pea protein isolates are alternatives components for the mulberry filling's structure. The purpose of this study was to develop dried mulberry using the incorporation of selected hydrocolloids and isolated pea protein that were heated at elevated temperature using hot-air drying to evaluate drying rate, antioxidant retention, and quality attributes. According to the findings, the inclusion of selected hydrocolloids and isolated pea protein (IPP) increased heat stability, slightly decreased the drying rate while retaining antioxidant capacity by 22% for xanthan gum (XG+IPP), 16% for guar gum (GG+IPP), and 10% for beta-glucan (BG+IPP) when compared to the dried mulberry solely (MS). In comparison, the cohesion of isolated pea protein with xanthan gum improved textural characteristics by enhancing cohesiveness and gumminess when compared to GG+IPP, BG+IPP, and MS, respectively. The color value (L^*) of mulberry filling was slightly affected. This study suggests that the incorporation of selected hydrocolloids and pea protein improves the antioxidant retention of dried mulberry and can be utilized to manufacture high-quality of bakery filling.

Keywords: Mulberry)*Morus rubra*(, Hot-air drying, Antioxidant, Textural characteristics, Bakery filling

1. Introduction

Bakery filling was historically popular, but nowadays consumers prefer foods that are low in sugar, high in antioxidants and also contain nutraceuticals. The locally cultivated mulberry (Morus rubra) is a fruit that has gained recognition due to its increased nutritional health benefits in a variety of food products. Mulberries' active ingredient is anthocyanin, a purple pigment present in mulberries that has antioxidant action and is attracting attention as a nutraceutical [1]. Mulberry fruit also possesses medicinal and nutraceutical benefits due to its antioxidant, lowering blood sugar and blood pressure, cholesterol-lowering agent, phenolic compounds that help reduce inflammation [2,3], inhibiting some types of cancer [4], neurological disorders that involve Alzheimer's disease [5], and some other behavioral disorders [6].

One methodology to make mulberry filled-cracker is drying process to reduce moisture content and increase the texture and stickiness of the product. These products consist of dried mulberries sandwiched between biscuits or used as a bakery filling. Conventional process, particularly hot-air drying, is widely employed by SMEs (Small and medium-sized enterprises) to preserve fruit for a lengthy period of time. Hot temperatures, on the other hand, can destroy the important vitamins and nutrients in mulberry, drastically reducing their quantity. Hydrocolloids have the potential to assist minimize the degradation of essential active compounds in fruit following drying and storage. Xanthan gum is a high molecular weight biopolymer that is commonly used to thickening food and stabilize bakery filling including cohesion. Xanthan and guar gum also have high water-binding capabilities and are utilized in a variety of application, including improving texture and heat stability in fruit filling [7]. Beta-glucan has been extracted from a variety of sources, including yeast cell walls (Saccharomyces cerevisiae), mushrooms, seaweed, algae, bacteria, cereal seeds, so on. Oral administration of yeast extracts containing beta-1,3/1,6 glucan has been proven in animal studies to activate the immune system [8], anti-cancer immune response in animal studies and patients with colon cancer [9], and enhances the immune system's response to influenza challenges in mice [10]. Pea protein (Pisum sativum) is being employed as an ingredient in baked products due to its improved nutritional value, and ability to act as an emulsifier and encapsulator for bioactive compounds [11]. The combination of pea protein (Pisum sativum) derived from legume seed and pectin has been interested in spray drying for encapsulation and oxidative stability in PUFA-rich oil [12]. Recently, pea protein with xanthan gum has been demonstrated to improve rheological properties and enable 3D printing in dysphagia diets [13]. According to the report, pea protein may have the potential to improve texture qualities, which is the structure for pulverized mulberry fruit.

*Corresponding author e-mail: montri.cha@rmutr.ac.th

Receive Date: 28 August 2023, Revise Date: 19 November 2023, Accept Date: 06 December 2023

DOI: 10.21608/EJCHEM.2023.232302.8512

There was a small amount of research reporting on gums and isolated pea protein in hot-air drying for bakery filling. The aim of this research was to investigate the effects of chosen hydrocolloids, namely xanthan gum, guar gum, and beta-glucan incorporated with isolated pea protein, on drying rate, texture, and antioxidant stabilization in fruit filled bakery product dried utilizing hot air drying. This research provided information for the development of bakery filling using mulberry fruit.

2. Materials and methods

2.1. Materials

Mulberry (Morus rubra) strain Chiang Mai 60 was obtained from a plantation from Nakhon Pathom province, Thailand. Green pea was purchased from a local market located in Nakhon Pathom province. Beta-glucan was obtained from a Specialty Natural Products Company (Chonburi, Thailand). Two hydrocolloids namely, xanthan gum and guar gum were purchased from Chemipan (Bangkok, Thailand).

