2018

Microencapsulated ascorbic acid: Development, characterization, and release profile in simulated gastrointestinal fluids

Michele Cristina Rodrigues da Cruz
Federal University of Paraná, Curitiba, Brazil

Camila Augusto Perussello
Technological University Dublin, camila.perussello@tudublin.ie

Maria Lúcia Masson
Federal University of Paraná, Curitiba, Brazil

Follow this and additional works at: <https://arrow.tudublin.ie/ehsiart>

Part of the [Food Science Commons](#), and the [Medicine and Health Sciences Commons](#)

Recommended Citation

da Cruz, MCR, Perussello, CA, Masson, ML. Microencapsulated ascorbic acid: Development, characterization, and release profile in simulated gastrointestinal fluids. *J Food Process Eng.* 2018; 41:e12922. <https://doi.org/10.1111/jfpe.12922>

This Article is brought to you for free and open access by the ESHI Publications at ARROW@TU Dublin. It has been accepted for inclusion in Articles by an authorized administrator of ARROW@TU Dublin. For more information, please contact arrow.admin@tudublin.ie, aisling.coyne@tudublin.ie, gerard.connolly@tudublin.ie.
Funder: CAPES Brazil—Coordination for the Improvement of Higher Education Personnel

ORIGINAL ARTICLE

Microencapsulated ascorbic acid: Development, characterization, and release profile in simulated gastrointestinal fluids

Michele Cristina Rodrigues da Cruz¹ | Camila Augusto Perussello² | Maria Lúcia Masson¹

¹Department of Chemical Engineering, Graduate Program in Food Engineering, Federal University of Paraná, Curitiba, Brazil

²Environmental Sustainability and Health Institute, Dublin Institute of Technology, Dublin, Ireland

Correspondence

Michele Cristina Rodrigues da Cruz, Department of Chemical Engineering, Graduate Program in Food Engineering, Federal University of Paraná, Av. Francisco Hoffmann dos Santos s.n., Curitiba CEP 81530-900, PR, Brazil.
Email: michelercruz@yahoo.com.br

Funding information

Coordenação de Aperfeiçoamento de Pessoal de Nível Superior; Coordination for the Improvement of Higher Education Personnel

Abstract

The aim of this study was to produce ascorbic acid (AA) microcapsules by complex coacervation, characterize them, and evaluate the release profile in simulated gastrointestinal fluids. The microcapsules were made of gelatin and pectin at concentrations of 1.0, 2.0, and 3.0%. The coacervated microcapsules were assessed for morphology (by optical microscopy and scanning electron microscopy), water activity (A_w), hygroscopicity, solubility, encapsulation efficiency (EE), thermal behavior, and AA release profile. The AA microcapsules were characterized by low solubility, low hygroscopicity, high thermal stability, and low diameter ($<10\ \mu\text{m}$), with EE ranging from 23.7 to 94.3%. The release profile was faster in gastric fluid (pH 1.8) than in intestinal fluid (pH 6.8). The sample containing 2.0% of wall material and 50% (w/w) of the simple emulsion (core) had the highest EE and the best in vitro release profile, with release of only 68% of AA in 120 min in the gastric fluid.

Practical applications

Ascorbic acid (AA) is widely used in the food industry as an antioxidant, but practical use becomes limited due to its high instability. Microencapsulation was proposed as an alternative to increase the stability of this compound. Gelatin and pectin were used as wall materials in the production of ascorbic acid microcapsules by complex coacervation. Both solubility and hygroscopicity were low while the thermal stability of the active material improved with the microencapsulation process. Ascorbic acid release was more pronounced in gastric fluid (pH 1.8) and slower in intestinal fluid (pH 6.8). The results of this study demonstrate that the microcapsules produced represent a feasible method of increasing AA availability in food products, especially for those samples that presented high encapsulation efficiency.

1 | INTRODUCTION

Ascorbic acid (AA) is a bioactive compound known for its health benefits as it is a source of vitamin C. In addition to the nutritional function, AA is used as antioxidant in meat products and to reduce enzymatic browning in vegetables and fresh fruits (Abbas, Da Wei, Hayat, & Xiaoming, 2012; Damodaran, Parkin, & Fennema, 2010). However, this additive is highly unstable and reactive, undergoing rapid degradation when exposed to temperature and pH variations, light, high concentrations of oxygen, and high water activity (Damodaran et al., 2010). Due to its high instability, the incorporation of AA into food

matrices is not straightforward. The food industry has been using microencapsulation to increase the stability of heat labile compounds, as well as to control their release into the desired medium (Abbas et al., 2012; Dias, Ferreira, & Barreiro, 2015).

Microencapsulation is defined as a process in which the active material is surrounded by a coating or wall, forming small capsules that protect the encapsulated material from adverse external conditions (i.e., light, heat, moisture, oxygen, interaction with other compounds) and hence increase its stability, while maintaining the viability of the active compound throughout processing and storage (Abbas et al., 2012; Desai, Liu, & Park, 2006; Dias et al., 2015; Gharsallaoui,

Roudaut, Chambin, Voilley, & Saurel, 2007; Gibbs, Kermasha, Alli, & Mulligan, 1999; Shahidi & Han, 1993).

Different methods of microencapsulation have been studied to improve the stability of AA, such as spray chilling (De Matos, Comunian, Thomazini, & Favaro-Trindade, 2017; Matos, Di Sabatino, Passerini, Favaro-Trindade, & Albertini, 2015) spray drying (Alvim, Stein, Koury, Dantas, & Cruz, 2016; Desai & Park, 2005), microfluidic technique (Comunian, Abbaspourrad, Favaro-Trindade, & Weitz, 2014; Comunian et al., 2017), solvent evaporation (Uddin, Hawlader, & Zhu, 2001) and complex coacervation (Comunian et al., 2013; Rodrigues da Cruz, Andreotti Dagostin, Perussello, & Masson, 2019).

Microencapsulation via coacervation is one of the methods used to encapsulate heat labile and unstable compounds as it produces a true capsule, completely protecting the active compound within the wall material (Dong et al., 2011; Nori et al., 2011). Complex coacervation is the phase separation of two hydrocolloids from the initial solution with subsequent deposition of the coacervate phase around the active ingredient suspended or emulsified in the same reaction medium (De Kruif, Weinbreck, & De Vries, 2004; Eghbal & Choudhary, 2018; Gouin, 2004). Complex coacervation can be used for the formation of films for packaging and the production of emulsions or food gels. The coacervates formed are also used as additives, emulsifiers, and viscosity modifiers in the food industry (Eghbal & Choudhary, 2018; Sing, 2017). In addition, the microcapsules produced by complex coacervation are resistant to high temperatures and have good characteristics for controlled release (Dong et al., 2011).

The controlled release of bioactive compounds ensures the preservation of the nutritional and sensory characteristics of the food, prolonging its shelf life. The release mechanisms depend on many factors, such as the wall material, the substance to be encapsulated, the ratio between wall material and core, the geometry and morphology of the microcapsule, and the release conditions (i.e., solvent, pH, ionic strength, temperature) (Dong et al., 2011; Eghbal & Choudhary, 2018; Maderuelo, Zarzuelo, & Lanao, 2011; Moschakis & Biliaderis, 2017). Most studies on microencapsulation by complex coacervation involves hydrophobic agents, such as in the research carried out by Dong et al. (2011) with peppermint oil, and by Rutz et al. (2017) with palm oil. However, hydrophilic active agents, such as AA, cannot be encapsulated directly by complex coacervation, requiring an additional step comprising the emulsification of the solution containing the active agent in the oil. This was done in the study conducted by Comunian et al. (2013) in the encapsulation of AA, by Rocha-Selmi, Theodoro, Thomazini, Bolini, & Favaro-Trindade (2013) in the encapsulation of sucralose, and by Santos et al. (2014) in the encapsulation of xylitol. There are only a few studies involving the release of hydrophilic compounds, such as AA, in simulated gastric and intestinal fluids. Therefore, the objective of this study was to develop microcapsules of AA, analyze their morphological, physicochemical, and thermal characteristics, and evaluate the *in vitro* release of microencapsulated AA in simulated gastric and intestinal fluids, aiming at their industrial application.

