

Research Article

Comparison of Antioxidant and Alpha-Glucosidase Inhibitory Properties of *Moringa peregrina* and *Ferulago carduchorum* Leaf Extracts and Microencapsulation of Superior Plant

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Today, medicinal plants have a crucial role in treating diseases such as diabetes and cancer. These plants do not impose any side effects owing to their bioactive compounds in comparison with chemical drugs. Several studies have demonstrated antioxidant capacity and α -glucosidase inhibitory effects of bioactive compounds from medicinal plants. According to previous studies, *Moringa peregrina* (MP) and *Ferulago carduchorum* (FC) are two promising plants in terms of antioxidant capacity and α -glucosidase inhibitory effects. This research followed a three-stage study. In the first stage, the antioxidant and α -glucosidase inhibitory properties of MP and FC ethanolic extracts, native to Iran, were compared using spectrophotometric methods. The results showed that the ABTS^{•+} radical scavenging and α -glucosidase enzyme inhibitory activities of both plants were dependent on extract concentration. MP exhibited lower IC₅₀ values in both tests, 1.01 and 4.96 mg·mL⁻¹, respectively. Accordingly, the extract of MP was selected for further experiments. In the second stage, total phenolic content (TPC) and GC-MS analysis were conducted on MP extract to investigate the reason behind its antioxidant capacity and α -glucosidase inhibitory properties. Results of assessing total phenolic content (TPC) using the Folin-Ciocalteu method revealed a strong positive correlation between TPC with antioxidant activity ($r = 0.94$, $p < 0.05$). GC-MS was used to identify phytoconstituents of the extract, leading to the determination of 35 components whose major one was vitamin E (10.2%). To ensure its suitability for food fortification, in the third stage, encapsulation of the MP extract was followed. Microencapsulation was performed using three polymer coatings, and the effects of carriers were investigated on moisture content, solubility, bulk density, microencapsulation yield, particle size, antioxidant activity, and TPC. According to the experiments, antioxidant activity and TPC were retained well in all carriers. Moreover, SEM, DSC, and FTIR analyses confirmed that the extract was well-coated and no surface fractures were observed. The results indicated that MP can be a promising plant for food fortification as a natural antioxidant and antidiabetic source.

1. Introduction

Today, medicinal plants are employed to treat many chronic diseases, such as diabetes and cancer due to their bioactive compounds and lack of side effects unlike chemical drugs [1]. In addition, there is an increasing interest in producing fortified foods with natural extracts as health promoters [2]. A number of studies have

demonstrated antioxidant capacity and α -glucosidase inhibitory effects of bioactive compounds from medicinal plants [3–5]. Moreover, the use of synthetic antioxidants in foods, such as butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT), has declined because of their carcinogenic effects and consumers' rejection. Therefore, there exists a strong desire to use natural antioxidants [6].

Diabetes can be prevented and controlled by consuming natural products rich in antioxidants [4]. On the other hand, the breakdown of disaccharides to glucose for subsequent absorption can be catalyzed by the α -glucosidase enzyme. Generally, inhibition of starch-digesting enzymes reduces the metabolism rate of intestinal carbohydrates, which is commonly used to control blood glucose levels [7].

Moringa peregrina (MP) belongs to the *Moringaceae* family found in southeast Iran (Sistan-Baluchistan and Hormozgan provinces), where it is commonly known by the local name of “Gas-e-roghani” or “Gaz Rokh” [8, 9]. *Moringa* plant native to different regions has been considered by many researchers for its nutritional and pharmaceutical properties and they have reported its chemical composition, antioxidant potential, antidiabetic, antiinflammatory, and antimicrobial activities of its seeds and leaves [8, 10–14].

The plant *Ferulago carduchorum* (FC) with the Persian name of “Chevill” is one of the important, valuable, and aromatic native plants of western Iran, belonging to the family “*Apiaceae*” [9]. Locals use this plant as a spice and flavoring agent for dairy products. Traditionally, the plant has been used to treat digestive disorders, intestinal worms, and hemorrhoids [15]. Moreover, there are reports on anticholinesterase potential, antimicrobial, antioxidant, and biological activities, and also the composition of some *Ferulago* species, such as *F. angulata*, *F. cassia*, and *F. longistylis* [16–20].

According to previous studies, MP and FC are two valuable and rich medicinal plants [18, 21]. Our in-depth literature review revealed that geographical and ecological conditions, as well as plant species, have a significant influence on the phytoconstituent content and biological activities of both plants [17, 20, 21]. Therefore, in this work, the plants, MP and FC, native to Iran, were studied. Previously, the antioxidant activity of methanolic leaf extract of Iranian MP was investigated [22]. To the best of our knowledge, while studies have been conducted on antioxidant capacity and anti-diabetic properties of MP and FC in other countries [23], there exist no studies on antioxidant capacity and anti-diabetic properties of MP and FC ethanolic leaf extracts, native to Iran.

Encapsulation insulates core material from environmental influences. Choosing the right wall material is the most important factor in microencapsulation, coming in both natural and synthetic forms. Maltodextrin, Arabic gum, and modified starch are common hydrophilic wall materials for microencapsulation. Due to their natural taste and odor, high encapsulation efficiency, good density properties, suitable protection of core material, good storage ability, and long thermal stability, in this study, they were selected for microencapsulation [24–27]. Also, studies have indicated that the microcoating technique can preserve antioxidant properties of the extract during processing, and covers unpleasant taste and appearance of the extract [27, 28].

Therefore, the objective of this work was to evaluate the antioxidant properties and alpha-glucosidase inhibition of ethanolic extracts of MP and FC. For facilitating the use of plant extract as a natural antioxidant and hypoglycemic agent in the food industry, encapsulation of superior plants

was considered the second objective. It is important to note that this is the first time that encapsulation of MP (superior plant) extract is performed and its antioxidant properties are studied.

