Food Hydrocolloids for Health 2 (2022) 100051

Contents lists available at ScienceDirect

Food Hydrocolloids for Health

journal homepage: www.elsevier.com/locate/fhfh

Alginate based encapsulation as a tool for the protection of bioactive compounds from aromatic herbs

Alessandra Cristina Tomé*, Flávio Alves da Silva

Department of Food Engineering, School of Agronomy, Federal University of Goiás, Goiânia road / Nova Veneza, Km 0 – Mailbox 131, Zip code 74001-970, Goiânia, Goiás, Brazil

ARTICLE INFO

Keywords:
Plant extracts
Antioxidant capacity
Encapsulation

ABSTRACT

The objective of this study was to evaluate the total phenolic compound content (TPC), antioxidant capacity by the •DPPH, ABTS•+ and FRAP methods in aqueous (AqE), hydroethanolic (HE), and ethanolic (EOH) extracts of six species of aromatic herbs, Basil (*Ocimum basilicum*), Parsley (*Petroselinum crispum*), Rosemary (*Rosmarinus officinalis*), Thyme (*Thymus vulgaris*), Chervil (*Anthriscus cerefolium*) and Chive (*Allium fistulosum*), with subsequent microencapsulation, using the dripping technique, only for the ones who had better TPC contents. The TPC values presented a variation between (142.81±7.06 mg GAE/g) for the HE of rosemary and (30.52±0.95 mg GAE/g) for the HE of chervil. In this study, the lowest antioxidant activity obtained by the •DPPH was for the EOH of parsley, and the highest antioxidant activity was for the HE of rosemary. The best results found by the ABTS•+ was for the HE of rosemary 40.44±0.19 (μmol of Trolox/g), and the lowest values were for the AqE of basil 9.13±0.97 (μmol of Trolox/g), the results obtained in this assay, found higher ferric reducing power for AqE of rosemary 46.78±0.25 (μmol FeSO4/g) and lower value for EOH of chervil 8.99±0.13 (μmol FeSO4/g). The use of scanning electron microscopy revealed the presence of microparticles with the desired shape, sizes ranging from 920.08±11.63 to 754.28±16.62 μm, and encapsulation efficiency, from 68.24±0.15 to 93.39±0.01%. These results indicate that the application of microencapsulated plant extracts has potential for use in the food industry. They present good results for phenolic compounds and antioxidant capacity levels, presenting a good microencapsulation efficiency.

1. Introduction

Besides folk medicine, aromatic herbs have always been used for other purposes, including food preservation, which has progressively expanded around the world (Potortì et al., 2020; Putnik et al., 2016) with growing interest in the food industry because raw extracts of herbs, spices and other plant materials rich in phenolics can increase acceptability, delay the oxidative degradation of lipids and even improve the nutraceutical value of food products (Celano et al., 2017; El-sayed & Youssef, 2019).

Oxidative lipid degradation is one of the leading causes of food quality deterioration. Many aromatic herbs have shown promise as natural antioxidants, with the possibility of applications in various forms: whole, milled, in the form of extracts, or essential oils (Giannenas et al., 2020).

However, the application of these natural antioxidants in food products can suffer several limitations due to the possibility of being degraded if exposed to oxygen, light, enzymatic activities, adverse conditions of temperature and pH, metallic ions, and water, which leads to alteration of its beneficial properties (Rakmai et al., 2018; Rezaei et al., 2019; Sharma et al., 2019).

Microencapsulation can be employed as an exciting alternative to stabilize phenolics from natural extracts (Lee & Chang, 2020; Mar et al., 2020; Vinceković et al., 2017), and therefore, in recent years, it has attracted significant interest from food, pharmaceutical, nutraceutical, and cosmetic industries, with wide application in the design of functional products, such as food or food ingredients (Teng et al., 2019).

The microencapsulation process consists of enclosing the active agent in a carrier (matrix) to protect it against external factors, such as different time combinations of temperature, oxygen, light, humidity, and chemicals, volatile losses, or additional interactions with components such as proteins (Elena & St, 2021). There, the mechanism involves the design of a protective shell to enclose the sensitive compound, promoting its controlled release (Tarone et al., 2020).

Based on the above considerations, the objectives of this study were to evaluate the content of total phenolic compounds and antioxidant activity through the application of three in vitro assays on three different extracts, in a variety of six aromatic herbs, and also microencapsulate

* Corresponding author.
E-mail address: alessandra.tome@ifgoiano.edu.br (A.C. Tomé).

https://doi.org/10.1016/j.fhfh.2021.100051
Received 1 September 2021; Received in revised form 15 December 2021; Accepted 21 December 2021

the extracts that presented the highest contents of total phenolic compounds, using the dripping process based on sodium alginate.

2. Material and methods

2.1. Plant material

Six plant materials such as basil (*Ocimum basilicum*), curly leaf parsley (*Petroselinum crispum*), rosemary (*Rosmarinus officinalis*), thyme (*Thymus vulgaris*), chervil (*Anthriscus cerefolium)*, and chives (*Allium fistulosum*), were cultivated in the horticultural sector of the *Instituto de Educação, Ciência e Tecnologia Goiano* - Morrinhos campus (Goiás, Brazil), the cultivation time varying between 60 to 120 days according to each species. The seeds from plant materials were donated by the company *ISLA Sementes* (Porto Alegre, RS, Brazil).

2.2. Preparation of the aromatic herbs

The harvesting of leaves for the study was carried out in the morning, between 8 am and 10 am, then washed in running water, sanitized with a sodium hypochlorite solution at (100 μL.L-1) for 15 min, then rinsed in ultrapure water, arranged on trays, and dried in an incubator with air circulation at 40 °C to reduce the moisture content (< 10%), then milled in a knife mill (Fortnox FT 50), using a 30 mesh steel sieve to obtain a homogeneous powder, labeled and adequately packaged in laminated bags and stored at room temperature for further analysis.

2.3. Extract preparation

A modified version of the methodology described by Vieitez et al. (2018) was used to obtain the extracts. Briefly, the conventional technique was used at an initial ratio of 1:20 plant by volume of solvent (m:v), the ethanol extract (absolute ethanol), hydroethanolic (50% ultrapure water + 50% absolute ethanol), and aqueous (ultrapure water), under stirring at room temperature for 1 hour in a Q261-22 magnetic stirrer (Quimis, São Paulo, Brazil) and then the solution was filtered with Whatman Nº4 filter paper, and the final volume was adjusted to 50 mL with the respective solvent, later, the extracts were placed in amber glass flasks, sealed and stored in a freezer (-18°C) for further analysis.

According to Prat et al. (2015), ethanol and water are solvents of low toxicity and have a minor impact on the environment, enabling the subsequent application of microencapsulated extracts in food products.

2.4. Total phenolic compound content (TPC)

The TPC of the aqueous, hydroethanolic, and ethanolic extracts was determined using a Folin-Ciocalteu colorimetric method (Andrew L Waterhouse, 2002) with some modifications. The standard curve prepared with gallic acid in the range of 10 to 80 mg.L^{-1} was used as a reference for quantification.

Briefly, 2 mL of sample extract or gallic acid standard solution was added to a 10 mL volumetric flask mixed with 3 mL of Folin-Ciocalteu reagent diluted ten times and let settle for 3 min. The final volume was adjusted to 10 ml with sodium carbonate at 7.5% (w/v). After 30 min of incubation in dark conditions at room temperature, the absorbance of the mixture was measured at 750 nm using an *AAKER-BEL Photonics* spectrophotometer (Medianeira, Porto Alegre, Brazil). Results were expressed in milligrams of gallic acid equivalent (GAE) per gram of sample.