2 | MATERIALS AND METHODS

2.1 | Materials

Bovine gelatin type B (Gelita South América, Mococa, Brazil) and citrus pectin (Vetec-Sigma Aldrich, St. Louis, Brazil) were used as wall materials, while pure ascorbic acid (AA) (Biotec, Pinhais, Brazil) was used as core material. Polyglycerol polyricinoleate (PGPR 90) (SGS Agricultura e Indústria Ltda, Ponta Grossa, Brazil) was used as emulsifier, and corn oil (Cargill, Mairinque, Brazil) was used to produce the simple emulsion.

2.2 | Methods

2.2.1 | Preparation of microcapsules

The microcapsules were prepared based on the methodology described by Comunian et al. (2013), however the process conditions (oil phase: aqueous phase ratio, stirring rate, and stirring time) were modified throughout the experiments, aiming at an enhanced encapsulation efficiency (EE). One hundred milliliters of 1.0, 2.0, and 3.0% of pectin and gelatin solutions were prepared separately by heating at $45 \pm 2^\circ\text{C}$. The pH of each solution was adjusted to 7.5 using sodium hydroxide (1 mol/L) before making up the volume to 100 ml. A simple emulsion W/O was prepared with 30% (w/w) AA solution, corn oil at a 1:1 mass ratio (1 g of AA solution : 1 g of oil) and the lipophilic surfactant polyglycerol polyricinoleate (PGPR 90) at a concentration of 0.8% (w/w) in relation to the total mass of emulsion. The blend was homogenized with an Ultra-Turrax (T 25 digital, Ika, Germany) at 22,000 rpm for 4 min. The pH was adjusted again to 7.5 with NaOH 1 mol/L and this emulsion was considered as core and used in the calculations of wall:core proportions. Then the simple emulsion was added to 100 ml of aqueous gelatin solution at concentrations of 1.0, 2.0, and 3.0% at $45 \pm 2^\circ\text{C}$ for the formation of the double emulsion (W/O/W). The emulsification was carried out with an Ultra-Turrax homogenizer for 3 min at 14,000 rpm. The pH was adjusted again to 7.5 with NaOH 1 mol/L. The pectin solutions were slowly added to the double emulsions under constant magnetic stirring at $45 \pm 2^\circ\text{C}$ for 5 min. The pH was adjusted to 3.5 by addition of HCl (1 mol/L) to promote the complex coacervation. The solution was cooled to approximately $10 \pm 2^\circ\text{C}$ in an ice bath, and the coacervate material was stored for 24 hr at 7°C to allow phase separation. After 24 hr, the water was discarded, the samples were centrifuged at 3,600 rpm for 15 min, the coacervates were transferred to vials coated with aluminum foil, and were then frozen (-18°C) for 24 hr and freeze-dried (LIOBRAS, LIOTOP model L101) at -52°C for 48 hr. After freeze-dried, the samples were added into glass vials coated with aluminum foil and stored in a desiccator. To select the best ratio between the core and the polymer pair, a 3^2 full-factorial design coupled with response surface methodology (RSM) was applied. Table 1 shows the variables and levels used in the experimental design.

2.2.2 | Ascorbic acid content determination

The AA content in the solution was determined according to AOAC's 967.21 method (AOAC International, 2010), known as the 2,6-dichlorophenolindophenol titration method, with the modifications described by Benassi and Antunes (1988). Approximately 50 mg

TABLE 1 Variables and levels used in experimental design

Sample	Wall concentration g% (w/v) ^a	Core (%) ^b	Wall : Core (w/w)
S1	1.0 (−1) ^c	25 (−1)	1:0.25
S2	1.0 (−1)	50 (0)	1:0.5
S3	1.0 (−1)	75 (+1)	1:0.75
S4	2.0 (0)	25 (−1)	1:0.25
S5	2.0 (0)	50 (0)	1:0.5
S6	2.0 (0)	75 (+1)	1:0.75
S7	3.0 (+1)	25 (−1)	1:0.25
S8	3.0 (+1)	50 (0)	1:0.5
S9	3.0 (+1)	75 (+1)	1:0.75

^a The mass ratio of gelatin:pectin was fixed at 1:1.

^b Percentage of simple emulsion (core) in relation to the total amount of polymers (w/w).

^c The numbers in parentheses represent the coded variables of the experimental design.

of 2,6-dichlorophenolindophenol were dissolved in 100 ml of hot water (40–50 °C) containing 42 mg of NaHCO₃, and the volume of the solution was completed to 200 ml. The treatments were dissolved with oxalic acid solution at 0.03% and titrated with 2,6-dichlorophenolindophenol solution. The 2,6-dichlorophenolindophenol solution was standardized with a standard solution of L-ascorbic acid. The concentration of ascorbic acid in the samples was calculated according to the amount of solution used in the titration compared with the standard solution of 2,6-dichlorophenolindophenol.

2.2.3 | Characterization of the microcapsules

Morphological characteristics

The morphological characteristics of the freeze-dried microcapsules were examined by a scanning electron microscope (SEM) (Vega3 LMU, Tescan Orsay Holding, Brno, Czech Republic), operating in the secondary electron mode with a 15 kV acceleration voltage. The microscope has a resolution of 3 nm and controlled pressure between 3 and 500 Pa. For the SEM observations, freeze-dried microcapsules were fixed in metallic adhesive tapes adhered in metallic stubs. The stubs were covered with a thin layer of gold evaporator (Balzer SCD50—Lichtenstein, Austria) for 180 s under a 40 mA current.

Mean diameter

The mean diameter of the freeze-dried microcapsules was determined by the AxioVision LE-Observer D1 (Carl Zeiss® Group, Aalen, Jena, Germany) inverted optical microscope coupled to an Axiocam MR3 camera and connected to a computer. The images obtained by the Axio Vision SE64 software (Carl Zeiss® Microscopy, Tornwood, New York, NY) were used to calculate the mean diameter of the particles with the aid of the free software ImageJ. The diameter was calculated based on 150 microparticles from each different formulation.

Water activity (A_w) and moisture content

Water activity was determined by direct reading in an electronic meter (Aqualab, Pullman). The moisture content (expressed in wet basis) was determined by gravimetric analysis in an oven set at

105 °C until the samples reached a constant weight (AOAC International, 2010).

Solubility

The microcapsules' solubility was determined by the gravimetric method, according to Cano-Chauca, Stringheta, Ramos, and Cal-Vidal (2005), with some modifications. One gram of sample was added to 100 ml of distilled water at 40 °C and stirred for 5 min with a magnetic stirrer. The solution was filtered using filter paper (pore size 14 µm) and a 20 ml aliquot of the filtrate was transferred into a previously weighed Petri dish, which was then heated in an oven at 105 °C for 5 hr. The percentage solubilized was calculated by mass difference.