2. Materials and Methods

2.1. Materials. Fresh *Moringa peregrina* (MP) and *Ferulago carduchorum* (FC) leaves were collected in late June in Sistan and Baluchistan and Kermanshah provinces, Iran, respectively. For extraction, ethanol (96%, Merck, Germany) was used. Sodium phosphate buffer (Merck, Germany), α -glucosidase enzyme (*Saccharomyces cerevisiae*, Sigma-Aldrich, Germany), and p-Nitrophenyl α -D-glucopyranoside (pNPG) substrate (Sigma-Aldrich, Germany) were used to conduct α -Glucosidase Inhibition analysis. 2,2'-azinobis-3-ethylbenzothiazoline-6-sulfonic acid (ABTS) (Sigma-Aldrich, Germany), potassium persulfate (Merck, Germany), and Trolox (Sigma-Aldrich, Germany) were employed to determine the Antioxidant Capacity of extracts. Folin-Ciocalteu reagent (Sigma-Aldrich, Germany) and Gallic acid (Merck, Germany) were used to measure the Total Phenolic Content (TPC) of the MP extract. Maltodextrin (MD) (Merck GmbH, Germany), Arabic gum (AG) (Merck GmbH, Germany), and Modified starch (MS) (Merck GmbH, Germany) were employed as wall materials in the encapsulation process.

2.2. Preparation of Plant Samples. Fresh MP and FC leaves were collected in late June in Sistan and Baluchistan (Fanouj city: 26.576282, 59.645719) and Kermanshah (Bisotun city: 34.395870, 47.445040) provinces, Iran, respectively. MP and FC plants were verified by the herbarium of the Faculty of Pharmacy of Islamic Azad University, Pharmaceutical Sciences Branch. For avoiding changes in the composition of the purified plants, they were dried in shade. Then, the dried plants were completely crushed using an electric grinder to increase the contact area of the solvent with the dry matter in order to increase extraction efficiency. Finally, they were stored in a dry, cool, and dark place for experimental use.

2.3. Preparation of Extracts. For extraction, the method proposed by Gu et al. was used with a slight modification [3]. For this purpose, 100 g of each plant's leaf powder was continuously stirred with 500 mL of ethanol for 6 h. Then, the mixture was transferred to separatory funnels. This procedure was repeated three times as long as the output of ethanolic extract contained any effective extract. The extracts from each stage were collected in a dark container protected from light and heat. Solutions of the extracts were evaporated at 38°C under vacuum using a rotary evaporator. Finally, leaf extracts were stored at 4°C for further experiments.

2.4. The α -Glucosidase Inhibition Assay. This assay was performed based on the method introduced by Fang et al. with some minor modifications [29]. Sodium phosphate

buffer (100 mM, pH 6.8), extracts at different concentrations (1.97–15.80 mg·mL⁻¹), α -glucosidase enzyme (0.25 U·mL⁻¹), and p-Nitrophenyl α -D-glucopyranoside (pNPG) substrate (5 mM) were prepared in a 96-well microplate and were incubated for 15 min at 37°C. Finally, the reaction was halted by adding sodium carbonate (0.2 M). Absorbance at 405 nm was recorded using an enzyme-linked immunosorbent assay (ELISA) microplate reader (BioTek, USA). This experiment was performed in three replicates for each concentration. The IC₅₀ (concentration of each extract required to inhibit 50% of enzyme activity) was calculated by regression equation of enzyme inhibition percentage at different concentrations.

2.5. Determination of Antioxidant Capacity. In this study, ABTS^{•+} radical scavenging capacity was used to evaluate the antioxidant activity of ethanolic extracts of MP and FC plants as described in Zulueta et al. [30]. ABTS^{•+} solution was prepared using potassium persulfate and was kept overnight in a dark place at room temperature. Absorbance was read using a spectrophotometer (Unico 2100 - USA) to adjust its absorbance to 0.7 ± 0.02 at 734 nm. Different concentrations of each extract (1.97–15.80 mg·mL⁻¹) were added to the ABTS^{•+} solution. The mixture was incubated at 25°C for 5 min and then, the absorbance of the samples was measured at 734 nm. Trolox was used as standard control to prepare standard curve, and the IC₅₀ values of extracts were compared with that of Trolox.

2.6. Determination of Total Phenolic Content (TPC). The Folin-Ciocalteu spectrophotometric method described by Carloni et al. was used to investigate the TPC of the extract in the range of 1.97–15.80 mg·mL⁻¹ [31]. Gallic acid with a concentration range of (16.65–250 mg·mL⁻¹) was used as a standard to plot the calibration curve. The TPC was obtained from linear regression equation and results were expressed as mg of Gallic acid equivalent/g of extract dried weight (GAE·g⁻¹).

2.7. GC-MS Analysis of the Extract. Gas chromatography-mass spectrometry (GC-MS) analysis of the MP extract was carried out using Agilent 6890/5973GC/MSD (USA) and HP-5 MS capillary column (30 m × 0.25 mm × 0.25 μ m of film thickness), 5%-phenyl-methyl polysiloxane. Ionization energy was equal to 70 eV. The temperature of the column oven was programmed from 50 (holding for 5 min) to 280°C with a temperature increase rate of 8°C/min (holding for 29 min). As carrier gas, Helium (99.99%) was used with a flow rate of 1 mL·min⁻¹, and the sample injection volume was equal to 1 μ L with a split ratio of 1:10. Retention times and mass spectra of the compounds were compared with those of known compounds stored in the national institute for standards and technology (NIST) library [10].

2.8. Encapsulation of MP Extract

2.8.1. Production of Microcapsules. In this study, Maltodextrin (MD), Maltodextrin/Arabic gum (MAG) (1:1), and Maltodextrin/Modified starch (MMS) (3:1) were used as

wall material (20% w/v). A spray dryer (Mini Spray Dryer B-290, BUCHI, Switzerland) was used to prepare the microcapsules. Operating conditions of the spray dryer were set as follows: Inlet temperature of $130 \pm 2^\circ\text{C}$ and outlet temperature of $85 \pm 2^\circ\text{C}$, aspirator 90%, feed pump speed of 15 mL·min⁻¹, nozzle diameter of 6–7 mm, and dry airflow rate of 50 L·h⁻¹. Microencapsulation was performed in 3 control and 3 sample treatments. MP extract was gradually added to wall material and was homogenized (2 min at 10,000 rpm and then 3 min at 25,000 rpm) using a homogenizer (Ultra Turrax IKA T25 Basic, Germany). For protecting powders from moisture absorption, they were placed in regular conical tubes and were stored at 4°C until further testing [25].

2.8.2. Moisture Content. Microcapsules were placed in a predried and preweighed Petri dish in the oven (Mettler, UFE 600, Germany) ($105 \pm 2^\circ\text{C}$, 3 h) and were weighed after cooling in the desiccator. The moisture content of the powders was calculated from weight differences before and after drying in the oven [27].