2.2. Chemicals

Chemicals and reagents were purchased from Sigma Chemicals (Sigma-Aldrich, Steinheim, Germany. DPPH (2,2-diphenyl-1-picrylhydrazyl radical) was purchased from Sisco Research Laboratories Pvt, Ltd.

2.3. Preparation of Isolated Green Pea Protein

Green pea protein isolate was prepared according to the method of Ladjal-Ettoumi et al., (2016) [14] with slight modifications. Briefly, the green pea was ground, dried, and sifted through a sieve. Green pea flour (100 g) was mixed with distilled water at a 1:10 ratio (w/v) and adjusted to pH 8.00 using 1 M NaOH with stirred at 500 rpm for 45 min at room temperature (25 °C). The suspension was centrifuged at 4500×g for 20 min at 4 °C to collect the supernatant. The pellet was suspended in distilled water at a ratio of 1:5 (w/v) and adjusted to pH 8.00 with stirred further for 45 min, followed by centrifugation (4000×g, 20 min, 4 °C). Both supernatants were pooled and adjusted to pH 4.8 using 0.1 M HCl to precipitate the protein. The protein was then separated by centrifugation and collected. The precipitate was washed twice with distilled water (4 °C) and re-dispersed in distilled water with adjusted pH to 7 with 1 M NaOH, before freeze-dried. The protein content of the isolates was determined in triplicate using a Lowry method and calculated protein content by %N × 6.25. Green pea protein content was 83.5 ± 0.2 % of fresh matter.

2.4. Heat stability

The heat stability was measured using modification the method of Rudra et al., (2020) [15] with some modification. Physical stability of mulberry with selected hydrocolloids and isolated pea protein was determined by transferring 15 g (F0) of sample to test tubes with 70 mm high and 20 mm diameter sealed with plastic caps to prevent evaporation. Test tubes were subjected to the water batch at 80 °C for 30 min and then dispersions were centrifuged for 20 min at 4000 rpm. After heating, the water was poured out of the test tube. The heat stability were calculated by equation: % Heat stability = (F1/F0)*100 where F0 is initial weight of sample and F1 is final weight of sample after water releasing.

2.5. Hot air-drying

Mulberry was harvested during fully-ripened stage due to high phenolic content and antioxidant capacity [16] and storage at -20 °C. Frozen mulberry was first thawed at room temperature for 30 min, and then crushed using fruit blender for 3 min. The crushed mulberry (98% w/w) were mixed with isolated pea protein (1.5% w/w) and selected hydrocolloid (1.5% w/w of xanthan gum, guar gum, or beta-glucan) dispersions were mixed using a high-speed dispersing (Ultra-Turrax T25 apparatus, KIKA Labortechnik) with 18,000 rpm for 5 min. All materials were dried in a tray with mulberry dispersion high 3 cm. at 80 °C using a hot air-drying technique (Electric oven, model LR-ES-36 (CE), Kluaynamthai Trading Group, Thailand). Drying process were done until the moisture content decreased to 9% dry weight (initial moisture content equal to 87% dry weight). Moisture content of dried mulberry was determined according to AOAC method [17]. The dried mulberry fruit powder was weighted and keep in aluminum foil polyethylene bag. The packed powder was store at 30°C under humidity of 50-70%. Water activity (aw) of dried mulberry was measured by a water activity meter (Aqualab CX-2, Decagon Devices, Inc., USA).

2.6. Color measurement

Using a Hunter colorimeter Model D 25 optical Sensor (Hunter Associates Laboratory Inc., Reston, VA, U.S.A.), the colors of the mulberry filling were assessed. According to the CIE International Commission on Illumination & Colorimetry, the color of samples was measured using the L^*, a^*, and b^* parameters, where L^* denotes lightness, a^* denotes redness to greenness, and b^* denotes yellowness to blueness. Each measurement was performed in triplicate [18].

2.7. Texture Analysis

A TA-XT texture analyser (Stable Micro Systems Ltd, Vienna Court, Surrey, UK) equipped with a 50 kg load cell and compression plate (10 cm in diameter) were utilized to conduct texture profile analysis [19]. At room temperature, samples were compressed to 50% of their original height for two cycles at a compression speed of 2.0 mm/s. The results were expressed as hardness, adhesiveness, cohesiveness, and gumminess calculated using the resistance force-deformation curves. The experiments were done in five replications and the average values were calculated.