Hygroscopicity

About 0.2 g of the sample were distributed into Petri dishes and stored for 7 days in a dessicator containing a saturated solution of CaCl₂ (75% RH) at 25 °C. The hygroscopicity was expressed as grams of water absorbed per 100 g of sample (Cai & Corke, 2000).

Encapsulation efficiency

The amount of total AA was determined by the methodology described by Alvim et al. (2016), with some modifications. To determine the EE, 100 mg of the freeze-dried microcapsules were completely dissolved in approximately 80 ml of oxalic acid (0.03%) at 40 °C for 5 min. Then, the resulting solution was transferred to a 100 ml-volumetric flask. After preparation of the samples, the AA content in the solution was determined as described previously for the ascorbic acid content. The amount of surface AA was determined according to the methodology used by Ribeiro, Arellano, and Grosso (2012), with some adaptations. About 100 mg of the freeze-dried microcapsules and 10 ml of Tween 80 (0.1%) surfactant solution were added into 125 ml-erlenmeyers and shaken at 100 rpm for 5 min in an incubator provided with orbital shaking. The suspension was then filtered on filter paper and the amount of AA in the filtrate was determined as described in Section 2.2.2.

The EE was calculated by subtracting the amount of superficial ascorbic acid (AA_{superficial}) from the total ascorbic acid (AA_{total}) found in the sample, as shown in Equation (1):

$$EE (\%) = \frac{AA_{total} - AA_{superficial}}{AA_{total}} \times 100 \quad (1)$$

2.2.4 | Thermogravimetric analysis

The thermogravimetric curves (TG) were obtained using a TGA equipment (PerkinElmer, model 4,000, The United States) under a nitrogen atmosphere, with flow rate of 50 ml/min, heating rate of 10 °C/min, and temperature range from 30 to 800 °C.

2.2.5 | Fourier transform infrared spectroscopy

Spectroscopy analyses of the wall materials (gelatin and pectin), pure AA, and microcapsules related to sample S5 (the one that presented the best EE) were performed by means of Fourier transform infrared (FTIR) spectroscopy on a Vertex 70 equipment (Bruker, The United States) in the range of 4,000 to 600 cm^{−1}.

2.2.6 | Release profile

The release profile of the AA microcapsules was evaluated by an in vitro assay simulating gastric and intestinal conditions following the procedure described by Gbassi, Vandamme, Ennahar, and Marchioni (2009) and Wang, Ju, He, Yuan, and Aluko (2015), with some modifications. Approximately 30 mg of each microparticle was weighed in triplicate at eight different time intervals (0, 30, 60, 120, 180, 240, 300, and 360 min). The microcapsules were added to 20 ml of simulated gastric fluid (NaCl = 9 g/L and pepsin = 3 g/L, pH 1.8) and incubated at 37 °C under agitation during the first 120 min. One milliliter of each sample was withdrawn from the flask after 0, 30, 60, and 120 min and centrifuged at 4,000 rpm for 5 min. The supernatant was used to determine the amount of AA, according to Section 2.2.2. The samples were then added of 20 ml of simulated intestinal fluid (NaCl = 9 g/L, pancreatin = 10 g/L, and bile salts = 3 g/L, pH 6.8) and incubated at 37 °C under stirring. During this period, 1 ml of the mixture was withdrawn after 240, 300, and 360 min centrifuged at 4,000 rpm for 5 min, and the supernatant was used to determine the amount of AA as described previously. The amount of sample withdrawn for the analyses was replaced by the same amount of the respective solutions.

2.2.7 | Statistical analysis

The experimental determinations were submitted to analysis of variance (ANOVA). When the results showed significant differences among the means, these were compared by the Tukey's post-hoc test at the 5% level of significance ($p \leq .05$). A 3^2 full-factorial design coupled with response surface methodology (RSM) was applied to evaluate the effects of wall material concentration and core ratio on EE.

3 | RESULTS AND DISCUSSION

3.1 | Physicochemical properties of AA microcapsules

Table 2 presents the results of mean diameter, moisture, water activity, solubility, hygroscopicity, and EE of the freeze-dried AA microcapsules. The mean diameter ranged from 6.1 to 9.3 μm , and is directly related to the core: wall material ratio, as the proportion of core

increased with an increase in the microparticles' mean diameter. Samples S7 (6.1 μm) and S9 (9.3 μm) had the smallest and the largest mean diameters, respectively. De Souza et al. (2018) obtained a mean diameter of 116 μm in the encapsulation of cinnamon extract by complex coacervation using the same wall materials as in this study. Many factors may interfere with particle diameter, such as the microencapsulation method, the concentration of the wall material, and the process conditions, such as speed and homogenization time, which in these studies were very different from the ones used in the present work.

Saravanan and Rao (2010) encapsulated indomethacin by complex coacervation using gelatin/pectin and obtained mean diameters of 120.9 and 110.6 μm with moisture content ranging from 5.1 to 13.7%. Samples S4 (13.7%) and S5 (13.1%) exhibited the highest moisture content, while S2 (6.1%), S3 (6.2%), and S6 (6.3%) had the same moisture content ($p > .05$). The same behavior was observed for samples S1 (7.9%) and S9 (7.3%). The lowest moisture content was found in samples S7 and S8 (5.5 and 5.1%, respectively), which had the lowest A_w , as expected in lyophilized products (Damodaran et al., 2010).

The solubility of freeze-dried microcapsules ranged from 8.2 to 35.6%. The samples S7 (18.8), S8 (26.8%), and S9 (35.6%) presented higher solubility in water. As these samples were prepared with a wall polymers' concentration of 3.0%, the increased solubility may be attributed to this increase in concentration. Sample S8 exhibited low EE (34.8%), which can explain the high solubility since the material that was not encapsulated remained on the surface of the microcapsule and solubilized in water more quickly. Rocha-Selmi, Bozza, Thomazini, Bolini, and Fávaro-Trindade (2013) observed the same behavior (i.e., high solubility and low EE) in some samples when they encapsulated aspartame by complex coacervation. The remaining samples presented relatively low solubility, which is expected in the case of microcapsules produced by coacervation.

Hygroscopicity ranged from 12.4 to 16.5%. Sample S9 (16.5%) presented the highest hygroscopicity, as well as the highest solubility and the highest mean diameter. The behavior for the hygroscopicity was similar to that of solubility, that is, samples with the highest concentration of wall material had higher hygroscopicity. Sample S5, in addition to having the highest EE (94.3%), showed the lowest hygroscopicity, 12.4%, implying that hygroscopicity as well as solubility depends on EE. In the studies carried out by Mendanha et al. (2009), Nori et al. (2011), and Silva, Favaro-Trindade, Rocha, and Thomazini (2012), higher hygroscopicity values were found in the encapsulation

TABLE 2 Physicochemical properties of AA microcapsules produced by complex coacervation