2.8.3. Solubility. According to the method proposed by Cano-Cano-Chauca et al., 1 g of each powder was added to 100 mL of distilled water with continuous stirring (500 rpm, 5 min) [32]. The resulting dispersion was centrifuged under $3,000 \times g$ (gravity) for 5 min. Then, the supernatant was poured into a pre-weighed Petri dish and was dried in an oven (Mettler, UFE 600, Germany) (105°C , 5 h). The difference in weight of the dried substance compared to the initial powder was used to determine solubility (%).

2.8.4. Determination of Bulk Density. The bulk density of the powders was measured by the tapping method. For this purpose, 0.5 g of the powders was poured into a 5-mL graduated cylinder. Then, the cylinder was continuously tapped on a flat surface until the ceasing of volume change in powder (average of 100 taps). Finally, the ratio of the powder mass to volume was used as bulk density (g·mL⁻¹) [33].

2.8.5. Encapsulation Yield. Microencapsulation yield was determined based on the weight ratio between obtained microcapsules and the initial solid solution before spray drying according to the following equation [25]:

$$\text{EY} = \frac{\text{weight of obtained microcapsule}}{\text{initial solid solution}} \times 100. \quad (1)$$

2.8.6. Analysis of Particle Size. For analyzing the particle size of the loaded microcapsules, dynamic light scattering (DLS) was used [34]. A dispersion containing 100 mg·mL⁻¹ of nanoparticles was prepared and analyzed using a particle size analyzer (Brokhaeven, ZetaPlus, USA). The results were expressed as mean particle size (nm) obtained from three replications.

2.8.7. Morphology of the Microcapsules. Microcapsules' surface morphology was studied using a scanning electron microscopy (SEM) device (Prox, Phenom Co, Netherlands). Samples were fixed on aluminum stubs and were covered with a thin layer of gold. Using SEM micrographs, the surface structure of the particles was evaluated.

2.8.8. Differential Scanning Calorimetry (DSC) Analysis. Thermal analysis of the samples was performed by differential scanning calorimetry (DSC8231, Rigaku, Japan). On an aluminum plate, 22 mg of each sample were placed. The blank plate was considered as a reference. The device was heated at a scanning rate of $10^{\circ}\text{C min}^{-1}$ in a range of $25\text{--}360^{\circ}\text{C}$ under a nitrogen atmosphere with a flow rate of $50\text{ mL}\cdot\text{min}^{-1}$ [24].

2.8.9. Fourier-Transform Infrared (FTIR) Analysis. The FTIR spectra of the extract and microcapsules were recorded using an FTIR spectrophotometer (SHIMADZU 8400S, Japan) in the wave-number range of $400\text{--}4,000\text{ cm}^{-1}$. Samples were mixed separately with potassium bromide (KBr) and were pelleted under 80 PSI of hydraulic pressure for 10 min. 16 scans at a resolution of 4 cm^{-1} for each spectrum were obtained.

2.9. Statistical Analysis. Analysis was performed using SPSS statistical software version 22. Data were analyzed using analysis of variance (ANOVA), and means were compared using Duncan's multiple-range test at $p < 0.05$. Results were expressed as mean \pm standard error (SE) of 3 replicates.

3. Results and Discussion

3.1. Antioxidant and α -Glucosidase Inhibitory Activities of MP and FC. A dose-dependent activity was observed for both plants such that, enzyme inhibitory and antioxidant activities were increased by increasing the extract concentration, shown in Tables 1 and 2 respectively.

3.1.1. α -Glucosidase Inhibitory Activity. Table 1 shows a dose-dependent inhibitory activity of MP and FC plants, where increasing the concentration of the extracts ($p < 0.05$), results in α -Glucosidase inhibitory activity growth. Concentration-dependent inhibitory activity of *Moringa* and *Ferulago* plants has been observed in several pieces of research. Ullah et al. reported the inhibitory effect of MP hydroalcoholic extract native to Saudi Arabia in a concentration-dependent manner, and they detected a moderate inhibition of α -glucosidase [23]. In another study, Magaji et al. experimented with methanolic extract of Nigerian indigenous *Moringa oleifera* leaves and observed a dose-dependent α -glucosidase inhibitory activity [35]. In a study on methanolic extract of three different species of *Ferulago* plant (*F. blancheana*, *F. pachyloba*, and *F. trachycarpa*) native to Turkey, Karakaya et al. observed that the inhibition percentage of α -glucosidase is dependent on the concentration of the extract [19].

The IC_{50} values of MP and FC extracts were determined by the regression equation of enzyme inhibition percentage at different concentrations. A lower IC_{50} value indicates higher enzyme inhibition activity. According to Table 1, the IC_{50} of MP was equal to $4.96\text{ mg}\cdot\text{mL}^{-1}$ and the IC_{50} of FC was equal to $7.41\text{ mg}\cdot\text{mL}^{-1}$. Therefore, it was concluded that the MP extract was more effective in inhibiting α -glucosidase.

3.1.2. Antioxidant Activity. Antioxidant activity of MP and FC extracts was determined using the ABTS assay. Among the advantages of this method are rapid response, cheapness, simplicity, and resistance to pH changes [30]. Table 2 shows a concentration-dependent antioxidant activity of MP and FC plants, where increasing the concentration of the extracts, increases antioxidant activity ($p < 0.05$). Concentration-dependent antioxidant activity of *Moringa* and *Ferulago* plants has been reported in a number of studies. Al-Dabbas assessed antioxidant activity using ABTS and DPPH methods in several extracts of MP native to Jordan and realized the existence of a dose-dependent antioxidant activity in all of them [36]. In another study, Jayawardana et al. reported concentration-dependent radical scavenging activity of *Moringa oleifera* leaves in chicken sausage [37]. Alizadeh et al. investigated the antioxidant activity of ethanolic extract of *Ferulago Angulata*, native to Iran, at three different concentrations, and observed concentration-dependent antioxidant activity [16].

The IC_{50} values for each extract were determined based on dose-response curves, calculated from the ABTS test, and were compared with that of the standard antioxidant Trolox. According to Table 2, the IC_{50} values were equal to 1.01, 19.32, and $0.05\text{ mg}\cdot\text{mL}^{-1}$ for MP, FC, and Trolox, respectively. Therefore, the antioxidant activity of MP and FC was much lower than that of Trolox. Comparing MP with FC, the antioxidant power of the former was much higher than that of the latter. Hence, ethanolic extract of MP was chosen for further experiments.