2.8. Determination of antioxidant capacity

DPPH assay was determined according to the method described by Amarowicz [20]. For methanolic extracted samples and standard, 0.2 mL of methanolic solution containing 0.05 to 0.2 mg of extract was dispersed with 1 mL of distilled water. Then, the solution was added into a tube containing 3 mL DPPH solution (2,2-diphenyl1-picryhydrazyl, 60 M in Methanol). The mixture was mixed using vortex for 1 min. The decrease in absorbance at 515 nm (UV/VIS spectrophotometer: model B-530 spectrophotometer, Jasco International Co., Ltd. Tokyo, Japan) was recorded versus blank incubation at room temperature after 30 min. For an antioxidant capacity assays, the selected incubation time (30 min) was the required time for the reaction to reach a plateau. Samples were analyzed in triplicate and the results were expressed in □mol Trolox equivalent antioxidant scavenging per g dry weight.

2.9. Total phenolic content

The total phenolic content in the extract was determined according to the procedure described by Gutfinger (1981)

[21] using Folin-Ciocalteu's reagent. The reaction mixture in this procedure consisted of 0.5 mL of extract, 5.0 mL of distilled water, and 0.5 mL of the Folin-Ciocalteu reagent. After 3 minutes, 1.0 mL of saturated sodium carbonate solution was given. This mixture was shaken and then placed aside for 1 hour. The absorbance was measured at 725 nm using UV/VIS spectrophotometer (model B-530 spectrophotometer, Jasco International Co., Ltd. Tokyo, Japan). Gallic was utilized as the standard curve (R^2=0.09843). The results were expressed as mg gallic acid equivalent per 100 g dry weight.

2.10. Total anthocyanin content

The total anthocyanins content was determined by different pH method using different buffer system namely, potassium chloride buffer pH 1.0 (0.025 M) and sodium acetate buffer, pH 4.5 (0.4 M) [22]. Briefly, 1 ml of extracts was diluted with 9 ml of such buffers and detected against distilled water at 510 and 700 nm. The absorbance values (A) were measured using a spectrophotometer (UV/VIS spectrophotometer: model B-530, Jasco International Co., Ltd. Tokyo, Japan) and calculated using equation: $A = (A_{510}-A_{700})_{pH1.0}-(A_{510}-A_{700})_{pH4.5}$. The anthocyanin content was express as mg cyanidin 3-glucoside/g dry weight calculated as TAC = (A.Mw.Df.100)/ε, where Mw: molecular weight (449.2 g/mol); Df: dilution factor; ε: molar extinction coefficient of cyanidin-3-glucoside (26,900 L/mol.cm); Df: dilution factor.

2.11. Statistical analysis

All analyses were measured in triplicate and expressed as the mean ± standard deviation. The differences of results were determined by analysis of variance (ANOVA) and Duncan's multiple range test. The significance of differences between mean values was indicated at the 95% confidence level.

3. Results and discussion

3.1. Heat stability

In our case, heat stability is an important parameter for determining the stability of the main constituent to prevent mulberry water release during drying. When the mulberry water is released from the main constituent during drying, this part is directly heated and easily evaporates as free water, resulting in considerable antioxidant loss. The results had showed that xanthan gum (XG+IPP) had higher heat stability than guar gum (GG+IPP) and beta-glucan (BP+IPP), respectively (Table 1). Xanthan gum (XG+IPP) had a significantly different in heat stability then beta-glucan (BP+IPP). From this experiment, it can be seen experimentally that the drying rate of beta-glucan (BP+IPP) was faster than xanthan gum (XG+IPP) due to less heat stability. The incorporation of isolated pea protein and selected hydrocolloids enhanced the heat stability due to their high-water absorbency of these two components established stable structure. Another factor, a higher viscosity value may cause a lower rate of water movement as a slower movement of oils droplets in a mayonnaise [15]. In the case of isolated pea proteins or hydrocolloids alone, the heat stability was substantially lower than the combination of the two components. This result was consistent with the drying rate experimental result. Furthermore, the heat stability test has the potential to estimate the drying rate and antioxidant retention before the actual drying experiment, allowing the number of trials to be reduced.

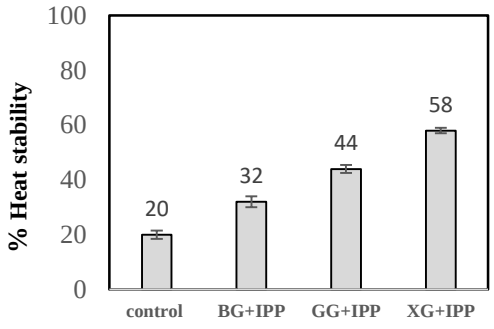

Fig. 1. Effect of selected hydrocolloids and isolate pea protein on drying rate of mulberry filling: (MS) mulberry solely, (XG+IPP) xanthan gum-isolated pea protein, (GG+IPP) guar gum-isolated pea protein, (BG+IPP) beta-glucan-isolated pea protein.