Sample	Mean diameter (μm)	Moisture (%)	A_w	Solubility (%)	Hygroscopicity (g/100 g)	EE (%)
S1	6.7 ^{cd} $\pm$ 2.33	7.9 ^b $\pm$ 0.91	0.25 ^c $\pm$ 0.008	11.4 ^c $\pm$ 1.05	13.5 ^{bc} $\pm$ 0.25	23.7 ^f $\pm$ 3.32
S2	7.2 ^{bcd} $\pm$ 3.03	6.1 ^{cd} $\pm$ 0.63	0.23 ^{cd} $\pm$ 0.005	8.2 ^c $\pm$ 0.18	14.9 ^{abc} $\pm$ 0.20	77.0 ^c $\pm$ 2.72
S3	7.8 ^{bc} $\pm$ 2.35	6.2 ^{cd} $\pm$ 0.47	0.23 ^{cd} $\pm$ 0.005	12.9 ^c $\pm$ 0.68	13.9 ^{abc} $\pm$ 1.15	35.4 ^e $\pm$ 3.01
S4	6.5 ^{cd} $\pm$ 3.40	13.7 ^a $\pm$ 1.11	0.54 ^a $\pm$ 0.012	13.5 ^c $\pm$ 0.42	13.8 ^{abc} $\pm$ 0.22	55.3 ^d $\pm$ 2.17
S5	7.6 ^{bc} $\pm$ 4.50	13.1 ^a $\pm$ 1.41	0.37 ^b $\pm$ 0.04	11.5 ^c $\pm$ 0.35	12.4 ^c $\pm$ 0.40	94.3 ^a $\pm$ 1.04
S6	8.3 ^{ab} $\pm$ 3.29	6.3 ^{cd} $\pm$ 0.29	0.22 ^d $\pm$ 0.015	14.2 ^c $\pm$ 0.31	14.8 ^{abc} $\pm$ 0.79	81.4 ^b $\pm$ 1.29
S7	6.1 ^d $\pm$ 3.54	5.5 ^d $\pm$ 0.31	0.18 ^e $\pm$ 0.010	18.8 ^{bc} $\pm$ 1.73	15.5 ^{ab} $\pm$ 0.23	51.0 ^d $\pm$ 3.15
S8	6.6 ^{cd} $\pm$ 2.46	5.1 ^d $\pm$ 0.44	0.14 ^f $\pm$ 0.06	26.8 ^{ab} $\pm$ 5.62	15.1 ^{abc} $\pm$ 1.49	34.8 ^e $\pm$ 3.21
S9	9.3 ^a $\pm$ 4.06	7.3 ^{bc} $\pm$ 0.50	0.25 ^c $\pm$ 0.08	35.6 ^a $\pm$ 6.34	16.5 ^a $\pm$ 0.52	51.6 ^d $\pm$ 2.83

There are no significant differences between samples assigned with the same letters in the same column ($p < .05$).

of hydrolyzed casein (20.08–26.0 g/100 g), propolis extract (34.46–34.10 g/100 g), and lycopene (33.4–36.8 g/100 g).

EE is an important parameter in the evaluation of an encapsulation process, especially when the substance to be encapsulated is hydrophilic. Generally, hydrophilic substances (such as AA) cannot be encapsulated directly by complex coacervation as they require oil-in-water type simple emulsion, which is considered as core in the encapsulation process. This additional step in the coacervation process was carried out in this study and in the works developed by Comunian et al. (2013) in the encapsulation of AA, by Rocha-Selmi, Theodoro, et al. (2013) in the encapsulation of sucralose, by Rocha-Selmi, Bozza, et al. (2013) in the encapsulation of aspartame, and by Santos, Bozza, Thomazini, and Favaro-Trindade (2015) in the encapsulation of xylitol.

Table 2 presents the results obtained for the EE of AA. The EE ranged from 23.7 to 94.3%, with significant differences between samples S1, S2, and S3, while S1 had the lowest EE (23.7%) ($p < .05$). A low EE confirms the hypothesis that the concentration of the wall material (pectin + gelatin) was not high enough to allow the encapsulation of the entire core, resulting in a high concentration of free nucleus. Samples S3 (35.4%) and S8 (34.8%) exhibited the same EE ($p > .05$), even when prepared with different wall material concentrations and core : wall ratios. The same behavior was observed in samples S4 (55.3%), S7 (51.0%), and S9 (51.6%), indicating that both the wall material concentration and the core: wall ratio influenced the EE. The highest EE was determined in sample S5 (94.3%), followed by sample S6 (81.4%), both produced with 2.0% of wall material. Most studies involving encapsulation of bioactive compounds resulted in high EE values with an increase in the concentration of the polymers used as wall material. In this study an increase in EE was achieved when the concentration of the wall material increased from 1.0 to 2.0%, however this behavior was not observed with a higher concentration (3.0%). This indicates that wall material concentrations above 2.0% did not yield a greater interaction between the wall polymers.

FIGURE 1 Response surface for the effects of wall material concentration and core ratio on encapsulation efficiency

Figure 1 presents the response surface for the effects of wall material concentration and core ratio on EE. This graph confirms the conclusions discussed above in that the best condition for AA encapsulation corresponds to 2.0% wall material and 50% core (sample S5). As it exhibited the best EE, sample S5 was assessed for morphology, thermal behavior, FTIR, and release in gastric and intestinal fluids.

Nori et al. (2011) encapsulated propolis extract by complex coacervation using isolated soy protein and pectin as encapsulating agents. The EE decreased from 72.01% to 66.12% when the wall material concentration was increased from 2.5 g/100 ml to 5.0 g/100 ml. The authors concluded that greater concentrations of wall material in relation to the core did not give higher EEs, as observed in this study. De Souza et al. (2018) encapsulated cinnamon extract by complex coacervation using gelatin and pectin as wall materials and obtained an EE of 85%. García-Saldaña et al. (2016) obtained about 18% of EE using gelatin and pectin to encapsulate sulforaphane from broccoli seeds by complex coacervation, which is lower than the efficiency obtained in this work using the same wall materials.

3.2 | Morphology

The morphology of the AA microcapsules from the S5 sample is presented in Figure 2. The optical microscopy of the wet coacervated particles (Figure 2a) and the freeze-dried particles is shown in Figure 2b. The images show spherical, reservoir-type microcapsules filled with small droplets of the AA solution inside the oil droplet, both immersed in the gelatin solution, confirming the formation of the double emulsion.

Figure 2c,d show the SEM image of the freeze-dried microcapsules. Figure 2c reveals an agglomerated structure, which is very common in particles that have undergone lyophilization. This same behavior was observed by Comunian et al. (2016), Rocha-Selmi, Theodoro, et al. (2013), and Butstraen and Salaün (2014). Figure 2d shows microcapsules with continuous walls and without cracks and pores. Porous microcapsules are undesirable because they can facilitate the entry of oxygen and the diffusion of water into the microcapsule, consequently resulting in rapid release of AA to the medium. These results confirm that the drying process chosen was adequate as no damage was found along the microcapsules' surface. An undamaged surface is important to ensure greater protection and retention of the encapsulated material.

3.3 | Thermogravimetric analysis

Thermal analysis was performed to evaluate the thermal behavior of the AA microcapsules. As the samples showed similar behavior, only the results for the sample with highest EE, S5 (94.3%), will be shown hereinafter. Figure 3 shows the thermograms (TG) of the microcapsules of sample S5, gelatin, pectin, and pure AA and their respective derivatives. The gelatine thermogram exhibited two events of thermal degradation, an event between 31 and 139 °C, related to approximately 9% of weight loss as result of water removal from the sample. The second thermal event occurred at 271.8 to 391.3 °C, representing 56% of weight loss as a result of protein degradation.