2.5. Determination of the antioxidant capacity

The determination of the antioxidant capacity was determined in the aqueous, hydroethanolic and ethanolic extracts using the 2.2 diphenyl-1-picrylhydrazyl DPPH method (Cuvelier & Berset, 1995) modified by Borguini et al. (2009), FRAP method (Ferric ion reducing antioxidant power; (Benzie & Strain, 1996) and 2.2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) ABTS●+ process; (Re et al., 1999) using the *AAKER-BEL Photonics* spectrophotometer (Medianeira, Porto Alegre, Brazil). The •DPPH method was based on the measurement of the degree of discoloration of the DPPH radical by the action of antioxidants after 20 minutes of reaction, which was measured spectrophotometrically in *AAKER-BEL Photonics* (Medianeira, Porto Alegre, Brazil) at 517 nm in the aqueous, hydroethanolic and ethanolic extracts, to read the absorbance in the spectrophotometer, it was necessary to dilute the extract in a concentration of 0.2 mg.mL^{-1} of sample. Results were expressed in % discoloration according to equation 1.

$$\%\ discoloration\ of\ \bullet\text{DPPH} = \left(1 - \left(\frac{abs\ sample\ - abs\ blank}{abs\ control}\right)\right) \times 100 \quad (1)$$

Where: abs sample is the absorbance of the sample; abs blank is the absorbance of blank (750 μL extract + 1.5 mL methanol), and abs control is the absorbance of the control (750 μL methanol + 1.5 mL of •DPPH).

The FRAP method is based on the ability to reduce Fe^{3+} to Fe^{2+} at low pH values. In the presence of 2, 4, 6-tripyridyl-S-triazine (TPTZ) and an antioxidant, an intense blue complex (Fe^{2+}-TPTZ) is formed, which can be determined by spectrophotometry at an absorbance of 593 nm. The results were expressed in μmol of $FeSO_4$/g.

The ABTS method is based on the capacity of the antioxidant to scavenge the $ABTS^{\bullet+}$ radical formed by the reaction between the aqueous solution of ABTS (7 μMol) and potassium persulfate (2.45 mM), which is measured in a spectrophotometer at 734 nm. Results were expressed in μmol Trolox/g.

2.6. Obtaining the microcapsules

Of the eighteen extracts obtained in this study, only six were microencapsulated: the aqueous extract of chives and hydroethanolic extracts of basil, parsley, rosemary, thyme, and chervil, because they were the ones that obtained the best levels of TPC. The microspheres were obtained according to Dalponte Dallabona et al. (2020) with modifications. Two grams of Sodium Alginate $(C_6H_7NaO_6)_n$, Analytical Grade, CAS Number: 9005-38-3, Brand: Dynamic, was mixed with 100 ml of the extract under magnetic stirring. Once homogenized, the solution (alginate + extract) was left to stand for ~2 h to remove any air bubbles. The solution was then added into 80 mL of calcium chloride solution 4% (w/v) from a burette and subjected to the dripping method.

The processing factors of the microcapsules were controlled by adding a pipette tip with a capacity of 200 μL at the end of the burette (to obtain the desired diameter for microcapsules), the drip speed was defined at 30 mL/h, and the distance established between the tip and the surface of the $CaCl_2$ solution was 7 cm. The capsules formed in this process were kept in the $CaCl_2$ solution for ~15 min under magnetic stirring at 120 rpm. Then they were filtered through Whatman filter paper and washed three times using ultrapure water. Afterward, the microspheres were air-dried at 25 °C for 24 h and kept in a desiccator at 25 °C.

2.7. Scanning electron microscopy of the microcapsules

The surface morphology and structure of the microcapsules were observed by SEM (scanning electron microscope) (JEOL–JSM 6610, Tokyo, Japan), using accelerating voltages of 2.5 kV. The microcapsules were mounted on aluminum support or “stubs” using double-sided adhesive tape, then placed in a sputter coater (*Balzers, SCD–040*) to be coated with a layer of gold. Images were captured at 30x magnification.

2.8. Determination of the total phenolic compound content of the microcapsules and the encapsulation efficiency

The Encapsulation efficiency analysis was performed according to a previously described study (Dalponte Dallabona et al., 2020) with minor modifications. 10 mg aliquots of microspheres were dissolved into 5 mL of sodium citrate (5% w/v) and centrifuged for 20 min at 3000

Table 1
Contents of Total Phenolic Compounds (TPC) and antioxidant capacity by •DPPH, $ABTS^{\bullet+}$ and FRAP methods in aqueous (AqE), hydroethanolic (HE) and ethanolic (EOH) extracts of basil (*Ocimum basilicum*), curly leaf parsley (*Petroselinum crispum*), rosemary (*Rosmarinus officinalis*), thyme (*Thymus vulgaris*), chervil (*Anthriscus cerefolium*) and chive (*Allium fistulosum*).

Herbs	Extract	TPC (mg GAE/g)*	DPPH (%)	$ABTS^{\bullet+}$ (μmol de Trolox/g)	FRAP (μmol de $FeSO^4$/g)
Basil	AqE	63.54±2.26^b	17.05±0.46^b	9.13±0.97^c	20.53±0.25^b
	HE	76.30±2.54^a	29.65±0.53^a	28.93±0.93^a	26.61±0.70^a
	EOH	34.74±2.71^c	17.66±0.53^b	13.20±0.81^b	19.96±0.16^b
Curly leaf parsley	AqE	75.05±3.13^a	16.13±0.46^a	33.85±1.04^a	25.72±0.84^a
	HE	79.01±2.68^a	16.28±1.86^a	32.61±0.22^a	23.89±0.41^b
	EOH	39.42±1.31^b	12.59±1.41^b	25.06±0.39^b	17.87±0.25^c
Rosemary	AqE	79.68±4.02^b	19.04±3.07^b	38.44±0.64^b	46.37±0.68^a
	HE	142.81±7.06^a	50.99±1.16^a	40.44±0.19^a	27.32±0.39^b
	EOH	88.95±4.12^b	46.69±4.64^a	35.45±0.56^c	14.63±0.15^c
Thyme	AqE	78.02±4.38^a	26.26±1.22^b	24.36±0.45^c	26.40±1.46^b
	HE	84.42±5.87^a	37.01±2.32^a	36.69±0.53^a	40.75±4.24^a
	EOH	76.19±0.45^a	25.65±0.53^b	29.87±0.61^b	14.19±0.73^c
Chervil	AqE	44.63±7.12^b	17.05±1.66^b	19.62±3.74^a	12.16±0.17^b
	HE	66.87±1.08^a	20.12±2.93^{ab}	15.85±0.35^{ab}	25.77±0.48^a
	EOH	30.52±7.26^b	25.34±1.38^a	11.48±0.24^b	8.99±0.03^c
Chive	AqE	72.24±4.59^a	18.12±0.53^a	33.50±1.44^a	29.31±0.21^b
	HE	55.41±0.89^b	16.12±1.66^{ab}	33.50±1.45^a	38.59±1.29^a
	EOH	44.32±5.37^c	15.05±0.53^a	32.04±0.11^a	37.81±1.63^a

Values consist of the mean ± standard deviation. Different letters differ significantly by Tukey's test ($p < 0.05$).
*GAE = Gallic acid equivalent.

rpm. Encapsulation efficiency was calculated according to the equation below:

$$EE(\%) = \frac{TPCe}{TPCi} \times 100 \quad (2)$$

TPCe corresponds to the total phenolic content encapsulated in the capsules, and TPCi corresponds to the initial total phenolic content of the solution used in the encapsulation process.