3.2. Effect of selected hydrocolloids and isolate pea protein on drying rate of mulberry filling

Moisture content is an important parameter to determine the stability of dried mulberry during storage in order to control food quality and safety. The bad condition of dried mulberry has high level of moisture contents lead to microbial growth, texture, appearance, and off flavors from lipid oxidation. Although the moisture content below than 3% is suitable for dried products, however, dried fruits can be used at maximum 10% moisture content or water activity less than 0.6 for avoid microbial deterioration [23]. After the bag was opened, the moisture of dried product will increase due to moisture from the environment [24].

In many reports, the drying rate reduced substantially as the temperature increased from 60 to 80 °C [25]. A temperature of 80 °C was chosen for this experiment to examine drying performance. Hydrocolloid-infused samples found a greater moisture content than the untreated sample, but it was still less then 10% moisture content after drying in the range of 8.3 to 9.7%. Addition of gums and isolated pea protein influenced on the interaction within the sample which caused difficulty moisture release form samples during drying. According to the finding, hydrocolloid-infused sample shown delay releasing of moisture compared to untreated sample, especially initial drying phase. The sample treated with beta-glucan and isolated pea protein (BP+IPP; 1.6 gwater/gdw.(h)) showed slow drying rate than guar gum (GG+IPP; 1.9 gwater/gdw.(h)), xanthan gum (BP+IPP; 1.8 gwater/gdw.(h)), and mulberry solely (MS; 1.4 gwater/gdw.(h)) as shown in figure 2. The low drying rate resulted in a decrease in the productivity of mulberry especially, XG+IPP sample. This result was most likely caused by the significant interaction of xanthan gum and pea protein in mulberry constituents, which resulted in a hard wall structure and limited water evaporation. However, the drying rate of mulberry infused with guar gum (GG+IPP) and xanthan gum (XG+IPP) was not far-off from beta-glucan (BP+IPP) samples and mulberry solely (MS).

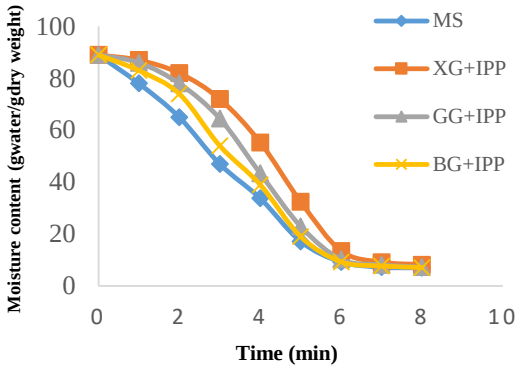

Fig. 2. Effect of selected hydrocolloids and isolate pea protein on drying rate of mulberry filling: (MS) mulberry solely, (XG+IPP) xanthan gum-isolated pea protein, (GG+IPP) guar gum-isolated pea protein, (BG+IPP) beta-glucan-isolated pea protein.

3.3. Color attributes of mulberry filling

Despite the addition of hydrocolloid and isolated pea protein at such concentration to the mulberry's constitution, the black color remained unchanged after drying as shown in figure 3.

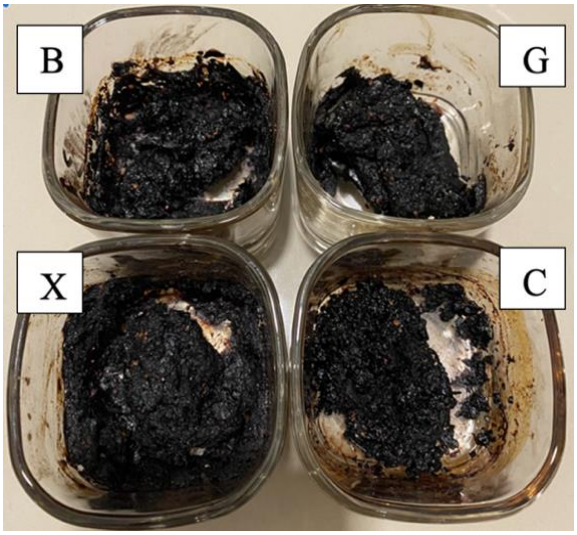

Fig. 3. General appearance of mulberry filling: (C) mulberry solely, (XG+IPP) xanthan gum-isolated pea protein, (GG+IPP) guar gum-isolated pea protein, (BG+IPP) beta-glucan-isolated pea protein.