3.2. Total Phenolic Compounds (TPCs). In the previous experiments, MP has been found to have higher antioxidant and α -glucosidase enzyme inhibitory activities than FC ($p < 0.05$). Many studies have reported a positive correlation between phenolic compounds in plants and α -glucosidase enzyme inhibition as well as antioxidant properties [3, 10, 38]. Therefore, the TPC of MP extract was investigated in this study using the Folin-Ciocalteu method. Also, the correlation of phenolic content was investigated with antioxidant activity. Figure 1(a) shows the presence of high phenolic content in a dose-dependent manner. The obtained data were in agreement with those reported by Jafari et al. investigating the phenolic content of the MP essential oil [39]. According to regression analysis, there was a significant positive correlation ($r = 0.94$, $p < 0.05$) between TPC and antioxidant activity, as demonstrated in Figure 1(b). Thus, 87% of the antioxidant activity of the extract was due to phenolic compounds, which was in line with other studies that have found positive correlations between TPC and

TABLE 1: α -Glucosidase enzyme inhibitory activity of *Moringa peregrina* and *Ferulago carduchorum* ethanolic extracts¹.

Concentration (mg·mL ⁻¹)	α -glucosidase inhibition (%)		IC ₅₀ values (mg·mL ⁻¹)	
	<i>Moringa Peregrina</i>	<i>Ferulago Carduchorum</i>	<i>Moringa Peregrina</i>	<i>Ferulago Carduchorum</i>
15.80	142.7 ± 2.0 ^a	106.9 ± 1.1 ^a		
7.90	113.4 ± 1.2 ^a	92.8 ± 1.7 ^a		
3.95	40.9 ± 2.5 ^b	Not detected	4.96	7.41
1.97	Not detected	Not detected		
0.98	Not detected	Not detected		

¹values are expressed as mean ± standard error ($n = 3$) and different lowercase letters in the same columns show significant differences ($p < 0.05$).

TABLE 2: Antioxidant properties of *Moringa peregrina* and *Ferulago carduchorum* ethanolic extracts¹.

Concentration (mg·mL ⁻¹)	ABTS ^{•+} radical scavenging activity (%)		IC ₅₀ values (mg·mL ⁻¹)		
	<i>Moringa Peregrina</i>	<i>Ferulago Carduchorum</i>	Trolox	<i>Moringa Peregrina</i>	<i>Ferulago Carduchorum</i>
15.80	98.1 ± 1.1 ^a	41.3 ± 1.0 ^a			
7.90	94.7 ± 0.4 ^a	13.3 ± 1.6 ^b			
3.95	58.5 ± 1.9 ^b	10.0 ± 0.6 ^b	0.05	1.01	19.32
1.97	42.9 ± 2.1 ^c	Not detected			

¹values are expressed as mean ± standard error ($n = 3$) and different lowercase letters in the same columns show significant differences ($p < 0.05$).

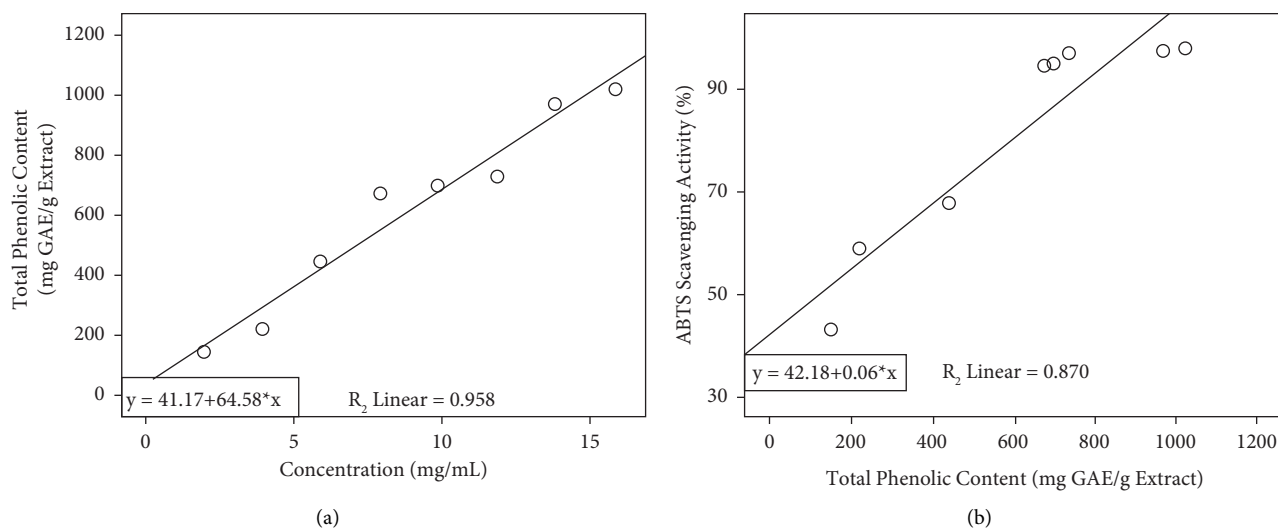


FIGURE 1: (a) Total phenolic content of *Moringa Peregrina* extract at experimented concentrations, (b) Correlation between total phenolic content and antioxidant activity of *Moringa peregrina* extract.

antioxidant activity in *Moringa*'s alcoholic extract [3, 35]. Moreover, the regression analysis demonstrated a significant correlation between the TPC of the extract and α -glucosidase inhibitory activity ($r = 0.97$, $p < 0.05$). Phenolic compounds can inhibit carbohydrate hydrolyzing enzymes such as α -glucosidase, due to their biological properties [40, 41]. Mahmoud et al. investigated antidiabetic, antioxidant, and chemical functionalities of *Moringa oleifera* methanolic leaf extract. They found a high correlation between the phenolic content and α -glucosidase inhibitory activity [42], which agrees with current research.

3.3. GC-MS Analysis. Table 3 shows the GC-MS analysis of MP extract, which revealed the presence of 35 various compounds, such as fatty acids, alcohols, aromatics, fatty acid esters, and triterpenes in chromatograms (99.6% of the

total extract). The highest percentage of the identified compounds was related to vitamin E (10.2%), which is a fat-soluble vitamin that has a phenolic ring in its structure and possesses antioxidant properties [43]. A number of the identified compounds showed different biological properties, which is probably the reason for high antioxidant and α -glucosidase inhibitory activities in this plant, including 9-Hexadecenoic acid or palmitoleic acid (antioxidant and antidiabetic), n-Hexadecanoic acid, or palmitic acid (strong antioxidant and hypocholesterolemic compound), Hexadecanoic acid, ethyl ester (antioxidant and antiandrogen), vitamin E (antioxidant, antidiabetic, and anti-inflammatory), Lupeol (antioxidant and antihyperglycemic), and β -sitosterol (antioxidant, antibacterial, anti-inflammatory, and hypoglycemic) [44–46]. Raafat and Hdaib in a study on Kenyan *Moringa Oleifera* hexane extract found that 82% of the peaks contained sterols and the constituent with the

TABLE 3: GC-MS analysis of *Moringa peregrina* extract.