2.9. Fourier-transform infrared spectroscopy (FTIR)

Fourier-transform infrared spectroscopy (FTIR) was used to measure changes in the chemical structure of capsules filled with sodium alginate and the bioactive compound from hydroethanolic extracts of basil (*Ocimum basilicum*), curly leaf parsley (*Petroselinum crispum*), rosemary (*Rosmarinus officinalis*), thyme (*Thymus vulgaris*), and chervil (*Anthriscus cerefolium*) and aqueous extract of chive (*Allium fistulosum*).

The emission spectra were acquired by the attenuated total reflectance (ATR) technique in an FT-IR spectrometer (*Bruker Vertex 70v*) with the Bruker Platinum A255 ATR unit accessory. The sample was placed on a diamond crystal cell (2 × 2 mm), operating on a single reflection mode with a 45° incidence angle. The spectra were acquired in the spectral range of 4000 to 600 cm^{-1}, with a resolution of 4 cm^{-1}, the final result being the average of 64 measurements.

2.10. Differential scanning calorimetry (DSC)

Differential scanning calorimetry was used to evaluate the thermal stability of sodium alginate capsules loaded with the bioactive compound of hydroethanolic extracts of basil (*Ocimum basilicum*), curly leaf parsley (*Petroselinum crispum*), rosemary (*Rosmarinus officinalis*), thyme (*Thymus vulgaris*), and chervil (*Anthriscus cerefolium*) and aqueous extract of chive (*Allium fistulosum*).

Thermograms were obtained using a Netzsch DSC 204 F1 Nevio instrument. Each sample (5 mg) was accurately weighed into aluminum crucibles (40 μL). The crucibles were sealed and kept isothermally at 25 °C for 1 min, DSC scanning was performed from 25 °C to 350 °C at a heating rate of 10 °C/min^{-1} under dry nitrogen purge of 50 mL/min^{-1}.

2.11. Statistical analysis

The experiments were conducted according to a randomized design with three experimental replicates ($n = 3$) and four analytical replicates. The results were submitted to Analysis of Variance (ANOVA) with subsequent Tukey's range test at the level of 5% probability ($p < 0.05$) using the Statistical Software Action Stat.

3. Results and discussion

3.1. Total phenolic compound content and the determination of the antioxidant capacity

As shown in Table 1, the results of total phenolic compounds (TPC) and antioxidant capacity, evaluated by the •DPPH, ABTS•+ and FRAP methods of basil, parsley, rosemary, thyme, chervil, and chives extracts, show us that the TPC levels presented a variation between (142.81±7.06 mg GAE/g) for the hydroethanolic extract of rosemary and (30.52±0.95 mg GAE/g) for the ethanolic extract of chervil.

Slimestad et al. (2020) more significant quantities of phenolic compounds in methanol extracts of rosemary, when TPC's content was analyzed in ten most essential herbs in Norwegian commercial kitchens.

Castronuovo et al. (2019) reported that when evaluating samples of basil in methanolic extracts at 80%, the TPC values ranged from 54.1±3.5 to 48.8±2.0 mg GAE/g, with lower values when compared to the results of aqueous extracts and hydroethanolic extracts in this study, which found values of 63.54 and 76.30 mg GAE/g respectively, and higher values when compared to the results found for the 34.74mg GAE/g ethanol extract. Our results are superior to those found by Tang et al. (2015), who measured the total phenolic content in parsley as 42.31±0.50 mg GAE/g, and who found 21.63±1.81mg GAE/g. However, El-Sayed et al. (2018) reported that parsley exhibited a higher level of polyphenols (121.95±2.15mg GAE/g of extract).

The total phenolic content in chives from these extracts was higher than those prepared by Huang et al. (2009), who found 18.1±0.5 mg GAE/g in acidified methanol extract.

According to the data presented, it is observed that the concentration of phenolic compounds found in thyme showed values higher than those reported by Habashy et al. (2018), who found a maximum concentration of 44.16 mg GAE/g in aqueous extracts of thyme and lower than those found by Aouam et al. (2019) of 135.8 mg GAE/g for ethanolic extracts and 88.7 mg GAE/g for aqueous extracts.

These variations in the total polyphenol content may be related to genetic variations, growing conditions, extraction time or extraction solvent, and also free phenolic acids or derivatives present in the form of ester or ether, which is found in variable amounts in plant tissues in

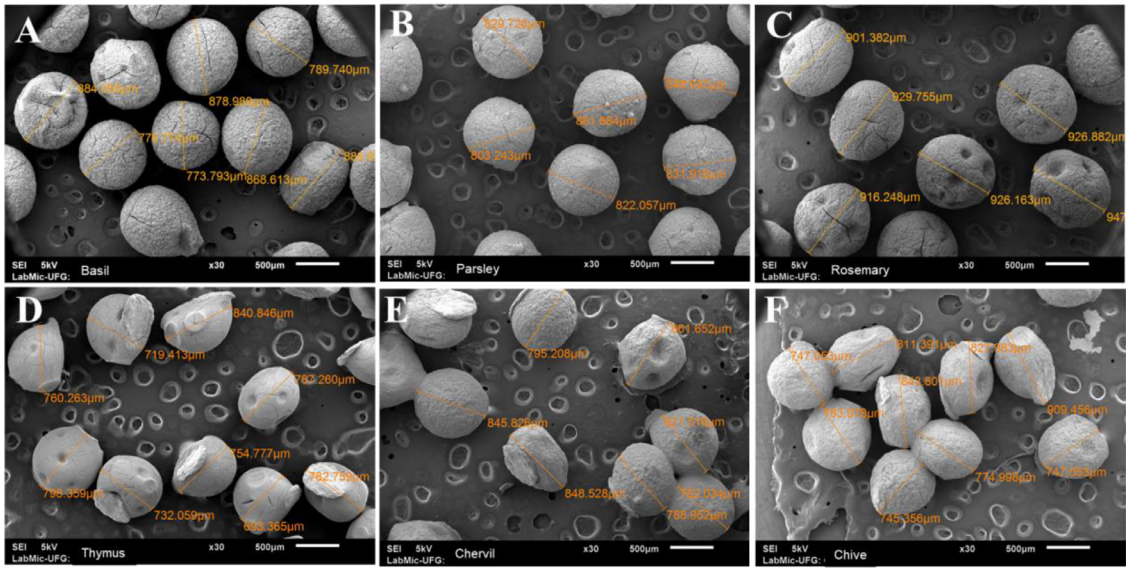

Fig. 1. Surface morphologies of microcapsules filled with (A) hydroethanolic extracts of basil (*Ocimum basilicum*), (B) parsley (*Petroselinum crispum*), (C) rosemary (*Rosmarinus officinalis*), (D) thyme (*Thymus vulgaris*), (E) chervil (*Anthriscus cerefolium*) and (F) aqueous extract of chive (*Allium fistulosum*), done by SEM, with a magnification of 30x.

response to characteristic synthetic patterns, resulting from encounters with different forms of environmental stress (Wong & Kitts, 2006).