FIGURE 2 Micrographs of the coacervated microcapsules: (a) sample S5 by optic microscopy (1,600 $\times$); (b) freeze-dried sample S5 by optic microscopy (1,600 $\times$); (c) freeze-dried sample S5 by scanning electron microscopy (60 $\times$); (d) freeze-dried sample S5 by scanning electron microscopy (5,000 $\times$)

FIGURE 3 Thermal behavior of sample S5, gelatin, pectin, and pure AA and their respective derivatives

Pectin showed three thermal events. The first event occurred from 32 to 70 $^{\circ}\text{C}$ and is related to a 5.7% of weight loss ascribed to removal of water from the sample. The second thermal event took place at 190–208 $^{\circ}\text{C}$ and is attributed to the loss of acidic side chain groups and pyranoside ring carbons. The third and most pronounced thermal event occurred at a temperature range of 222–256 $^{\circ}\text{C}$, where 25% of the weight was lost due to the pyrolytic decomposition of the sample. This decomposition consists in a primary and secondary decarboxylation involving the acid side group and a carbon in the ring of the pectin molecule (Monfregola, Bugatti, Amodeo, De Luca, & Vittoria, 2011).

The pure AA showed two thermal events, the first one at 193–250 $^{\circ}\text{C}$, related to 31.8% of weight loss, and the second one at

275–311 $^{\circ}\text{C}$, corresponding to a weight loss of 12.4%. The main event related to the thermal decomposition of AA arises in the temperature range of 191–268 $^{\circ}\text{C}$, where the simultaneous loss of one molecule of water and one molecule of CO_2 occurs (Jingyan, Yuwen, Zhiyong, & Cunxin, 2013). The thermal degradation of the sample S5 developed in three parts: the first event at 34.5–79.9 $^{\circ}\text{C}$ (5% weight loss), the second one at 216.8–250.6 $^{\circ}\text{C}$ (7.6% of weight loss), and the third one at 296.4–353.4 $^{\circ}\text{C}$, which gave the highest weight loss (~24%). In this temperature range weight loss is usually associated with the release of the nucleus together with the decomposition of the polysaccharide (pectin) and the protein (gelatin) used as wall materials (Lv, Zhang, Zhang, Abbas, & Karangwa, 2013). The sample S5 presented thermal events similar to those of AA (core) and of gelatin and pectin

(wall materials). The encapsulation process increased the stability of the pure AA, as shown in Figure 3.

3.4 | FTIR analysis

FTIR analysis is able to detect interactions that occur during the process of complex coacervation between gelatin and pectin. Figure 4 shows the spectra obtained for the sample S5, for the wall materials (gelatin and pectin), and for pure AA. The gelatin spectrum can be seen in Figure 4d.

A peak around $3,313\text{ cm}^{-1}$ is observed in the gelatin's spectrum, which is characteristic of amine groups, which are positively charged. Other peaks also can be observed in the gelatin molecule in the region of $1,550$ to $1,650\text{ cm}^{-1}$. The peak at $1,631\text{ cm}^{-1}$ in the gelatin spectrum refers to amide I and is due to the stretching at C[dbond]O and the deformation of NH. According to Anvari and Chung (2016), this wavelength may be linked to the secondary structure of proteins (β -sheets), favoring the interaction between biopolymers in the complex formation process.

The spectrum of pectin (Figure 4b) shows a region characteristic to carboxylic groups ($1,750$ – $1,350\text{ cm}^{-1}$), with the highest intensity for the esterified band at $1,750\text{ cm}^{-1}$ and for the deprotonated COO group ($1,600$ – $1,650\text{ cm}^{-1}$), which may be related to the pectin's degree of esterification (DE), an important parameter for these compounds (Fellah, Anjukandi, Waterland, & Williams, 2009; Saravanan &

Rao, 2010). The DE confirms that the pectin used in this study is of low degree of methoxylation (García-Saldaña et al., 2016). The pure AA's spectrum is shown in Figure 4a. The spectrum showed peaks at $1,140$, $1,310$, and $1,699\text{ cm}^{-1}$, which according to Desai and Park (2005) can be attributed to the C[dbond]O and OH groups present in the AA molecule and also in the S5 sample, confirming the presence of the active compound in the microcapsules.

3.5 | Release profile in simulated gastric and intestinal fluids

The release of encapsulated compounds involves several steps, such as absorption of solvent by the microcapsule, dissolution of the wall, dissolution of the filling, permeation of the filling by the wall, and diffusion of the filling by the solvent (Pereira et al., 2009). The coating used in the microencapsulation process is of great importance since it is responsible for the controlled release of the core. The release mechanism may be modulated by the molecular weight of the polymer used: the higher its molecular weight, the slower the *in vitro* release of the encapsulated compounds (Kumari, Yadav, & Yadav, 2010).

Figure 5 and Table 3 shows the *in vitro* release profile of AA microcapsules with different concentrations of wall material and core. In general, all samples had a similar behavior, with a faster release in the first 120 min of analysis, which corresponds to the gastric fluid, and a slower release in the intestinal fluid, from 120 min onward. In the first 30 min of analysis, (gastric fluid), samples S3 (64.9%) and S8 (63.0%) showed a higher release rate when compared with the others. Although these samples were prepared with different concentrations of wall material and with different core: wall proportions—sample S3 (1.0% wall material and 75% core) and sample S8 (3.0% wall material and 50% core)—they exhibited virtually the same EE ($p > .05$). These samples also presented low EE, S3 (35.4%) and S8 (34.8%), resulting in more fragile microcapsules with a larger amount of active material adhered to their surface, hence the faster release. Samples S5 (2.0% wall material and 50% core) and S6 (2.0% wall material and 75% core) showed a slower release during the first 120 min (gastric fluid), which

FIGURE 4 Spectra obtained by FTIR for (a) pure AA, (b) pectin, (c) sample S5, and (d) gelatin

FIGURE 5 Release profile of AA microcapsules in simulated gastric and intestinal fluids

TABLE 3 Release profile of AA microcapsules in simulated gastric and intestinal fluids

Sample	Time (min)						
	0	30	60	120	240	300	360
S1	35.6 ^{ABf} ± 1.67	56.1 ^{Ce} ± 2.17	70.1 ^{CDd} ± 2.19	90.7 ^{Ac} ± 2.34	92.5 ^{Abc} ± 0.80	97.0 ^{Aab} ± 1.30	97.8 ^{Aa} ± 1.50
S2	30.2 ^{De} ± 0.82	34.8 ^{Ee} ± 1.99	73.8 ^{ABcd} ± 0.86	81.8 ^{Cc} ± 1.61	87.1 ^{CDEb} ± 0.92	93.6 ^{ABa} ± 3.63	95.2 ^{ABCa} ± 0.96
S3	32.1 ^{BCDf} ± 1.63	64.9 ^{Ae} ± 1.59	75.6 ^{ABd} ± 1.16	88.1 ^{ABc} ± 1.05	89.8 ^{ABCDbc} ± 0.78	92.5 ^{ABb} ± 0.80	96.8 ^{ABa} ± 1.67
S4	37.5 ^{Ae} ± 2.35	56.7 ^{Cd} ± 1.83	71.9 ^{BCDc} ± 1.09	85.4 ^{BCb} ± 0.73	86.3 ^{DEb} ± 1.65	88.9 ^{Bab} ± 0.77	93.4 ^{BCa} ± 2.30
S5	25.8 ^{Eg} ± 1.54	36.0 ^{Ef} ± 1.15	57.9 ^{Ee} ± 1.21	68.6 ^{Dd} ± 2.51	85.0 ^{Ec} ± 2.36	90.8 ^{Bb} ± 2.31	97.7 ^{Aa} ± 1.48
S6	36.5 ^{Ad} ± 1.07	38.3 ^{Ed} ± 1.46	59.7 ^{Ec} ± 1.61	69.4 ^{Db} ± 1.27	89.7 ^{BCDa} ± 0.79	91.7 ^{Ba} ± 2.03	92.5 ^{Ca} ± 0.80
S7	34.6 ^{ABCe} ± 0.73	59.5 ^{BCd} ± 1.56	73.7 ^{ABCc} ± 1.65	88.1 ^{ABc} ± 1.10	88.9 ^{BCDb} ± 0.77	90.8 ^{Bb} ± 1.45	95.1 ^{ABCa} ± 0.96
S8	30.3 ^{CDe} ± 1.91	63.0 ^{ABd} ± 1.30	77.3 ^{Ac} ± 0.40	90.8 ^{Ab} ± 1.45	91.6 ^{ABb} ± 0.83	93.3 ^{ABb} ± 0.90	97.9 ^{Aa} ± 1.21
S9	38.3 ^{Ad} ± 1.45	48.9 ^{Dc} ± 1.81	68.4 ^{Db} ± 0.83	88.1 ^{ABa} ± 1.09	90.5 ^{ABCa} ± 1.27	90.7 ^{Ba} ± 0.45	91.6 ^{Ca} ± 0.84