No.	Compound name	Retention time (min)	(%) Content
1	Butanenitrile 3 methyl	3.50	0.8
2	Butanenitrile 3 methyl	3.60	8.7
3	Cyclotrisiloxane hexamethyl	6.31	1.9
4	Cyclotrisiloxane hexamethyl	6.40	0.9
5	Cyclotetrasiloxane octamethyl	16.89	1.0
6	D-limonene	17.59	0.4
7	Levogluosenone	21.26	4.9
8	Cyclopentasiloxane, decamethyl	22.64	2.1
9	3,6-octadecadienoic acid, methyl ester	23.96	1.9
10	Cyclohexasiloxane, dodecamethyl	27.70	2.3
11	1,6,6-trimethyl-7-(3-oxobut-1-enyl)-3,8-dioxatricyclo[5.1.0.0(2,4)]octan-5-one	29.17	0.8
12	Pyrrolizine-1,7-dione-6-carboxylic acid, methyl(ester)	29.80	0.3
13	5,9-undecadien-2-one, 6,10-dimethyl-, (Z)-	30.91	6.7
14	Cycloheptasiloxane, tetradecamethyl	32.12	0.4
15	12,15-octadecadienoic acid, methyl ester	32.42	0.4
16	Dodecanoic acid, 3-hydroxy	33.02	0.7
17	9-octadecen-12-ynoic acid, methyl ester	33.96	1.6
18	9-hexadecenoic acid	34.43	2.0
19	Dodecanoic acid, 3-hydroxy	38.32	3.7
20	Ethanol, 2-(9-octadecenyloxy)-, (Z)	39.60	2.6
21	Dodecanoic acid, 3-hydroxy	39.74	0.9
22	Ethanol, 2-(9-octadecenyloxy)-, (Z)-	40.10	1.1
23	Ethanol, 2-(9-octadecenyloxy)-, (Z)-	40.47	1.9
24	5,9,13-pentadecatrien-2-one,6,10, 14-trimethyl-, (E,E)-	41.24	5.5
25	1-heptatriacotanol	41.32	0.6
26	n-hexadecanoic acid	42.30	5.3
27	Hexadecanoic acid, ethyl ester	42.68	2.1
28	9-hexadecenoic acid	42.76	0.4
29	Ethyl iso-allocholate	44.54	0.3
30	Octadecanal, 2-bromo	44.92	1.7
31	9-hexadecenoic acid	45.53	0.7
32	9,12,15-octadecatrienoic acid, 2,3-dihydroxypropyl ester, (Z,Z,Z)-	45.87	1.3
33	Hexadecanoic acid, ethyl ester	46.33	0.5
34	9-hexadecenoic acid	49.07	0.7
35	2,6,10,14-hexadecatetraen-1-ol, 3,7,11,15-tetramethyl-, acetate, (E,E,E)-	49.54	2.7
36	Cholestan-3-one, cyclic 1,2-ethanediy l aetal, (5 β)-	51.06	0.4
37	Ethyl iso-allocholate	51.42	0.3
38	9,12,15-octadecatrienoic Acid, 2-[(trimethylsilyl)oxy]- 1-[[trimethylsilyl]oxy]methyl]ethyl ester, (Z,Z,Z)-	52.14	0.3
39	Ethyl iso-allocholate	55.89	0.5
40	1-heptatriacotanol	56.07	1.3
41	Ethyl iso-allocholate	56.17	0.6
42	3-[3-bromo phenyl]-7-chloro-3,4-dihydro-10-hydroxy-1,9(2H,10H)-acridinedione	56.56	1.5
43	3-[3-bromo phenyl]-7-chloro- 3,4-dihydro-10-hydroxy-1,9 (2H,10H)-acridinedione	56.86	2.5
44	Octadecanal, 2-bromo	57.11	0.7
45	Ethyl iso-allocholate	57.96	0.7
46	17-(1,5-dimethylhexyl)-2,3-dihydroxy-10, 13-dimethyl-1,2,3,7,8,9,10,11,12,13,14,15,16,17-tetradecahydrocycl	59.24	1.6
47	Ethyl iso-allocholate	59.44	0.6
48	Stigmastan-3,5-diene	59.72	2.3
49	Vitamin E	60.27	10.2
50	Ethyl iso-allocholate	61.61	1.1
51	β -sitosterol	62.70	2.4
52	Lupeol	63.88	2.8
	Total		99.6

highest percentage was β -sitosterol (44.6%), which had an inhibitory effect on α -glucosidase enzyme [47]. Moreover, in another study by Vats and Gupta on the hydroethanolic

extract of *Moringa oleifera* leaves, the percentage of palmitic acid, Hexadecanoic acid ethyl ester, Lupeol, and β -sitosterol, which are all antioxidant compounds, was lower than the

present study [46]. Also, Elbatran et al. identified β -sitosterol (28.8%) as the major steroidal compound in an extract obtained from the aerial part of *Moringa peregrina* native to Egypt [48]. Yet in another study, several extracts of MP leaves native to Oman were found to contain none of the compounds observed in this study except for Hexadecanoic acid ethyl ester. However, the authors found many antioxidants and bioactive compounds, such as phytol [10]. In conclusion, both types of solvent and geographical area influence the presence of phytoconstituents and the bioactivity of the compounds [10, 13, 46, 49].

3.4. Encapsulation. According to the results of the previous experiments, the extract of MP was selected for microencapsulation, due to the superiority of its extract in terms of antioxidant and anti- α -glucosidase properties. Spray drying is commonly used for producing microcapsules as it offers finer particles, better appearance and texture, bulk density, dispersibility, and solubility. It is also suitable for heat-sensitive products and can be easily implemented on an industrial scale [50]. This technique preserves the antioxidant properties of an extract during processing and also conceals its flavor and appearance [27, 28, 51].