Studies performed by Chang et al. (2013); Huang et al. (2009), who used alcoholic extracts of chive to study the antioxidant properties, demonstrated that the suppression capacity of the DPPH radical of the extracts was approximately 60–90%.

In the study by Tohidi et al. (2017), comparisons were made between ten species of *Thymus* spp. and investigated the ability of their extracts to eliminate the •DPPH free radical and found high antioxidant capacity.

Aouam et al. (2019) also evaluated the antioxidant activity of *Thymus riatarum* by the •DPPH method and found better results in ethanol extracts than aqueous extracts. In this study, the lowest antioxidant activity obtained by the DPPH method was for the ethanolic extract of parsley, and the highest antioxidant activity was for the hydroethanolic extract of rosemary.

The improved ABTS•+ method established by Re et al. (1999) was used successfully in this study to systematically evaluate the total antioxidant capacity of aromatic herb extracts, which is one of the most common methods, simple, fast, reliable, cheap, and also very adaptable to hydrophilic and lipophilic antioxidant systems.

Komes et al. (2011) reported a value of 24.40 µmol of Trolox/g of dry plant for the $ABTS^{\bullet+}$ elimination activity of an aqueous extract of *Thymus serpyllum*, results similar to that of this study which presented values of 24.36±0.45, 36.69±0.53, and 29.87±0.61µmol of Trolox/g for the aqueous, hydroethanolic and ethanolic extracts respectively, and the best results found by the $ABTS^{\bullet+}$ method were for the hydroethanolic extract of rosemary 40.44±0.19 µmol of Trolox/g and the worst ones were for the aqueous extract of basil 9.13±0.97 µmol Trolox/g.

Zengin et al. (2019) and Aouam et al. (2019) attested in their research an essential antioxidant activity of several extracts of *Thymus* spp. through the FRAP method and reported that the aqueous extract had significantly greater iron reducing power when compared to other extracts analyzed, the results obtained in this assay found higher ferric reducing power for the aqueous extract of rosemary 46.78±0.25 (µmol of FeSO4/g) and lower value for the ethanol extract of chervil 8.99±0.13 (µmol of FeSO4/g).

A variety of results from the evaluation of the antioxidant capacity of aromatic herb species extracts are available in the literature, with different types of methods and solvents used in the extraction, as well as other protocols used in carrying out these assays (Bahcesular et al., 2020; Derouich et al., 2020; Singh et al., 2020).

In light of the preceding, the need to carry out comprehensive tests that can integrate both the capacity of lipophilic and hydrophilic compounds based on their mechanisms (hydrogen transfer, electron transfer, and metal chelators) is justified (Zengin et al., 2019).

It is also worth mentioning that it is not possible to fully reflect the "total antioxidant capacity" of a plant extract with the application of only one method to assess antioxidant capacity.

3.2. Scanning electron microscopy of microcapsules

The surface morphologies of the microcapsules performed by scanning electron microscopy (SEM) are shown in Fig. 1.

The use of scanning electron microscopy revealed the presence of spherical, rugged, spongy-looking microparticles, presenting heterogeneous surface morphologies, characteristics of materials subjected to drying because of the partial collapse of the polymeric gel network (Aceval Arriola et al., 2016).

De Cássia Sousa Mendes et al. (2021) reported similar aspects and morphologies to this study in obtaining microcapsules loaded with extracts of jabuticaba skin and seeds (*Myrciaria cauliflora*).

3.2. Determination of the total content of phenolic compounds in microcapsules, microcapsule diameter, and encapsulation efficiency

According to Aguiar et al. (2016), capsules can be classified according to their size, structure, and morphology; concerning size, they can be named macrocapsules, microcapsules, or nanocapsules if they have diameters greater than 5000 μm, about 1–5000 μm or smaller than 1 µm, respectively. Thus, according to the diameter shown in Table 2, we can classify the capsules produced in this microcapsule study, demonstrating that the dripping method was adequate for the production of regular capsules, uniform size and desired diameter for microcapsules, ranging from 920.08±11.63 at 754.28±16.62µm, with encapsulation efficiency between 68.24±0.15 to 93.39±0.01%.

To obtain capsules with the desired size, with regular spheres and uniform size, it is necessary to control several factors such as the tip diameter, distance between the tip and the collecting solution, surface tension, and agitation speed (Dalponte Dallabona et al., 2020; Pasukamonset et al., 2016b).