However, based on analysis of L^* value of all formulas (Table 1), the value of control sample was slightly decrease ($p<0.05$) from other formulations. It's possible that hydrocolloids prevent the browning reaction between isolated protein and sugar from mulberry fruit during drying and the original color of the hydrocolloids and the color of the isolated pea protein incorporated in dried mulberry's constitution. The a^* and b^* values of the mulberry fillings with the incorporation of selected hydrocolloids was not significantly different when compared to control sample.

Table 1. Effects of selected hydrocolloids and isolated pea protein on color attributes of mulberry filling

	Parameters		
Sample	L^*	a^*	b^*
MS	29.88 ± 0.49^{a}	6.60 ± 0.30^{ns}	3.71 ± 0.79^{ns}
BG+IPP	30.98 ± 0.83^{ab}	6.63 ± 0.70^{ns}	3.72 ± 0.64^{ns}
GG+IPP	31.33 ± 0.77^{b}	6.53 ± 0.83^{ns}	3.64 ± 0.83^{ns}
XG+IPP	31.45 ± 0.63^{b}	6.69 ± 0.63^{ns}	3.75 ± 0.73^{ns}

L^*: Light to dark; a^*: red to green; b^*: yellow to blue

*Control: Control mulberry fillings prepared by mulberry without hydrocolloids and pea protein. Values (mean ± standard deviations; n = 3) in the same row followed by different letters are significantly different ($p<0.05$). ns: not statistically significant. (MS) mulberry solely, (XG+IPP) xanthan gum-isolated pea protein, (GG+IPP) guar gum-isolated pea protein, (BG+IPP) beta-glucan-isolated pea protein.

3.4. Effect of selected hydrocolloids and isolated pea protein on texture attributes of mulberry filling.

The texture results of dried mulberry obtained by different selected hydrocolloids and isolated pea protein were shown in Table 2. According to the findings, hydrocolloids and isolate pea protein incorporation reduced the hardness (the force required to initial compression) but increased the gumminess (energy required to disintegrate a semisolid food) of dried mulberry. This could explain why selected hydrocolloids encourages protein molecules to create a more viscoelastic and slightly firmer interfacial layer [26]. From the results, xanthan (XG+IPP) and guar gum (GG+IPP) were more effective for cohesiveness (The strength of the internal bonds making up to food) and gumminess than beta-glucan (BG+IPP) as shown in figure 2. The results are in accordance with the findings presented by Azimi et al. [27], who found that increasing the amount of guar gum with a fixed amount of gelatin increased the cohesiveness of white mulberry pastille while decreasing in hardness. Beta-glucan is a yeast cell wall with possibly low water-binding capacity and negligible interaction with pea protein. This finding suggests that samples treated with xanthan gum or guar gum in the presence of isolated pea protein may have a potential utility for improved cohesiveness. The significant consideration is that the higher cohesiveness and gumminess values for stringier and stickier of dried mulberry are suited for the manufacturing of mulberry fillings.

The control sample was dry, slightly clumping, and lacked noticeable agglomerations. Even if the control sample adheres to one another, it can be easily separated. The dried mulberry pulp became stickier when hydrocolloids and isolated pea proteins were added except for the untreated samples. This characteristic was consistent with the stickiness of bread when guar gum and xanthan gum were added [28].

3.5. Antioxidant retention

The results of DPPH radical scavenging of the dried mulberry with selected hydrocolloids and isolated pea protein are shown in the Table 3. High-temperature hot air drying enhanced drying rate but significantly lowered antioxidant capacity. The overall antioxidant capacity evaluated by the DPPH method decreased by almost 50% in the control sample compared to fresh mulberry.

Incorporation of hydrocolloids and pea protein isolate in mulberry constituent is important to retard the lowering of antioxidant capacity after drying. Form the results, xanthan gum (XG+IPP) was able to slow down the decrease in antioxidant capacity more than guar gum (GG+IPP) and beta-glucan (BG+IPP), respectively. Incorporation of gum and pea protein retained antioxidant capacity by 22% for xanthan gum (XG+IPP), 16% for guar gum (GG+IPP), and 10% for beta-glucan (BG+IPP) comparing to the control. Although high temperature accelerates oxidative and hydrolytic enzymes, hydrocolloids and proteins act like wall structure, reducing the amount of heat reaching to the core of the mulberry dispersion. The result of antioxidant retention was consistent with the drying rate experimental result [29]. The low drying rate reduce antioxidant damage from heat.