Samples assigned with the same capital letters in the same column and with the same small letters in the same row do not differ statistically ($p < .05$).

may be related to their high EE (94.3% for S5 and 84.1% for S6). High EEs indicate that the amount of wall material was sufficient to cover the entire core, leaving little active material unencapsulated and consequently slowing the release rate of the core.

When intestinal fluid was added to gastric fluid (after 120 min of the beginning of the release analysis), there was a reduction in the release rate, and the remaining AA in the microcapsules was gradually released up to 360 min (Figure 5). In the human body, it is expected that the contents of the microcapsules are released in small amounts under gastric conditions, while the release is gradual and complete in intestinal conditions, since it is in the intestine that most nutrients are absorbed (Somchue, Sermsri, Shiwatana, & Siripinyanond, 2009). Sample S5 exhibited the best release profile: only 68.8% of the microcapsules content was released in the gastric fluid, while an almost complete release (97.7%) took place in the intestinal fluid, in 360 min.

Rutz, Borges, Zambiasi, da Rosa, and da Silva (2016) evaluated the carotenoid release profile of microcapsules produced with chitosan/sodium tripolyphosphate and chitosan/carboxymethylcellulose as wall materials. The particles produced with chitosan/carboxymethylcellulose showed faster release in the gastric fluid (about 81%) and slower release in the intestinal fluid. Rutz et al. (2017) studied the release of carotenoids from palm oil encapsulated in chitosan, xanthan gum and pectin by complex coacervation followed by freeze-drying and spray-drying. The authors achieved a release of about 80% of the carotenoids in 120 min for lyophilized microcapsules produced with chitosan/pectin, a result similar to those found in this study. In another research reported by Luo, Zhang, Whent, Yu, and Wang (2011), the release of tocopherol encapsulated in zein/chitosan was evaluated in different concentrations (10, 20, and 30%). The microcapsules with the highest concentration of tocopherol (30%) exhibited slower release when compared with the other concentrations. About 50% of tocopherol was released under gastric conditions and 30% under intestinal conditions, that is, the release was faster in gastric conditions and slower in tryptic conditions. The same behavior was observed in this study, a faster release in the gastric fluid (pH 1.8) compared with the intestinal fluid (pH 6.8).

The mechanism of release of AA from the microcapsules produced in this study may be related to the dissolution of the wall or even the degradation of the particle, since in addition to the pH variation, the presence of simulated gastric and intestinal fluid proteases in

the medium may have contributed to the dissolution of the gelatin present in the microcapsules' wall. According to Comunian and Favaro-Trindade (2016), microparticles produced by complex coacervation can be easily degraded as a result of pH variation which alters the solubility of the polymers used as wall materials, leading to the release of the nucleus.

The release behavior of the microcapsules can be easily modulated by adapting the process conditions to the target food application (i.e., antioxidant, source of vitamin C). Manufacturing conditions the food will undergo following addition of the microcapsules (pH, temperature) must be taking into account in this process. Further studies will be conducted by these authors to test the best microcapsule formulation and encapsulation conditions yielding the best release profile of the encapsulated AA in specific food matrices.

4 | CONCLUSION

In this work, microcapsules having a hydrophilic core (AA) were produced with the addition of an emulsification step of the active agent in oil prior to the complex coacervation process. Samples S2, S5, and S6 presented high EE values. The microcapsules also exhibited low solubility and hygroscopicity. The encapsulated AA was released faster in gastric fluid than in intestinal fluid. Sample S5 had the best in vitro release profile, with a release of only 68% of the contents of the microcapsules in 120 min in the presence of gastric fluid. These results indicate that the addition of the emulsification step prior to the complex coacervation together with the selected wall materials presented great potential to encapsulate AA, contributing to an enhanced stability and thus facilitating its incorporation in food matrices.

ACKNOWLEDGMENTS

The authors would like to thank CAPES Brazil—Coordination for the Improvement of Higher Education Personnel—for the financial support.