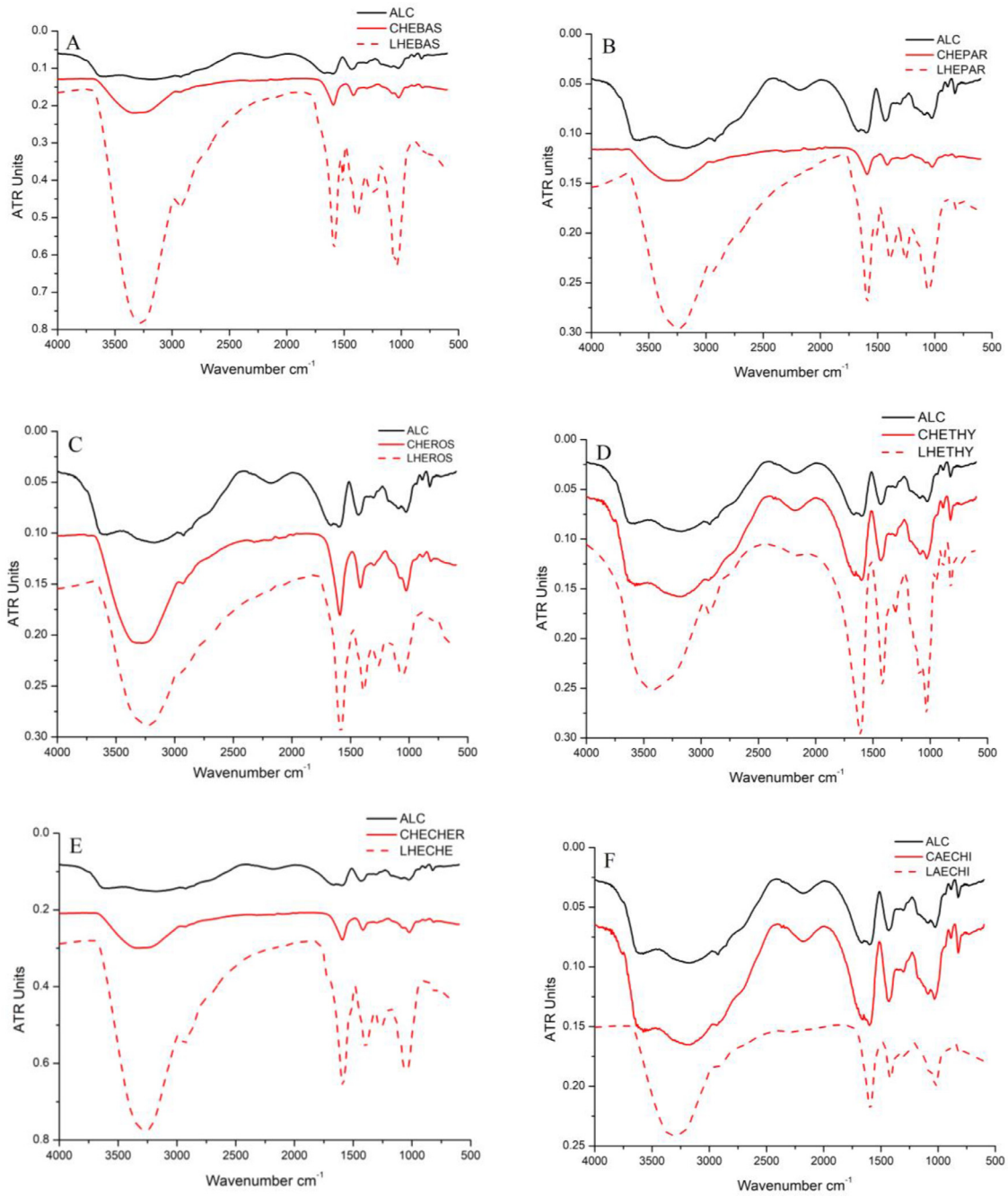

Fig. 2. FT-IR wavelengths. (ALC) Alginate capsules (without herbal extract), (CHEBAS) capsules loaded with hydroethanolic extract of basil (*Ocimum basilicum*), (CHEPAR) parsley (*Petroselinum crispum*), (CHEROS) rosemary (*Rosmarinus officinalis*), (CHETHY) thyme (*Thymus vulgaris*), (CHECHER) chervil (*Anthriscus cerefolium*), and (CAECHI) aqueous extract of chive (*Allium fistulosum*), and lyophilized extracts (LHEBAS) hydroethanolic extract of basil (*Ocimum basilicum*), (LHEPAR) curly leaf parsley (*Petroselinum crispum*), (LHEROS) rosemary (*Rosmarinus officinalis*), (LHETHY) thyme (*Thymus vulgaris*), (LHECHE) chervil (*Anthriscus cerefolium*), and (LAECHI) aqueous extract of chive (*Allium fistulosum*).

3.3. Fourier-transform infrared spectroscopy (FTIR)

FT-IR analysis allows identifying functional groups since each functional group absorbs radiation at a characteristic frequency of the infrared spectrum. The infrared spectra of samples of alginate capsules (without extract of the herbs) covering the area from 600 to 4000 cm−1, capsules filled with extract of aromatic herbs, and freeze-dried extract of herbs are shown in Fig. 2.

The peaks between 1400-1600 cm^{-1} correspond to the C=O of the aromatic ring, and the prominent peaks between 1400 and 1000 cm^{-1} are attributed to the C–O–C elongation vibrations of flavonoids (Arriola et al., 2019). The spectra showed the solid specific peaks in the region of 3450-3100 cm^{-1}. In general, strong peaks in this region correspond to the OH vibration, and the peaks between 1700-1600 cm^{-1} can also correspond to the symmetrical and asymmetrical elongation vibration for the carboxyl ion (COO); indicating the existence of carboxylic acid, ester, or carbonyl groups, these signs presented are in agreement with the results reported by por Dalponte Dallabona et al. (2020).

Arriola et al. (2019), Dalponte Dallabona et al. (2020) e Perumal et al. (2016) stated that wavelengths at 1504-1360 cm^{-1} correspond to the CO of the aromatic ring, and the prominent peaks between 1400 and 1000 cm^{-1} are attributed to elongation vibrations C–O–C and

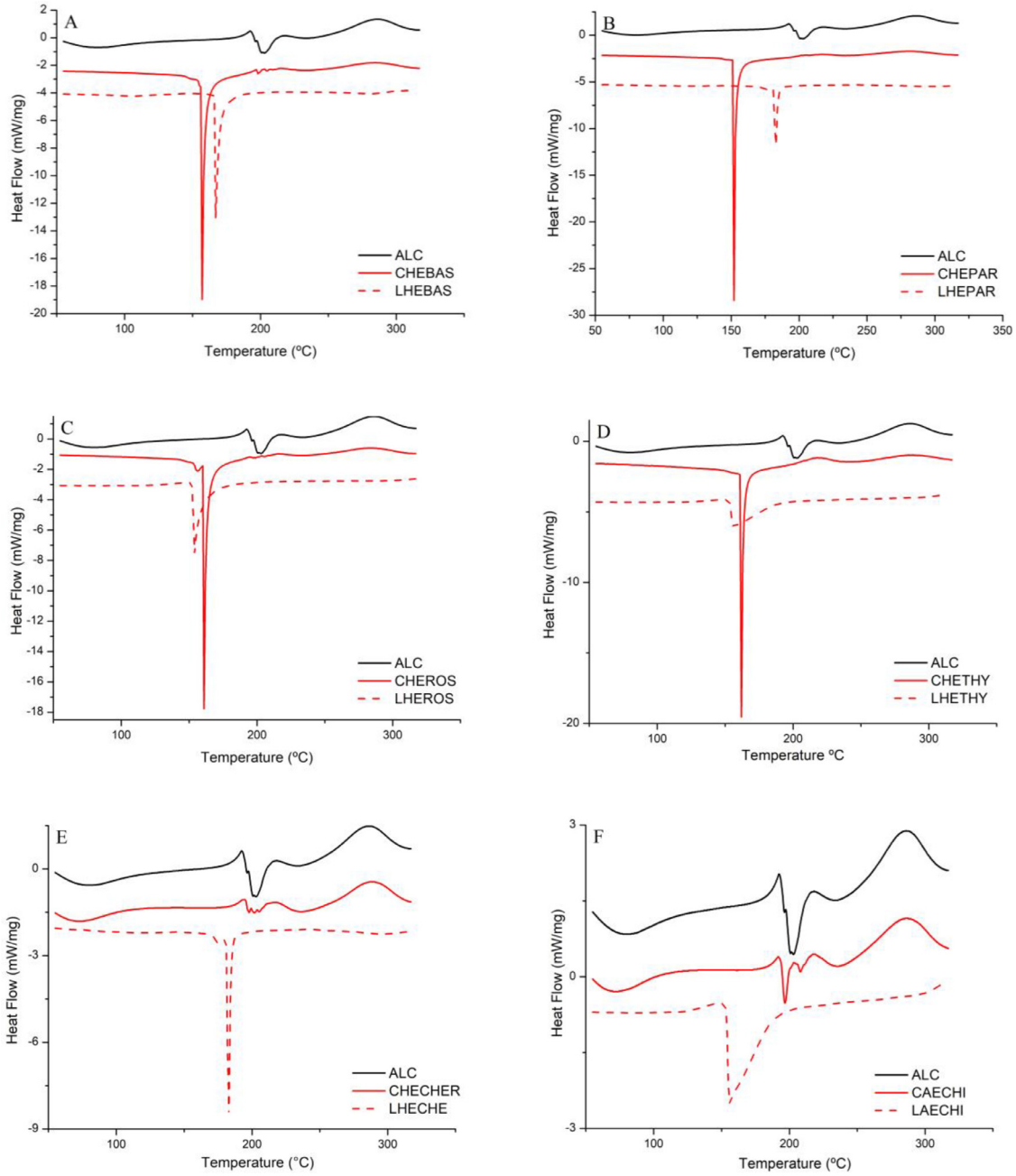

Fig. 3. DSC thermograms. (ALC) Alginate capsules (without herbal extract), (CHEBAS) capsules loaded with hydroethanolic extract of basil (*Ocimum basilicum*), (CHEPAR) parsley (*Petroselinum crispum*), (CHEROS) rosemary (*Rosmarinus officinalis*), (CHETHY) thyme (*Thymus vulgaris*), (CHECHER) chervil (*Anthriscus cerefolium*), and (CAECHI) aqueous extract of chive (*Allium fistulosum*), and lyophilized extracts (LHEBAS) hydroethanolic extract of basil (*Ocimum basilicum*), (LHEPAR) curly leaf parsley (*Petroselinum crispum*), (LHEROS) rosemary (*Rosmarinus officinalis*), (LHETHY) thyme (*Thymus vulgaris*), (LHECHE) chervil (*Anthriscus cerefolium*), and (LAECHI) aqueous extract of chive (*Allium fistulosum*).

bands between 1000 and 800 cm^{-1} may also be attributed to flexural vibration of the C=C bond, related to vibration of the aromatic ring. The results also showed similar peaks in the spectra of capsules filled with herbal extracts and freeze-dried extracts; such peaks did not appear in samples of alginate capsules (without extract), suggesting the successful encapsulation of herbal extracts.