Table 2. Effects of selected hydrocolloids and isolated pea protein on texture attributes of mulberry filling

Sample	Hardness (N)	Adhesiveness (N.s)	Cohesiveness (-)	Gumminess (N)
MS	30.44±1.11^{d}	-0.098±0.013^{d}	0.11±0.04^{a}	3.33±1.53^{a}
BG+IPP	22.23±1.25^{c}	-0.131±0.007^{c}	0.25±0.07^{b}	5.50±1.44^{b}
GG+IPP	15.67±1.74^{a}	-0.153±0.006^{b}	0.83±0.08^{c}	13.00±1.76^{c}
XG+IPP	19.63±1.81^{b}	-0.175±0.014^{a}	0.92±0.05^{d}	18.05±1.45^{d}

Data are expressed as mean ± SD. Different letters in the same column represent statistically different at $p<0.05$. (MS) mulberry solely, (XG+IPP) xanthan gum-isolated pea protein, (GG+IPP) guar gum-isolated pea protein, (BG+IPP) beta-glucan-isolated pea protein.

The phenolic compounds can scavenge DPPH radicals and act as powerful antioxidants in mulberry. The most prominent anthocyanins found in mulberry have been reported to be cyanidin-3-glucoside and cyanidin-3-rutinoside [30]. According to the experimental results, the total phenolic content increased presumably due to heating at elevated temperature compared to the fresh mulberry. This result was consistent with the report of Chang et al. [31] found that an impressive phenolic content at high temperatures. Phenolic substances can act as antioxidants by interacting with Fe^{2+} [32], resulting in a decrease in the number of hydroxyl ions due to the low breakdown of hydrogen peroxide (Fenton reaction) during stress. The hydrogen peroxide will then be eliminated by catalase in human. Antioxidant capacity was largely dependent on the concentration of phenolic compounds and some factors that can impact total antioxidant activity. The incorporation of selected hydrocolloids and isolated pea protein slightly lower the total phenolic content compared to the mulberry solely. Strong intense bond between protein or hydrocolloids with polyphenols can modestly inhibit the destruction of phenolic compounds in dried mulberry.

In terms of total anthocyanin content, samples containing guar gum and xanthan gum had total anthocyanin values corresponding to their antioxidant capacity. Xanthan gum (XG+IPP) was able to delay the loss of anthocyanin content more than guar gum (GG+IPP) and beta-glucan (BG+IPP), respectively. For the result of total anthocyanin content, the usage of hydrocolloids and isolate pea protein in mulberry constituent can postpone anthocyanin depletion following heating by hot-air drying. It was proposed that gum arabic created complex formation by the hydrophobic attraction between gum arabic and anthocyanins in an assembled nanostructure improved the antioxidant retention of anthocyanin after heating at 80 °C [33]. Additionally, it has been suggested that xanthan gum and the anthocyanins in black rice can interact through mainly hydrogen bonds and hydrophobic interactions [34]. The reduced thermal degradation of anthocyanin caused by our integration of hydrocolloids and separated pea protein was probably the cause of the improved retention of antioxidative capabilities. The antioxidant activity of mulberry was consistent with its anthocyanin concentration, which is a significant antioxidant in dried mulberry. According to this study, the thermal degradation of anthocyanin was significantly reduced by the addition of hydrocolloid and isolated pea protein.

Table 3. Effects of selected hydrocolloids and isolated pea protein on total antioxidant capacity, total phenolic content, and total anthocyanin content of mulberry filling

Sample	DPPH (mg TE/g db)	Total phenolic (mg GAE/g db)	Total anthocyanin (mg TAC/g db)
Fresh	133.32±14.45^{d}	12.33±0.12^{a}	18.31±0.23^{e}
MS	66.82±12.33^{a}	18.2±0.44^{d}	8.34±0.54^{a}
BP+IPP	76.44±7.46^{b}	17.4±0.74^{c}	12.53±0.33^{b}
GG+IPP	82.42±4.23^{c}	15.3±0.32^{b}	13.83±0.45^{c}
XG+IPP	87.33±2.82^{c}	15.2±0.43^{b}	14.32±0.22^{d}

Data are expressed as mean ± SD (n=3). Different letters in the same column represent statistically different at $p<0.05$. GAE: gallic acid equivalent; TE: Trolox equivalent; db: dry basis. (MS) mulberry solely, (XG+IPP) xanthan gum-isolated pea protein, (GG+IPP) guar gum-isolated pea protein, (BG+IPP) beta-glucan-isolated pea protein.