3.4.1. Characterization of Microcapsules. Microcapsules' properties, including moisture, solubility, and bulk-tapped density are reported in Table 4. Moisture content has a negative effect on production, quality control, and shelf life of the product, as it leads to stickiness in powders, core materials oxidation, and microbial growth [25]. The moisture content of the particles was in the range of 3.4–5.5%, close to the results of the study by Mahdi et al., where similar coatings for microencapsulation of the *fingered citron* extract were used [25]. According to Table 4, the moisture content in the MAGE microcapsules (Maltodextrin/Arabic gum/Extract) was higher than the other coatings due to hydrophilicity of AG and better moisture retention of hydrocolloids compared to starch derivative, which was in agreement with findings of the studies by Sarabandi et al. and Şahin-Nadeem et al. where they reported the highest moisture content for AG coatings compared to MD coatings [27, 52].

Solubility is one of the important rheological properties of the coated materials, as it is used as an ingredient in the food industry and needs to be solved appropriately, in order to avoid processing difficulties and lowering product quality [25]. As shown in Table 4, the solubility of the microcapsules was in the range of 44.8–63.1% and depended on the type and composition of the carrier. Solubility has an inverse relation with both particle size [50], and aggregation of microcapsules [27]. The solubility of ME (Maltodextrin/Extract) microcapsules was higher than that of other coatings. This could be because of its low viscosity. This finding was consistent with the research conducted by Cano-Chauca et al. stating that solubility was decreased by increasing the viscosity of the carrier [32]. Also, it was observed that solubility was decreased by reducing MD content. This result was in agreement with that of Wang and Zhou where MD,

TABLE 4: Physical properties of *Moringa peregrina* microcapsules¹.

Treatment	Moisture (%)	Solubility (%)	Bulk tapped density(g·mL ⁻¹)
ME ²	3.4 ± 0.1 ^c	63.1 ± 0.6 ^a	0.83 ± 0.00 ^b
MAGE ³	5.5 ± 0.2 ^a	44.8 ± 0.1 ^c	0.63 ± 0.00 ^c
MMSE ⁴	4.5 ± 0.2 ^b	50.9 ± 0.0 ^b	0.89 ± 0.01 ^a

¹values are expressed as mean ± standard error ($n=3$) and different lowercase letters in the same columns show significant differences ($p < 0.05$). ²maltodextrin/extract. ³maltodextrin/Arabic gum/extract. ⁴maltodextrin/modified starch/extract.

cellulose, and waxy starch were used to microencapsulate soy sauce [53]. They observed that the powders prepared with MD had higher solubility than the other coatings.

Bulk-tapped density is important for transportation and packaging. Particles with higher density are more desirable since they accommodate smaller containers [24]. In this study, MMSE (Maltodextrin/Modified Starch/Extract) had the highest tapped density, while MAGE had the lowest. Increasing the tapped density is dependent on particle size, because smaller microcapsules can be placed in gaps between the particles. Similar results were observed in microencapsulation of rosemary oil and sour cherry concentrate with polymer coatings in the studies by Fernandes et al. and Sarabandi et al. who reported that tapped density is correlated with particle size [24, 51].

Based on Figure 2, encapsulation yield is influenced by the MD ratio where reducing MD content decreases encapsulation yield. As demonstrated in Figure 2, the ME had the highest encapsulation yield, while MAGE had the lowest ($p < 0.05$). The reason for this observation is that, when the moisture content is increased, particles stick together and also adhere to the walls of the drying chamber so that, yield is decreased [39]. In agreement with the results of the study by Sarabandi et al. AG increased viscosity and produced larger droplets with higher aggregation and decreased encapsulation yield [27]. In another study, rosemary oil was coated with AG/MS/MD/inulin [24]. The authors reported that microcapsules containing MD/MS exhibited higher encapsulation yield than pure AG and AG/MD, which was consistent with the present study.

3.4.2. Morphology of the Particles. Figure 3 shows results regarding the relationship between carrier type and particle size. The mean particle size was in the range of 266.08–410.87 nm. The highest and lowest particle sizes were obtained in MAGE and MMSE, respectively. The addition of AG to MD increased particle size ($p < 0.05$), which can be attributed to the increased viscosity of the solution. Moreover, MD produces low viscosity at low concentrations, leading to the formation of smaller droplets [39]. Furthermore, it was observed that when MS was added to MD, particle size was reduced. This is because MD has a weak emulsifying ability and also lacks surface binding activities [24]. The addition of 25% of MS to MD improved surface binding activities and led to producing finer particles.

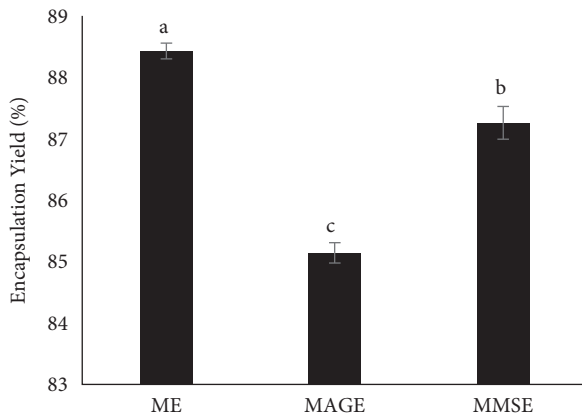


FIGURE 2: Encapsulation yield of microcapsules affected by different wall materials: maltodextrin/extract (ME), maltodextrin/arabic gum/extract (MAGE), maltodextrin/modified starch/extract (MMSE). different lowercase letters show statistically significant differences ($p < 0.05$).

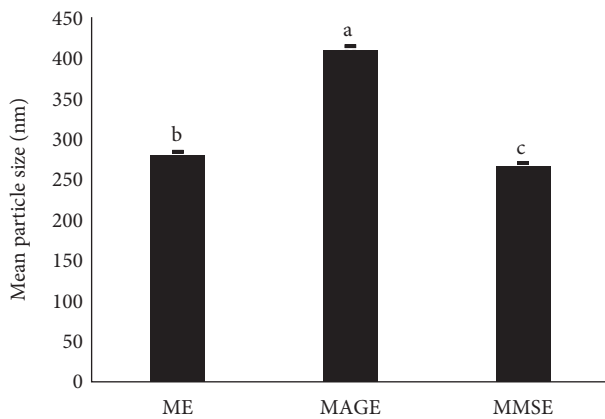


FIGURE 3: Mean particle size of MP microcapsules with different carrier materials: maltodextrin/extract (ME), maltodextrin/Arabic gum/extract (MAGE), maltodextrin/modified starch/extract (MMSE). different lowercase letters show statistically significant differences ($p < 0.05$).