3.4. Differential scanning calorimetry (DSC)

Thermal properties evaluated by DSC of samples (ALC) Alginate capsules (without herbal extract), (CHEBAS) capsules loaded with hydroethanolic extract of basil (*Ocimum basilicum*), (CHEPAR) parsley (*Petroselinum crispum*), (CHEROS) rosemary (*Rosmarinus officinalis*), (CHETHY) thyme (*Thymus vulgaris*), (CHECHER) chervil (*Anthriscus cerefolium*), and (CAECHI) aqueous extract of chive (*Allium fistulosum*), and lyophilized extracts (LHEBAS) hydroethanolic extract of basil (*Ocimum basilicum*), (LHEPAR) curly leaf parsley (*Petroselinum crispum*), (LHEROS) rosemary *(Rosmarinus officinalis*), (LHETHY) thyme (*Thymus vulgaris*), (LHECHE) chervil (*Anthriscus cerefolium*), and (LAECHI) aqueous extract of chive (*Allium fistulosum*) are shown in Fig. 3.

The DSC curves of the empty alginate capsules (ALC) showed an endothermic peak at 196°C. In contrast, the endothermic peaks mentioned in the thermograms of the capsule samples loaded with herbal extracts were displayed at lower temperatures, (CHEBAS) 157°C, (CHEPAR) 152°C, (CHEROS) 159°C, (CHETHY) 162°C, (CHECHER) 194°C and (CAECHI) 193°C, indicating a decrease in thermal resistance compared

Table 2
Efficiency of encapsulation (EE), TPC values of hydroethanolic extracts of basil (*Ocimum basilicum*), parsley (*Petroselinum crispum*), rosemary (*Rosmarinus officinalis*), thyme (*Thymus vulgaris*), chervil (*Anthriscus cerefolium*), and aqueous extract of chive (*Allium fistulosum*), and the diameters of the microcapsules.

Herbs	TPC_e (mg GAE/g^1)**	TPC_i (mg GAE/g^1)**	EE (%)	Diameter (μm)
Basil	64.35±2.03	76.30±2.54	76.70±0.18	839.09±52.94
Parsley	67.37±1.23	79.01±2.68	74.59±0.13	826.59±15.66
Rosemary	134.43±4.72	142.81±7.06	93.39±0.01	920.08±11.63
Thyme	72.09±1.64	84.42±5.87	83.50±0.17	842.81±69.60
Chervil	44.40±0.31	66.87±1.08	68.24±0.15	823.17±31.93
Chive	56.24±1.19	72.24±4.59	74.69±0.10	754.28±16.62

*Values consist of the mean (n = 3) ± standard deviation. TPCi initial total phenolic content of the extract solution used for the encapsulation process, TPCe Encapsulated total phenolic compounds. *GAE = Gallic acid equivalents.

to capsules without extract. This thermal behavior was also observed by Pasukamonset et al. (2016a).

They demonstrated a reduction in thermal resistance in capsules loaded with Clitoria ternatea extracts, decomposition peak at 188°C when compared to alginate capsules without extract, which showed decomposition peak at 197.8°C.

This behavior can be explained by the degree of complexity of the structure of the samples, for instance, the difference in molecular weight, electrostatic interactions, moisture content, phenolic compounds contents (Ezati & Rhim, 2020; Han & Song, 2021; Soleimanifar et al., 2021; Valencia et al., 2021).

On the other hand, for the freeze-dried extracts, the phase change occurred at lower temperatures, for most of the samples evaluated, compared to the endothermic peaks presented in the samples of microencapsulated extracts, indicating that the microencapsulation increased the thermal stability of the extracts.

Similar data were reported by Pasukamonset et al. (2016a) and Ra et al. (2014). They demonstrated increased thermal resistance in capsules loaded with Clitoria ternatea extracts and in capsules loaded with resveratrol, respectively, compared to decomposition peaks of the extracts before being microencapsulated. It is important to emphasize that this phase change process significantly influences the practical applications of extracts or microcapsules loaded with extracts.

Furthermore, Do et al. (2021) reported that when only one endothermic peak appears during the melting process, it indicates stable phase transfer material. This behavior was observed in most of the evaluated samples, suggesting that microencapsulated herb extracts may have good heat resistance.

4. Conclusion

The results obtained in this study showed that in the six species of herbs evaluated, the levels of phenolic compounds and the antioxidant capacity were significant, with more expressive results for rosemary HE.

Microscopic observation of the microcapsules obtained under ideal conditions revealed that the dripping method was adequate for producing microcapsules with regular spheres and uniform size.

The evaluation of the effectiveness of microencapsulation in the protection of bioactive compounds from aromatic herbs by determining the content of total phenolic compounds reached an efficiency of up to 93.39±0.01%, which is considered a high efficacy.

In light of the preceding, the application of microencapsulated aromatic herb extracts may present a potential alternative in preserving food and beverages.

Declaration of Competing interest

The authors declare that all are aware of the content of the manuscript and, in agreement with its submission, declare that there is no financial, personal or institutional conflict of interest with the information and results disclosed through this manuscript.

Acknowledgments

The authors are grateful to FAPEG (Research Foundation of the State of Goiás), who funded this study, with grant number (201810267000660), to the Institute of Education Science and Technology Goiano - Campus Morrinhos and the company *ISLA sementes*.

References

Aceval Arriola, N. D., De Medeiros, P. M., Prudencio, E. S., Olivera Müller, C. M., & De Mello Castanho Amboni, R. D. (2016). Encapsulation of aqueous leaf extract of Stevia rebaudiana Bertoni with sodium alginate and its impact on phenolic content. *Food Bioscience, 13*, 32–40. 10.1016/j.fbio.2015.12.001.

Aguiar, J., Estevinho, B.N., .& Santos, L. (2016). Microencapsulation of natural antioxidants for food application – The specific case of coffee antioxidants – A review. Trends in Food Science and Technology, 58, 21–39. 10.1016/j.tifs.2016.10.012

Aouam, I., EL Atki, Y., Taleb, M., Taroq, A., EL Kamari, F., Lyoussi, B. et al. (2019). Antioxidant capacities and total phenolic contents of thymus riatarum. Materials Today: Proceedings, 13, 579–586. 10.1016/j.matpr.2019.04.016

Arriola, N. D. A., Chater, P. I., Wilcox, M., Lucini, L., Rocchetti, G., Dalmina, M., et al. (2019). Encapsulation of stevia rebaudiana Bertoni aqueous crude extracts by ionic gelation – Effects of alginate blends and gelling solutions on the polyphenolic profile. *Food Chem., 275*, 123–134 April 2018. 10.1016/j.foodchem.2018.09.086.