ORCID

Michele Cristina Rodrigues da Cruz <https://orcid.org/0000-0002-7366-2270>

REFERENCES

- Abbas, S., Da Wei, C., Hayat, K., & Xiaoming, Z. (2012). Ascorbic acid: Microencapsulation techniques and trends—A review. *Food Reviews International*, 28(4), 343–374. <https://doi.org/10.1080/87559129.2011.635390>
- Alvim, I. D., Stein, M. A., Koury, I. P., Dantas, F. B. H., & Cruz, C. L. d. C. V. (2016). Comparison between the spray drying and spray chilling micro-particles contain ascorbic acid in a baked product application. *LWT—Food Science and Technology*, 65, 689–694. <https://doi.org/10.1016/j.lwt.2015.08.049>
- Anvari, M., & Chung, D. (2016). Dynamic rheological and structural characterization of fish gelatin–Gum arabic coacervate gels cross-linked by tannic acid. *Food Hydrocolloids*, 60, 516–524. <https://doi.org/10.1016/j.foodhyd.2016.04.028>
- AOAC International. (2010). *Official methods of analysis of AOAC International (Official Method 967.21)* (18th ed.). Gaithersburg, MD: AOAC.
- Benassi, M. T., & Antunes, A. J. (1988). A comparison of metaphosphoric and oxalic acids as extractant solutions for the determination of vitamin C in selected vegetables. *Arquivos de Biologia e Tecnologia*, 31(4), 507–513.
- Butstraen, C., & Salaün, F. (2014). Preparation of microcapsules by complex coacervation of gum Arabic and chitosan. *Carbohydrate Polymers*, 99, 608–616. <https://doi.org/10.1016/j.carbpol.2013.09.006>
- Cai, Y. Z., & Corke, H. (2000). Production and properties of spray-dried *Amaranthus betacyanin* pigments. *Journal of Food Science*, 65(7), 1248–1252. <https://doi.org/10.1111/j.1365-2621.2000.tb10273.x>
- Cano-Chauca, M., Stringheta, P. C., Ramos, A. M., & Cal-Vidal, J. (2005). Effect of the carriers on the microstructure of mango powder obtained by spray drying and its functional characterization. *Innovative Food Science and Emerging Technologies*, 6(4), 420–428. <https://doi.org/10.1016/j.ifset.2005.05.003>
- Comunian, T. A., Abbaspourrad, A., Favaro-Trindade, C. S., & Weitz, D. A. (2014). Fabrication of solid lipid microcapsules containing ascorbic acid using a microfluidic technique. *Food Chemistry*, 152, 271–275. <https://doi.org/10.1016/j.foodchem.2013.11.149>
- Comunian, T. A., Boillon, M. R. G., Thomazini, M., Nogueira, M. S., de Castro, I. A., & Favaro-Trindade, C. S. (2016). Protection of echium oil by microencapsulation with phenolic compounds. *Food Research International*, 88, 114–121. <https://doi.org/10.1016/j.foodres.2016.03.008>
- Comunian, T. A., & Favaro-Trindade, C. S. (2016). Microencapsulation using biopolymers as an alternative to produce food enhanced with phytosterols and omega-3 fatty acids: A review. *Food Hydrocolloids*, 61, 442–457. <https://doi.org/10.1016/j.foodhyd.2016.06.003>
- Comunian, T. A., Ravanfar, R., de Castro, I. A., Dando, R., Favaro-Trindade, C. S., & Abbaspourrad, A. (2017). Improving oxidative stability of echium oil emulsions fabricated by microfluidics: Effect of ionic gelation and phenolic compounds. *Food Chemistry*, 233, 125–134. <https://doi.org/10.1016/j.foodchem.2017.04.085>
- Comunian, T. A., Thomazini, M., Alves, A. J. G., de Matos Junior, F. E., de Carvalho Balieiro, J. C., & Favaro-Trindade, C. S. (2013). Microencapsulation of ascorbic acid by complex coacervation: Protection and controlled release. *Food Research International*, 52(1), 373–379. <https://doi.org/10.1016/j.foodres.2013.03.028>
- Damodaran, S., Parkin, K. L., & Fennema, O. R. (2010). *Fennema's food chemistry (Arthemed, Ed.)* (4th ed.). Porto Alegre: CRC Press.
- De Kruijff, C. G., Weinbreck, F., & De Vries, R. (2004). Complex coacervation of proteins and anionic polysaccharides. *Current Opinion in Colloid and Interface Science*, 9(5), 340–349. <https://doi.org/10.1016/j.cocis.2004.09.006>
- De Matos, F. E., Comunian, T. A., Thomazini, M., & Favaro-Trindade, C. S. (2017). Effect of feed preparation on the properties and stability of ascorbic acid microparticles produced by spray chilling. *LWT—Food Science and Technology*, 75, 251–260. <https://doi.org/10.1016/j.lwt.2016.09.006>
- De Souza, V. B., Thomazini, M., Echalar Barrientos, M. A., Nalin, C. M., Ferro-Furtado, R., Genovese, M. I., & Favaro-Trindade, C. S. (2018). Functional properties and encapsulation of a proanthocyanidin-rich cinnamon extract (*Cinnamomum zeylanicum*) by complex coacervation using gelatin and different polysaccharides. *Food Hydrocolloids*, 77, 297–306. <https://doi.org/10.1016/j.foodhyd.2017.09.040>
- Desai, K. G. H., & Park, H. J. (2005). Encapsulation of vitamin C in tripolyphosphate cross-linked chitosan microspheres by spray drying. *Journal of Microencapsulation*, 22(2), 179–192. <https://doi.org/10.1080/02652040400026533>
- Desai, K. G., Liu, C., & Park, H. J. (2006). Characteristics of vitamin C encapsulated tripolyphosphate-chitosan microspheres as affected by chitosan molecular weight. *Journal of Microencapsulation*, 23(1), 79–90. <https://doi.org/10.1080/02652040500435360>
- Dias, M. I., Ferreira, I. C. F. R., & Barreiro, M. F. (2015). Microencapsulation of bioactives for food applications. *Food & Function*, 6(4), 1035–1052. <https://doi.org/10.1039/C4FO01175A>
- Dong, Z., Ma, Y., Hayat, K., Jia, C., Xia, S., & Zhang, X. (2011). Morphology and release profile of microcapsules encapsulating peppermint oil by complex coacervation. *Journal of Food Engineering*, 104(3), 455–460. <https://doi.org/10.1016/j.jfoodeng.2011.01.011>
- Eghbal, N., & Choudhary, R. (2018). Complex coacervation: Encapsulation and controlled release of active agents in food systems. *LWT—Food Science and Technology*, 90, 254–264. <https://doi.org/10.1016/j.lwt.2017.12.036>
- Fellah, A., Anjukandi, P., Waterland, M. R., & Williams, M. A. K. (2009). Determining the degree of methylesterification of pectin by ATR/FT-IR: Methodology optimisation and comparison with theoretical calculations. *Carbohydrate Polymers*, 78(4), 847–853. <https://doi.org/10.1016/j.carbpol.2009.07.003>
- García-Saldaña, J. S., Campas-Baypoli, O. N., López-Cervantes, J., Sánchez-Machado, D. I., Cantú-Soto, E. U., & Rodríguez-Ramírez, R. (2016). Microencapsulation of sulforaphane from broccoli seed extracts by gelatin/gum Arabic and gelatin/pectin complexes. *Food Chemistry*, 201, 94–100. <https://doi.org/10.1016/j.foodchem.2016.01.087>
- Gbassi, G. K., Vandamme, T., Ennahar, S., & Marchioni, E. (2009). Microencapsulation of *Lactobacillus plantarum* spp in an alginate matrix coated with whey proteins. *International Journal of Food Microbiology*, 129(1), 103–105. <https://doi.org/10.1016/j.ijfoodmicro.2008.11.012>
- Gharsallaoui, A., Roudaut, G., Chambin, O., Voille, A., & Saurel, R. (2007). Applications of spray-drying in microencapsulation of food ingredients: An overview. *Food Research International*, 40(9), 1107–1121. <https://doi.org/10.1016/j.foodres.2007.07.004>
- Gibbs, B. F., Kermasha, S., Alli, I., & Mulligan, C. N. (1999). Encapsulation in the food industry: A review. *International Journal of Food Sciences and Nutrition*, 50, 213–224.
- Gouin, S. (2004). Microencapsulation: Industrial appraisal of existing technologies and trends. *Trends in Food Science and Technology*, 15(7–8), 330–347. <https://doi.org/10.1016/j.tifs.2003.10.005>
- Jingyan, S., Yuwen, L., Zhiyong, W., & Cunxin, W. (2013). Investigation of thermal decomposition of ascorbic acid by TG-FTIR and thermal kinetics analysis. *Journal of Pharmaceutical and Biomedical Analysis*, 77, 116–119. <https://doi.org/10.1016/j.jpba.2013.01.018>
- Kumari, A., Yadav, S. K. S. C., & Yadav, S. K. S. C. (2010). Biodegradable polymeric nanoparticles based drug delivery systems. *Colloids and Surfaces B: Biointerfaces*, 75(1), 1–18. <https://doi.org/10.1016/j.colsurfb.2009.09.001>
- Luo, Y., Zhang, B., Whent, M., Yu, L. L., & Wang, Q. (2011). Preparation and characterization of zein/chitosan complex for encapsulation of α -tocopherol, and its in vitro controlled release study. *Colloids and Surfaces B: Biointerfaces*, 85(2), 145–152. <https://doi.org/10.1016/j.colsurfb.2011.02.020>
- Lv, Y., Zhang, X., Zhang, H., Abbas, S., & Karangwa, E. (2013). The study of pH-dependent complexation between gelatin and gum arabic by morphology evolution and conformational transition. *Food Hydrocolloids*, 30(1), 323–332. <https://doi.org/10.1016/j.foodhyd.2012.06.007>
- Maderuelo, C., Zarzuelo, A., & Lanao, J. M. (2011). Critical factors in the release of drugs from sustained release hydrophilic matrices. *Journal of Controlled Release*, 154(1), 2–19. <https://doi.org/10.1016/j.jconrel.2011.04.002>
- Matos, F. E., Di Sabatino, M., Passerini, N., Favaro-Trindade, C. S., & Albertini, B. (2015). Development and characterization of solid lipid microparticles loaded with ascorbic acid and produced by spray congealing. *Food Research International*, 67, 52–59. <https://doi.org/10.1016/j.foodres.2014.11.002>
- Mendanha, D. V., Molina Ortiz, S. E., Favaro-Trindade, C. S., Mauri, A., Monterrey-Quintero, E. S., & Thomazini, M. (2009). Microencapsulation