4. Conclusions

The efficacy of different selected hydrocolloids and isolated pea protein combinations in the mulberry drying was evaluated using hot-air drying. The heat stability though our combination wall material must be considered as one of the key factors affecting the drying rate and antioxidant retention. The xanthan gum and isolated pea protein combination (XG+IPP) with mulberry fruit demonstrated a negligibly slower drying rate than guar gum (GG+IPP) and beta-glucan (BG+IPP). Even with such a concentration of hydrocolloids and isolated pea protein added to the mulberry's constituent, black color was slightly affected. In the antioxidant retention, the combination of xanthan gum and isolated pea protein outperformed the other ingredients in protecting the active material from elevated temperature. Xanthan gum or guar gum combined with isolated pea protein could be considered as a good wall material with good results for oxidative stability and bakery filling characteristic.

5. Conflicts of interest

"There are no conflicts to declare".

6. Formatting of funding sources

This work was financially supported by the National Research Council of Thailand (A9/2560). Researchers would like to thank the Rajamangala University of Technology Rattanakosin.

7. References

1. Ramappa, V. K., Srivastava, D., Singh, P., Kumar, U., Kumar, D., Gosipatala, S. B., ... & Raj, R. (2020). Mulberries: a promising fruit for phytochemicals, nutraceuticals, and biological activities. International Journal of Fruit Science, 20(sup3), S1254-S1279.

2. Guan, Y., & Zhong, Q. (2015). The improved thermal stability of anthocyanins at pH 5.0 by gum arabic. LWT-Food Science and Technology, 64(2), 706-712.
3. Kamiloglu, S., Serali, O., Unal, N., & Capanoglu, E. (2013). Antioxidant activity and polyphenol composition of black mulberry (Morus nigra L.) products. Journal of Berry Research, 3(1), 41-51.
4. Chen, P. N., Chu, S. C., Chiou, H. L., Kuo, W. H., Chiang, C. L., & Hsieh, Y. S. (2006). Mulberry anthocyanins, cyanidin 3-rutinoside and cyanidin 3-glucoside, exhibited an inhibitory effect on the migration and invasion of a human lung cancer cell line. Cancer letters, 235(2), 248-259.
5. Suttisansanee, U., Charoenkiatkul, S., Jongruaysup, B., Tabtimsri, S., Siriwan, D., & Temviriyanukul, P. (2020). Mulberry fruit cultivar 'Chiang Mai'prevents beta-amyloid toxicity in PC12 neuronal cells and in a Drosophila model of Alzheimer's disease. Molecules, 25(8), 1837.
6. Kang, T. H., Oh, H. R., Jung, S. M., Ryu, J. H., Park, M. W., Park, Y. K., & Kim, S. Y. (2006). Enhancement of neuroprotection of mulberry leaves (Morus alba L.) prepared by the anaerobic treatment against ischemic damage. Biological and Pharmaceutical Bulletin, 29(2), 270-274.
7. Janna, C., & Svetlana, P. (2013). Influence of different hydrocolloids on physicochemical and heat-stable properties of fruit fillings. The Annals of the University Dunarea de Jos of Galati. Fascicle VI-Food Technology, 37(2), 59-67.
8. Barton, C., Vigor, K., Scott, R., Jones, P., Lentfer, H., Bax, H. J., ... & Spicer, J. F. (2016). Beta-glucan contamination of pharmaceutical products: How much should we accept?. Cancer immunology, immunotherapy, 65(11), 1289-1301.
9. Chen, J., Zhang, X. D., & Jiang, Z. (2013). The application of fungal beta-glucans for the treatment of colon cancer. Anti-Cancer Agents in Medicinal Chemistry (Formerly Current Medicinal Chemistry-Anti-Cancer Agents), 13(5), 725-730.
10. Vetvicka, V., & Vetvickova, J. (2015). Glucan supplementation enhances the immune response against an influenza challenge in mice. Annals of translational medicine, 3(2).
11. Hadidi, M., Boostani, S., & Jafari, S. M. (2022). Pea proteins as emerging biopolymers for the emulsification and encapsulation of food bioactives. Food Hydrocolloids, 126, 107474.
12. Aberkane, L., Roudaut, G., & Saurel, R. (2014). Encapsulation and oxidative stability of PUFA-rich oil microencapsulated by spray drying using pea protein and pectin. Food and Bioprocess Technology, 7, 1505-1517.
13. Liu, Z., Chen, X., Dai, Q., Xu, D., Hu, L., Li, H., ... & Mo, H. (2023). Pea protein-xanthan gum interaction driving the development of 3D printed dysphagia diet. Food Hydrocolloids, 139, 108497.