Bahcesular, B., Diraz, E., Karaçocuk, M., Kulak, M., & Karaman, S. (2020). Industrial Crops & Products Seed priming with melatonin effects on growth, essential oil compounds and antioxidant activity of basil (Ocimum basilicum L .) under salinity stress. Industrial Crops & Products, 146(July 2019), 112165. 10.1016/j.indcrop.2020.112165

Benzie, I. F. F., & Strain, J. J. (1996). The Ferric Reducing Ability of Plasma (FRAP) as a Measure of "‘ Antioxidant Power. *The FRAP Assay, 76*, 70–76.

Borguini, R.G., .Ferraz, E.A., .Silva, D., Ferraz, E.A., .& Silva, D.A. (.2009). Tomatoes and Tomato Products as Dietary Sources of Antioxidants Tomatoes and Tomato Products as Dietary Sources of Antioxidants. 9129. 10.1080/87559120903155859

Castronuovo, D., Russo, D., Libonati, R., Faraone, I., Candido, V., Picuno, P., et al. (2019). Influence of shading treatment on yield, morphological traits and phenolic profile of sweet basil (Ocimum basilicum L.). *Scientia Horticulturae, 254*(September 2018), 91–98. 10.1016/j.scienta.2019.04.077.

Celano, R., Piccinelli, A. L., Pagano, I., Roscigno, G., Campone, L., De Falco, E., et al. (2017). Oil distillation wastewaters from aromatic herbs as new natural source of antioxidant compounds. *Food Res. Int., 99*, 298–307 March. 10.1016/j.foodres.2017.05.036.

Chang, T.-. C., Chang, H.-. T., Chang, S.-. T., Lin, S.-. F., Chang, Y.-. H., & Jang, H.-. D. (2013). A Comparative Study on the Total Antioxidant and Antimicrobial Potentials of Ethanolic Extracts from Various Organ Tissues of <i>Allium</i> spp. *Food and Nutrition Sciences, 04*(08), 182–190. 10.4236/fns.2013.48a022.

Cuvelier, M.E., .& Berset, C. (1995). Use of a Free Radical Method to Evaluate Antioxidant Activity. 30, 25–30.

Dalponte Dallabona, I., de Lima, G. G., Cestaro, B. I., Tasso, I. de S., Paiva, T. S., Laureanti, E. J. G., et al. (2020). Development of alginate beads with encapsulated jabuticaba peel and propolis extracts to achieve a new natural colorant antioxidant additive. *Int. J. Biol. Macromol., 163*, 1421–1432. 10.1016/j.ijbiomac.2020.07.256.

de Cássia Sousa Mendes, D., Asquieri, E. R., Batista, R. D., de Morais, C. C., Ramirez Ascheri, D. P., de Macêdo, I. Y. L., et al. (2021). Microencapsulation of jabuticaba extracts (Myrciaria cauliflora): Evaluation of their bioactive and thermal properties in cassava starch biscuits. *Lwt, 137* August 2020. 10.1016/j.lwt.2020.110460.

Derouich, M., Bouhlali, E. D. T., Hmidani, A., Bammou, M., Bourkhis, B., Sellam, K., et al. (2020). Assessment of total polyphenols, flavonoids and anti-inflammatory potential of three Apiaceae species grown in the Southeast of Morocco. *Scientific African,, 9*. 10.1016/j.sciaf.2020.e00507.

Do, J. Y., Son, N., Shin, J., Chava, R. K., Joo, S. W., & Kang, M. (2021). n-Eicosane-Fe3O4@SiO2@Cu microcapsule phase change material and its improved thermal conductivity and heat transfer performance. *Mater. Des., 198*, Article 109357. 10.1016/j.matdes.2020.109357.

Elena, G., & St, N. (2021). Microencapsulation of lycopene from tomatoes peels by complex coacervation and freeze-drying : Evidences on phytochemical profile, stability and food applications. 288(March 2020). 10.1016/j.jfoodeng.2020.110166

El-Sayed, M. M., Metwally, N. H., Ibrahim, I. A., Abdel-Hady, H., & Abdel-Wahab, B. S. A. (2018). Antioxidant Activity, Total Phenolic and Flavonoid Contents of Petroselinum crispum Mill. *Journal of Applied Life Sciences International, 19*(2), 1–7. 10.9734/jalsi/2018/45113.

El-sayed, S. M., & Youssef, A. M. (2019). Heliyon Potential application of herbs and spices and their effects in functional dairy products. *Heliyon, 5*, e01989 November 2018. 10.1016/j.heliyon.2019.e01989.

Ezati, P., & Rhim, J. W. (2020). pH-responsive pectin-based multifunctional films incorporated with curcumin and sulfur nanoparticles. *Carbohydr. Polym., 230*, Article 115638 October 2019. 10.1016/j.carbpol.2019.115638.

Giannenas, I., Sidiropoulou, E., Bonos, E., & Christaki, E. (2020). *Feed additives* perspectives. Elsevier Inc. 10.1016/B978-0-12-814700-9.00001-7.

Habashy, N. H., Abu Serie, M. M., Attia, W. E., & Abdelgaleil, S. A. M. (2018). Chemical characterization, antioxidant and anti-inflammatory properties of Greek Thymus vulgaris extracts and their possible synergism with Egyptian Chlorella vulgaris. *J. Funct. Foods, 40*, 317–328 November 2017. 10.1016/j.jff.2017.11.022.

Han, H. S., & Song, K. Bin (2021). Noni (Morinda citrifolia) fruit polysaccharide films containing blueberry (Vaccinium corymbosum) leaf extract as an antioxidant packaging material. *Food Hydrocolloids., 112*, Article 106372 September 2020. 10.1016/j.foodhyd.2020.106372.

Huang, Z., Wang, B., Eaves, D. H., Shikany, J. M., & Pace, R. D. (2009). Total phenolics and antioxidant capacity of indigenous vegetables in the southeast United States: Alabama Collaboration for Cardiovascular Equality Project. *Int. J. Food Sci. Nutr., 60*(2), 100–108. 10.1080/09637480701605715.

Komes, D., Belščak-Cvitanović, A., Horžić, D., Rusak, G., Likić, S., & Berendika, M. (2011). Phenolic composition and antioxidant properties of some traditionally used medicinal plants affected by the extraction time and hydrolysis. *Phytochem. Anal., 22*(2), 172–180. 10.1002/pca.1264.

Lee, Y., & Chang, Y. H. (2020). International Journal of Biological Macromolecules Microencapsulation of a maca leaf polyphenol extract in mixture of maltodextrin and neutral polysaccharides extracted from maca roots. *Int. J. Biol. Macromol., 150*, 546–558. 10.1016/j.ijbiomac.2020.02.091.

Mar, J. M., Silva, L. S., Rabelo, M. da S., Muniz, M. P., Nunomura, S. M., & Correa, R. F. (2020). Encapsulation of Amazonian Blueberry juices: Evaluation of bioactive compounds and stability. *Lwt, 124*, Article 109152 January. 10.1016/j.lwt.2020.109152.