Therefore, particle size depends on the type of carrier material, carrier viscosity, wall composition, and atomization method [39].

The microencapsulated particles of MP extract were studied by SEM. Figure 4 shows SEM images for each group of microcapsules. Generally, the produced microcapsules had spherical and hemispherical shapes, and in some samples, they had wrinkles and indentations. The generation of particles with irregular and wrinkled surfaces is a usual occurrence in spray drying of different products, primarily because of the quick formation of crusts on the surface of the droplets in the initial stages of drying [53]. According to Figure 4, the dried microcapsules had varying sizes and no broken microcapsules or cracks were visible in any coating. The ME microcapsules had a more uniform particle size, smoother surface, and less roughness and shrinkage compared to MAGE and MMSE microcapsules. High shrinkage at the surface of MAGE and MMSE microcapsules was due

to the formation of the gum layer as well as reinforcement of the coating by MS, and reduction in moisture leakage. These observations were in agreement with the study by Mehran et al. who reported smooth and spherical shapes for ME microcapsules compared to MMSE microcapsules [26]. Also, the findings in this study were consistent with other studies, which showed irregular spherical particles with much shrinkage and roughness on the surface [34, 54, 55].

3.4.3. Results of DSC Analysis. Thermal characteristics of microcapsules were studied using DSC analysis. Figure 5 demonstrates DSC thermograms for both MP extract and microcapsules, in a heating range of 25–360°C. Depending on the type of the carriers, all microcapsules exhibited two endothermic peaks. The first endothermic peak occurred between 25 and 10°C, which was due to dehydration and water loss from hydrophilic groups existing in the polymer structure. The second endothermic peak took place between 190 and 250°C that was because of the melting point and decomposition of the microcapsules [26]. Comparing the thermograms, it was observed that no new peak appeared in the presence of the extract. Therefore, it can be concluded that the thermal behavior of microcapsules was not influenced by the presence of the extract and it was completely incorporated into the microcapsules [56].

3.4.4. FTIR Spectroscopy. The FTIR spectrum was used to investigate the interactions between functional groups in coatings and extract. Absorption was measured in the range of 400–4,000 cm^{-1} . Table 5 reports infrared spectra's characteristic peaks of both extract and its microcapsules. According to Table 5, the addition of the extract changed the characteristic peaks significantly. Absorption bands of the MD microcapsules were observed at 3369.75 cm^{-1} (O-H stretching vibrations), 2928.04 cm^{-1} (C-H stretching vibration), 1645.33 cm^{-1} (COO^- asymmetric stretching vibrations), and 1417.73 cm^{-1} (COO^- symmetric stretching vibration). The addition of MD to AG and MS changed the location of the peak for the hydroxyl group from 3369 cm^{-1} to 3317 cm^{-1} , and 3389 cm^{-1} , respectively. Increasing hydrogen bonds of the particles may cause these changes. Similar results were obtained in the study by Mehran et al. [26].

The addition of MP extract led to a change in OH-stretching wave number for all microcapsules. Moreover, the stretching vibration wave number of the asymmetric carboxyl group in the MD shifted to a lower wave number in comparison with ME. These changes can be attributed to the interaction of these microcapsules with the extract and it can be considered an evidence of the presence of extract in microcapsules. Belščak-Cvitanović et al. reported the same results in the case of the encapsulated green tea extract with different polymer coatings [57]. Based on the current analysis, it can be concluded that the extract was well-loaded in all the carriers.

3.4.5. Antioxidant Capacity and TPC of the Encapsulated MP Extract. For verifying the efficacy of MP microcapsules to be used as a food additive, antioxidant properties, and TPC of

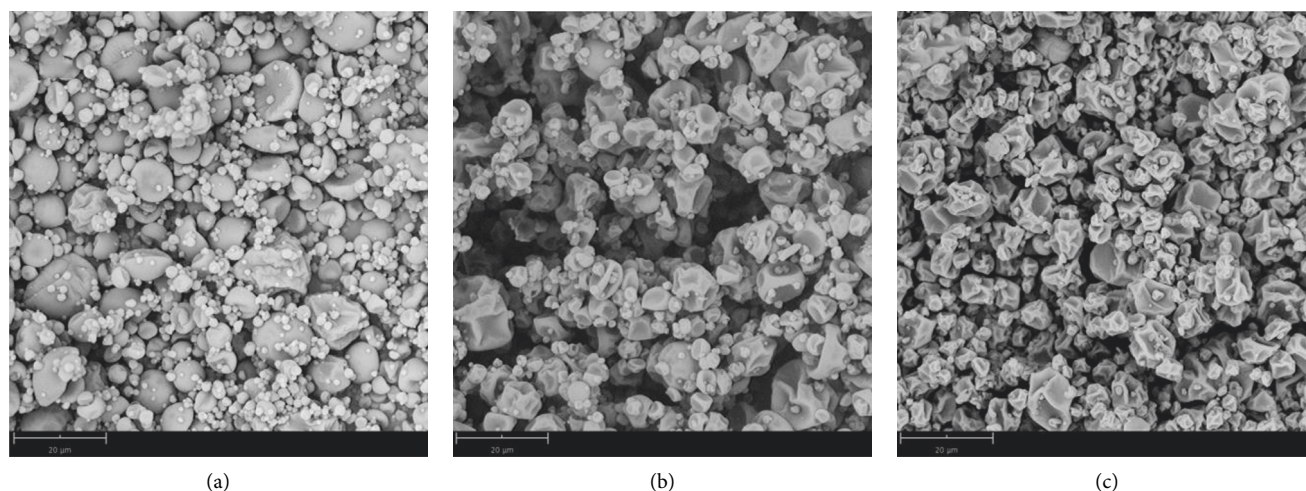


FIGURE 4: Microphotographs of microcapsules prepared by SEM. (a) Maltodextrin/extract, (b) Maltodextrin/Arabic gum/extract, (c) Maltodextrin/modified starch/extract.