14. Ladjal-Ettoumi, Y., Boudries, H., Chibane, M., & Romero, A. (2016). Pea, chickpea and lentil protein isolates: Physicochemical characterization and emulsifying properties. Food Biophysics, 11(1), 43-51.
15. Rudra, S. G., Hanan, E., Sagar, V. R., Bhardwaj, R., Basu, S., & Sharma, V. (2020). Manufacturing of mayonnaise with pea pod powder as a functional ingredient. Journal of Food Measurement and Characterization, 14, 2402-2413.
16. Mahmood, T., Anwar, F., Afzal, N., Kausar, R., Ilyas, S., & Shoaib, M. (2017). Influence of ripening stages and drying methods on polyphenolic content and antioxidant activities of mulberry fruits. Journal of Food Measurement and Characterization, 11, 2171-2179.
17. AOAC, (2000), Official Methods of Analysis of AOAC International. Seventeen editions. Association of Official Analytical Chemists. Gaithersburg, Maryland.
18. Žilić, S., Kocadağlı, T., Vančetović, J., & Gökmen, V. (2016). Effects of baking conditions and dough formulations on phenolic compound stability, antioxidant capacity and color of cookies made from anthocyanin-rich corn flour. LWT-Food Science and technology, 65, 597-603.
19. Bourne, M. C. (1978). Texture profile analysis. Food Technol., 32, 62-66.
20. Amarowicz, R., Naczk, M., & Shahidi, F. (2000). Antioxidant activity of crude tannins of canola and rapeseed hulls. Journal of the American Oil Chemists' Society, 77(9), 957.
21. Gutfinger, T. (1981). Polyphenols in olive oils. Journal of the American Oil Chemists' Society, 58(11), 966-968.
22. Fazaeli, M., Yousefi, S., & Emam-Djomeh, Z. (2013). Investigation on the effects of microwave and conventional heating methods on the phytochemicals of pomegranate (Punica granatum L.) and black mulberry juices. Food Research International, 50(2), 568-573.
23. Sornsomboonsuk, S., Junyusen, T., Chatchavanthatri, N., Moolkaew, P., & Pamkhuntod, N. (2019). Evaluation of physicochemical properties of spray dried bael fruit powder during storage. International Journal of Food Engineering, 5(3), 209-213.
24. Towns, J. K. (1995). Moisture content in proteins: its effects and measurement. Journal of Chromatography A, 705(1), 115-127.
25. Suna, S., & Özkan-Karabacak, A. (2019). Investigation of drying kinetics and physicochemical properties of mulberry leather (pestil) dried with different methods. *Journal of Food Processing and Preservation*, *43*(8), e14051.
26. Cai, Y., Deng, X., Liu, T., Zhao, M., Zhao, Q., & Chen, S. (2018). Effect of xanthan gum on walnut protein/xanthan gum mixtures, interfacial adsorption, and emulsion properties. Food Hydrocolloids, 79, 391-398.
27. Azimi, N., & Mortazavi, A. (2019). Evaluation on the effects of hydrocolloids on sensory, texture and color properties of mulberry pastille. Agricultural Engineering International: CIGR Journal, 21(3), 242-249.
28. Ahmed, J., & Thomas, L. (2018). Effect of xanthan and guar gum on the pasting, stickiness and extensional properties of brown wheat flour/β-glucan composite doughs. LWT, 87, 443-449.
29. Chen, Q., Li, Z., Bi, J., Zhou, L., Yi, J., & Wu, X. (2017). Effect of hybrid drying methods on physicochemical, nutritional and antioxidant properties of dried black mulberry. *Lwt*, *80*, 178-184.
30. Liu, X., Xiao, G., Chen, W., Xu, Y., & Wu, J. (2004). Quantification and purification of mulberry

anthocyanins with macroporous resins. Journal of Biomedicine and Biotechnology, 2004(5), 326.
31. Chang, C. H., Lin, H. Y., Chang, C. Y., & Liu, Y. C. (2006). Comparisons on the antioxidant properties of fresh, freeze-dried and hot-air-dried tomatoes. Journal of Food Engineering, 77(3), 478-485.
32. Andjelković, M., Van Camp, J., De Meulenaer, B., Depaemelaere, G., Socaciu, C., Verloo, M., & Verhe, R. (2006). Iron-chelation properties of phenolic acids bearing catechol and galloyl groups. Food chemistry, 98(1), 23-31.
33. Guan, Y., & Zhong, Q. (2015). The improved thermal stability of anthocyanins at pH 5.0 by gum arabic. LWT-Food Science and Technology, 64(2), 706-712.
34. Zhao, L., Pan, F., Mehmood, A., Zhang, Y., Hao, S., Rehman, A. U., ... & Wang, Y. (2020). Protective effect and mechanism of action of xanthan gum on the color stability of black rice anthocyanins in model beverage systems. International Journal of Biological Macromolecules, 164, 3800-3807.