Pasukamonset, P., Kwon, O., & Adisakwattana, S. (2016a). Alginate-based encapsulation of polyphenols from Clitoria ternatea petal flower extract enhances stability and biological activity under simulated gastrointestinal conditions. *Food Hydrocolloids., 61*, 772–779. 10.1016/j.foodhyd.2016.06.039.

Pasukamonset, P., Kwon, O., & Adisakwattana, S. (2016b). Food Hydrocolloids Alginate-based encapsulation of polyphenols from Clitoria ternatea petal fl ower extract enhances stability and biological activity under simulated gastrointestinal conditions. *Food Hydrocolloids., 61*, 772–779. 10.1016/j.foodhyd.2016.06.039.

Perumal, V., Manickam, T., Bang, K. S., Velmurugan, P., & Oh, B. T. (2016). Antidiabetic potential of bioactive molecules coated chitosan nanoparticles in experimental rats. *Int. J. Biol. Macromol., 92*, 63–69. 10.1016/j.ijbiomac.2016.07.006.

Potortì, A. G., Bua, G. D., Lo Turco, V., Ben Tekaya, A., Beltifa, A., Ben Mansour, H., et al. (2020). Major, minor and trace element concentrations in spices and aromatic herbs from Sicily (Italy) and Mahdia (Tunisia) by ICP-MS and multivariate analysis. *Food Chem., 313*, Article 126094 April 2019. 10.1016/j.foodchem.2019.126094.

Prat, D., Wells, A., Hayler, J., Sneddon, H., McElroy, C. R., Abou-Shehada, S., et al. (2015). CHEM21 selection guide of classical- and less classical-solvents. *Green Chem., 18*(1), 288–296. 10.1039/c5gc01008j.

Putnik, P., Kovačević, D. B., Penić, M., Fegeš, M., & Dragović-Uzelac, V. (2016). Microwave-Assisted Extraction (MAE) of Dalmatian Sage Leaves for the Optimal Yield of Polyphenols: HPLC-DAD Identification and Quantification. *Food Anal. Methods, 9*(8), 2385–2394. 10.1007/s12161-016-0428-3.

Ra, A., Yong, C., Chun, G., Keun, B., & Park, D.J. (.2014). Preparation of alginate – CaCl 2 microspheres as resveratrol carriers. 4612–4619. 10.1007/s10853-014-8163-x

Rakmai, J., Cheirsilp, B., Mejuto, J. C., Simal-Gándara, J., & Torrado-Agrasar, A. (2018). Antioxidant and antimicrobial properties of encapsulated guava leaf oil in hydroxypropyl-beta-cyclodextrin. *Ind. Crops Prod., 111*, 219–225 April 2017. 10.1016/j.indcrop.2017.10.027.

Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., & Rice-Evans, C. (1999). Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radic. Biol. Med., 26*(9), 1231–1237. 10.1016/S0891-5849(98)00315-3.

Rezaei, A., Fathi, M., & Jafari, S. M. (2019). Nanoencapsulation of hydrophobic and low-soluble food bioactive compounds within different nanocarriers. *Food Hydrocolloids., 88*, 146–162 June 2018. 10.1016/j.foodhyd.2018.10.003.

Sharma, S., Cheng, S., Bhattacharya, B., & Chakkaravarthi, S. (2019). Trends in Food Science & Technology E ffi cacy of free and encapsulated natural antioxidants in oxidative stability of edible oil : Special emphasis on nanoemulsion-based encapsulation. *Trends Food Sci. Technol., 91*, 305–318 August 2018. 10.1016/j.tifs.2019.07.030.

Singh, P., Bajpai, V., Khandelwal, N., Varshney, S., Gaikwad, A. N., Srivastava, M., et al. (2020). of. *J. Pharm. Biomed. Anal.*, Article 113707. 10.1016/j.jpba.2020.113707.

Slimestad, R., Fossen, T., & Brede, C. (2020). Flavonoids and other phenolics in herbs commonly used in Norwegian commercial kitchens. *Food Chem., 309*, Article 125678 April 2019. 10.1016/j.foodchem.2019.125678.

Soleimanifar, M., Jafari, S. M., Assadpour, E., & Mirarab, A. (2021). Electrosprayed whey protein nanocarriers containing natural phenolics; thermal and antioxidant properties, release behavior and stability. *J. Food Eng., 307*, Article 110644 April. 10.1016/j.jfoodeng.2021.110644.

Tarone, A.G., .Baú, C., Cazarin, B., Roberto, M., & Junior, M. (2020). Anthocyanins nthocyanins0" type="Other">ther">l., 2020" type="Othe. 133(December 2019). 10.1016/j.foodres.2020.109092

Teng, X., Zhang, M., & Devahastin, S. (2019). New developments on ultrasound-assisted processing and flavor detection of spices: A review. *Ultrason. Sonochem., 55*, 297–307 January. 10.1016/j.ultsonch.2019.01.014.

Tohidi, B., Rahimmalek, M., & Arzani, A. (2017). Essential oil composition, total phenolic, flavonoid contents, and antioxidant activity of Thymus species collected from different regions of Iran. *Food Chem., 220*, 153–161. 10.1016/j.foodchem.2016.09.203.

Valencia, M.S., .Júnior, Silva, da, M.F., Xavier-Júnior, F.H., .Veras, B.de O., Albuquerque, P.B.S.de et al. (2021). Characterization of curcumin-loaded lecithin-chitosan bioactive nanoparticles. Carbohydrate Polymer Technologies and Applications, 2, 100119. 10.1016/j.carpta.2021.100119

Vieitez, I., Maceiras, L., Jachmanián, I., & Alborés, S. (2018). Antioxidant and antibacterial activity of different extracts from herbs obtained by maceration or supercritical technology. *J. Supercrit. Fluids, 133*, 58–64 August 2017. 10.1016/j.supflu.2017.09.025.

Vinceković, M., Viskić, M., Jurić, S., Giacometti, J., Bursać Kovačević, D., Putnik, P., et al. (2017). Innovative technologies for encapsulation of Mediterranean plants extracts. *Trends Food Sci. Technol., 69*, 1–12. 10.1016/j.tifs.2017.08.001.

Waterhouse, Andrew L (2002). *Determination of total phenolics. Current Protocols in Food Analytical Chemistry, 6*. I1.1.1-I1.1.8. https://www.researchgate.net/profile/Roberto_Molteni5/post/How_can_get_accurate_total_phenolic_contents_by_Folin_Ciocalteus_reagent_method/attachment/5da38c6fcfe4a777d4e60a5c/AS%3A813644635992066%401570999406997/download/2002+total+polyphenol+methods.pdf .

Wong, P. Y. Y., & Kitts, D. D. (2006). Studies on the dual antioxidant and antibacterial properties of parsley (Petroselinum crispum) and cilantro (Coriandrum sativum) extracts. *Food Chem., 97*(3), 505–515. 10.1016/j.foodchem.2005.05.031.

Zengin, G., Atasagun, B., Zakariyyah, M., Saleem, H., Mollica, A., Babak, M., et al. (2019). Industrial Crops & Products Phenolic pro fi ling and in vitro biological properties of two Lamiaceae species (Salvia modesta and Thymus argaeus): A comprehensive evaluation. *Industrial Crops & Products, 128*, 308–314 October 2018. 10.1016/j.indcrop.2018.11.027.