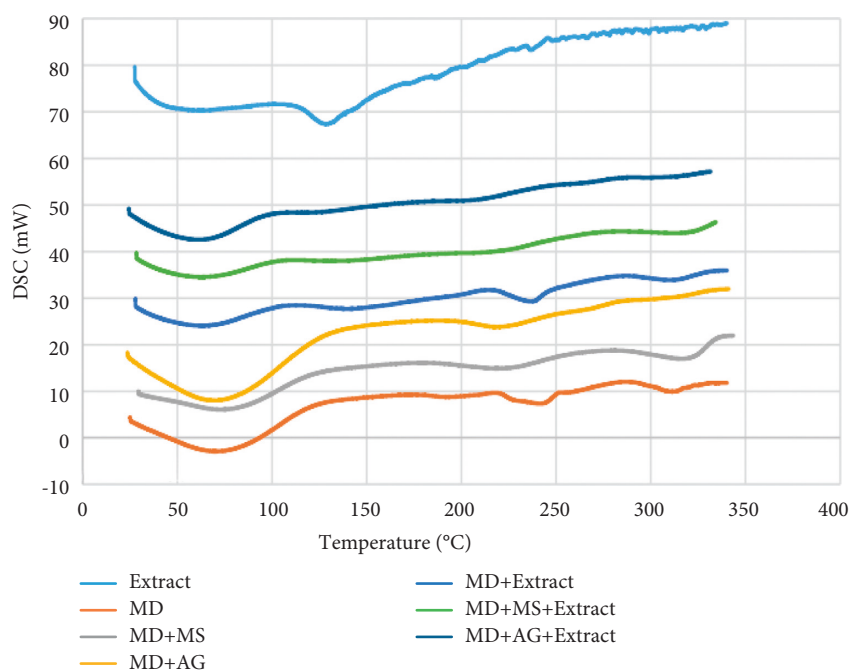


FIGURE 5: DSC curves of the extract and microcapsules. maltodextrin (MD), Arabic gum (AG), modified starch (MS).

TABLE 5: Infrared spectra's characteristic peaks of extract and microcapsules.

Bonds	Wave-number (cm^{-1})						
	Extract	ME ¹	MMSE ²	MAGE ³	MD ⁴	MMS ⁵	MAG ⁶
OH-stretching	3383.26	3337.68	3366.38	3366.91	3369.75	3389.68	3317.67
CH stretching	2854.74	2926.11	2926.11	2928.04	2928.04	2928.04	2929.97
Asymmetric COO^- stretching	1608.69	1643.41	1643.41	1622.19	1645.33	1647.26	1608.69
Symmetric COO^- stretching	1446.66	1415.80	1415.80	1419.66	1417.73	1417.73	1417.73

¹maltodextrin/extract. ²maltodextrin/modified starch/extract. ³maltodextrin/Arabic gum/extract. ⁴maltodextrin. ⁵maltodextrin/modified starch. ⁶maltodextrin/Arabic gum.

microcapsules were investigated. Table 6 shows results of evaluating antioxidant properties and TPC of the null microcapsules as well as coated extract with different wall

materials at a concentration of $15.80 \text{ mg}\cdot\text{mL}^{-1}$. The percentage of radical scavenging activity of microcapsules with and without extract was in the range of 76.3–88.5% and

TABLE 6: Antioxidant properties and total phenolic content of the coated extract with different wall materials¹.

Sample Formulation	Without extract	
	Mean inhibitory percentage of ABTS ^{•+} radical (%)	Total phenolic content (mg·GAE·g ⁻¹)
MD ²	Not detected	4.85 ± 0.10 ^c
MAG ³	17.7 ± 0.5 ^a	21.59 ± 0.11 ^a
MMS ⁴	10.8 ± 0.8 ^b	15.97 ± 0.00 ^b
Sample Formulation	With extract	
	Mean inhibitory percentage of ABTS ^{•+} radical (%)	Total phenolic content (mg·GAE·g ⁻¹)
ME ⁵	76.3 ± 0.5 ^c	332.70 ± 3.88 ^c
MAGE ⁶	80.2 ± 0.5 ^b	698.79 ± 4.85 ^b
MMSE ⁷	88.5 ± 0.7 ^a	747.91 ± 1.32 ^a

¹values are expressed as mean ± standard error ($n = 3$) and different lowercase letters in the same columns show significant differences ($p < 0.05$). ²maltodextrin. ³maltodextrin/Arabic gum. ⁴maltodextrin/modified starch. ⁵maltodextrin/extract. ⁶maltodextrin/Arabic gum/extract. ⁷maltodextrin/modified starch/extract.

10.8–17.7% respectively. Also, TPC varied in the range of 332.70–747.91 mg·GAE·g⁻¹ for coated extract, and 4.85–21.59 mg·GAE·g⁻¹ for null microcapsules. Based on Table 6, the wall material combination has a significant effect ($p < 0.05$) on both TPC and antioxidant capacity. MD wall material had no antioxidant activity and a very small amount of TPC. While the MAG and MMS contained small amounts of antioxidant activity and TPC. According to Table 6, by comparing each empty microcapsule with its corresponding coated extract, MMS wall material can provide superior protection for the core. On the other hand, MMSE had the highest percentage of free radical scavenging activity as well as the highest TPC. The reason for these observations may be the addition of starch to the coating formulation, which enabled it to protect sensitive phenolic compounds in the core. This result was consistent with studies conducted by Mehran et al. and Sansone et al., who reported that using MD/MS composition retained more phenolic compounds than the other coatings [26, 58]. Comparing the antioxidant activity and TPC of the microcapsules with that of the extract, at the concentration of 15.80 mg·mL⁻¹, shown in Table 2 and Figure 1(a) respectively, we can conclude that these properties have been decreased but still retained well after microencapsulation.

4. Conclusion

We compared the antioxidant and α -glucosidase inhibitory properties of MP and FC ethanolic extracts, native to Iran. Our results revealed that MP leaf extract is superior in terms of these properties compared to that of FC and MP selected for further experiments. Assessing TPC in the MP extract, a strong positive correlation was found between TPC with antioxidant activity and α -glucosidase inhibition. GC-MS analysis showed the presence of various phenolic compounds, with vitamin E accounting for about 10% of the total constituents. SEM, DSC, and FTIR analyses confirmed that the extract was completely incorporated into the microcapsules and no surface fractures were observed. According to the experiments, antioxidant activity and TPC were retained well in all carriers. For future studies, it is recommended to find suitable foods in order to use the encapsulated extract of this plant as a natural antioxidant and α -glucosidase enzyme inhibitor.

Data Availability

The manuscript contains most of the data generated and/or analyzed during this study. If there are other data not included in the manuscript, it is available from the corresponding author upon reasonable request.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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