- of casein hydrolysate by complex coacervation with SPI/pectin. *Food Research International*, 42(8), 1099–1104. <https://doi.org/10.1016/j.foodres.2009.05.007>
- Monfregola, L., Bugatti, V., Amodeo, P., De Luca, S., & Vittoria, V. (2011). Physical and water sorption properties of chemically modified pectin with an environmentally friendly process. *Biomacromolecules*, 12(6), 2311–2318. <https://doi.org/10.1021/bm200376c>
- Moschakis, T., & Biliaderis, C. G. (2017). Biopolymer-based coacervates: Structures, functionality and applications in food products. *Current Opinion in Colloid and Interface Science*, 28, 96–109. <https://doi.org/10.1016/j.cocis.2017.03.006>
- Nori, M. P., Favaro-Trindade, C. S., Matias de Alencar, S., Thomazini, M., de Camargo Balieiro, J. C., & Contreras Castillo, C. J. (2011). Microencapsulation of propolis extract by complex coacervation. *LWT-Food Science and Technology*, 44(2), 429–435. <https://doi.org/10.1016/j.lwt.2010.09.010>
- Pereira, H. V. R., Saraiva, K. P., Carvalho, L. M. J., Andrade, L. R., Pedrosa, C., & Pierucci, A. P. T. R. (2009). Legumes seeds protein isolates in the production of ascorbic acid microparticles. *Food Research International*, 42(1), 115–121. <https://doi.org/10.1016/j.foodres.2008.10.008>
- Ribeiro, M. D. M., Arellano, D. B., & Grosso, C. R. F. (2012). The effect of adding oleic acid in the production of stearic acid lipid microparticles with a hydrophilic core by a spray-cooling process. *Food Research International*, 47(1), 38–44. <https://doi.org/10.1016/j.foodres.2012.01.007>
- Rocha-Selmi, G. A., Bozza, F. T., Thomazini, M., Bolini, H. M. A., & Favaro-Trindade, C. S. (2013). Microencapsulation of aspartame by double emulsion followed by complex coacervation to provide protection and prolong sweetness. *Food Chemistry*, 139, 72–78. <https://doi.org/10.1016/j.foodchem.2013.01.114>
- Rocha-Selmi, G. A., Theodoro, A. C., Thomazini, M., Bolini, H. M. A., & Favaro-Trindade, C. S. (2013). Double emulsion stage prior to complex coacervation process for microencapsulation of sweetener sucralose. *Journal of Food Engineering*, 119(1), 28–32. <https://doi.org/10.1016/j.jfoodeng.2013.05.002>
- Rodrigues da Cruz, M. C., Andreotti Dagostin, J. L., Perussello, C. A., & Masson, M. L. (2019). Assessment of physicochemical characteristics, thermal stability and release profile of ascorbic acid microcapsules obtained by complex coacervation. *Food Hydrocolloids*, 87, 71–82. <https://doi.org/10.1016/j.foodhyd.2018.07.043>
- Rutz, J. K., Borges, C. D., Zambiasi, R. C., Crizel-Cardozo, M. M., Kuck, L. S., & Noreña, C. P. Z. (2017). Microencapsulation of palm oil by complex coacervation for application in food systems. *Food Chemistry*, 220, 59–66. <https://doi.org/10.1016/j.foodchem.2016.09.194>
- Rutz, J. K., Borges, C. D., Zambiasi, R. C., da Rosa, C. G., & da Silva, M. M. (2016). Elaboration of microparticles of carotenoids from natural and synthetic sources for applications in food. *Food Chemistry*, 202, 324–333. <https://doi.org/10.1016/j.foodchem.2016.01.140>
- Santos, M. G., Bozza, F. T., Thomazini, M., & Favaro-Trindade, C. S. (2015). Microencapsulation of xylitol by double emulsion followed by complex coacervation. *Food Chemistry*, 171, 32–39. <https://doi.org/10.1016/j.foodchem.2014.08.093>
- Santos, M. G., Carpinteiro, D. A., Thomazini, M., Rocha-Selmi, G. A., da Cruz, A. G., Rodrigues, C. E. C., & Favaro-Trindade, C. S. (2014). Coencapsulation of xylitol and menthol by double emulsion followed by complex coacervation and microcapsule application in chewing gum. *Food Research International*, 66, 454–462. <https://doi.org/10.1016/j.foodres.2014.10.010>
- Saravanan, M., & Rao, K. P. (2010). Pectin-gelatin and alginate-gelatin complex coacervation for controlled drug delivery: Influence of anionic polysaccharides and drugs being encapsulated on physicochemical properties of microcapsules. *Carbohydrate Polymers*, 80(3), 808–816. <https://doi.org/10.1016/j.carbpol.2009.12.036>
- Shahidi, F., & Han, X. (1993). Encapsulation of food ingredients. *Critical Reviews in Food Science and Nutrition*, 33(6), 501–547. <https://doi.org/10.1080/10408399309527645>
- Silva, D. F., Favaro-Trindade, C. S., Rocha, G. A., & Thomazini, M. (2012). Microencapsulation of lycopene by gelatin-pectin complex coacervation. *Journal of Food Processing and Preservation*, 36(2), 185–190. <https://doi.org/10.1111/j.1745-4549.2011.00575.x>
- Sing, C. E. (2017). Development of the modern theory of polymeric complex coacervation. *Advances in Colloid and Interface Science*, 239, 2–16. <https://doi.org/10.1016/j.cis.2016.04.004>
- Somchue, W., Sermisri, W., Shiwatana, J., & Siripinyanond, A. (2009). Encapsulation of α -tocopherol in protein-based delivery particles. *Food Research International*, 42(8), 909–914. <https://doi.org/10.1016/j.foodres.2009.04.021>
- Uddin, M. S., Hawlader, M. N., & Zhu, H. J. (2001). Microencapsulation of ascorbic acid: Effect of process variables on product characteristics. *Journal of Microencapsulation*, 18(2), 199–209. <https://doi.org/10.1080/02652040010000352>
- Wang, Z., Ju, X., He, R., Yuan, J., & Aluko, R. E. (2015). Effect of high pressure treatment on rapeseed protein microparticle properties and gastrointestinal release behavior of the encapsulated peptides. *Food Research International*, 77, 549–555. <https://doi.org/10.1016/j.foodres.2015.09.023>

How to cite this article: da Cruz MCR, Perussello CA, Masson ML. Microencapsulated ascorbic acid: Development, characterization, and release profile in simulated gastrointestinal fluids. *J Food Process Eng*. 2018;41:e12922. <https://doi.org/10.1111/jfpe.12922>