Iranian Food Science and Technology Research Journal
Homepage: https://ifstrj.um.ac.ir

Research Article
Vol. 22, No. 1, March- April, 2026, p. 17-34

Encapsulation of Vitamins E and C by Whey Protein Concentrate– Chitosan via Spray Drying: Optimization, Physicochemical Characterization, and Release Rate Analysis

P. Mojtahedi[1], M. Varidi[1*], A.M. Mortazavian[2]

1- Department of Food Science and Technology, Faculty of Agriculture, Ferdowsi University of Mashhad, Mashhad, Iran (*- Corresponding Author Email: m.varidi@um.ac.ir)
2- Department of Food Science and Technology, Faculty of Nutrition Sciences and Food Technology, National Nutrition and Food Technology Research Institute, Shahid Beheshti University of Medical Sciences, Tehran, Iran

Received: 22.10.2025
Revised: 09.01.2026
Accepted: 28.01.2026
Available Online: 02.05.2026

How to cite this article:
Mojtahedi, P., Varidi, M., & Mortazavian, A.M. (2026). Encapsulation of vitamins E and C by whey protein concentrate–chitosan via spray drying: Optimization, physicochemical characterization, and release rate analysis. *Iranian Food Science and Technology Research Journal*, *22*(1), 17-34.
https://doi.org/10.22067/ifstrj.2026.96109.1497

Abstract

Increasing concerns about human health have increased demand for functional bioactive compounds, such as vitamins E and C. However, their inherent instability and susceptibility to environmental conditions have restricted their direct application in food systems. Encapsulation strategies offer an effective approach to protect these vitamins from degradation and enable controlled release. This study aimed to develop a stable encapsulation system for vitamins E and C using a whey protein concentrate–chitosan (WPC–CS) complex through spray drying, enabling simultaneous delivery of hydrophilic and lipophilic vitamins. Response surface methodology (RSM) was applied to optimize the formulation with the objective of maximizing encapsulation efficiency while minimizing moisture content and hygroscopicity. The optimal microcapsule exhibited encapsulation efficiencies of 93.83% and 93.6% for vitamins E and C, respectively, with a low moisture content (3.17%) and controlled hygroscopicity (13.18%). Release studies demonstrated a phase-dependent behavior: vitamin C showed negligible release in simulated mouth fluid (0.07% after 10 min) but a substantial cumulative release in simulated gastric fluid (85.74% after 120 min). In contrast, vitamin E exhibited a slower and sustained release profile, with 31.45% released after 120 min in simulated gastric fluid and no detectable release under mouth conditions. Scanning electron microscopy (SEM) analysis revealed that the microcapsules had a predominantly spherical morphology with a wrinkled surface, characteristic of protein–polysaccharide matrices formed during rapid drying. Fourier transform infrared spectroscopy (FTIR) confirmed successful encapsulation of both vitamins through interactions between WPC and CS. These findings indicate that WPC–CS complexes produced via spray drying provide an effective co-encapsulation and controlled-release system, enhancing vitamin stability and offering promising applications in functional foods and pharmaceutical formulations.

Keywords: Controlled release, Microcapsules, Optimize, Spray drier

Introduction

Vitamins are indispensable micronutrients with potent antioxidant properties that play a fundamental role in maintaining cellular homeostasis and protecting biomolecules from oxidative stress. Among them, vitamin E (α-tocopherol) is a lipophilic antioxidant that prevents lipid peroxidation and contributes to

https://doi.org/10.22067/ifstrj.2026.96109.1497

the stabilization of biological membranes (Budinčić *et al*., 2021; Chen, Zhu, Zhao, Zhang, & Wang, 2022). In contrast, vitamin C (ascorbic acid, AA) is a hydrophilic antioxidant that exists in reduced (AA) and oxidized (dehydroascorbic acid, DHA) forms and is essential for scavenging free radicals in aqueous systems (Comunian *et al.*, 2013; Guldiken *et al.*, 2019). Together, these vitamins provide complementary protection against oxidative damage in both lipid and aqueous phases of biological systems. However, despite their recognized health-promoting effects, both vitamins exhibit inherent physicochemical limitations that restrict their practical applications. Vitamin E is poorly soluble in water and shows considerable instability when exposed to oxygen, heat, or light, which significantly limits its functionality in food and pharmaceutical formulations (Rabkin, Tirosh, & Kanner, 2022). Likewise, vitamin C undergoes rapid degradation due to its high sensitivity to oxidation, moisture, and environmental stressors such as temperature and light, leading to reduced stability and bioavailability (Katouzian & Jafari, 2016). These drawbacks highlight the urgent need for innovative approaches to preserve their activity and ensure efficient delivery. Microencapsulation has emerged as a promising solution to address these challenges. By entrapping bioactive molecules within protective wall materials, microencapsulation enhances stability, improves dispersibility, and provides controlled release at targeted sites. Moreover, this technique can be tailored to optimize the physicochemical and functional properties of encapsulated vitamins, thereby extending their shelf life and effectiveness. Various methods have been developed for microcapsule formation, including phase separation, extrusion, complex coacervation, polycondensation, and spray-drying. Among these, spray-drying has gained particular attention due to its scalability, cost-effectiveness, and ability to convert liquid formulations into stable powders with improved storage and release profiles (Chang & Nickerson, 2018; Lan, Ohm, Chen, & Rao, 2021; Li, Zhou, Huang, Li, & Xiao, 2022).

In this context, several studies have successfully applied spray-drying for vitamin encapsulation. For instance, vitamin C has been encapsulated using carriers such as gum Arabic/maltodextrin (Delaporte, Duchemin, Grisel, & Gore, 2024) and whey protein/maltodextrin (Agudelo-Chaparro, Ciro-Velásquez, Sepúlveda-Valencia, & Pérez-Monterroza, 2022) to reduce oxidative degradation. Similarly, vitamin E has been encapsulated with lipid-based or protein–polysaccharide matrices to improve its oxidative stability (Chen *et al.*, 2018; Yousefi, Rajaei, Nateghi, Nodeh, & Rashidi, 2023). Furthermore, co-encapsulation of vitamins C and E has been investigated to develop synergistic dual-phase antioxidant systems capable of functioning in both hydrophilic and lipophilic environments (Chang & Nickerson, 2018). Importantly, recent advances have focused on optimizing wall material composition, drying conditions, and encapsulation efficiency to maximize the stability and bioactivity of vitamins (Askari Vaselabadi *et al*., 2025).

WPC–CS spray-dried microcapsules offer distinct superiority over conventional carriers like liposomes or emulsions for co-encapsulating hydrophilic (e.g., vitamin C) and hydrophobic (e.g., vitamin E) compounds. Unlike liposomes, which suffer from inherent instability, leakage during storage/drying, poor scalability, and low entrapment efficiencies requiring complex stabilization, the protein–polysaccharide matrix yields robust, spherical powders with high efficiency, low moisture, and tailored pH-dependent release suitable for functional foods. This cost-effective, industrially scalable spray-drying process enhances environmental stability and controlled delivery, addressing key limitations of lipid-based systems.

Despite extensive investigations into individual encapsulating agents, the co-encapsulation of vitamins C and E using whey protein concentrate (WPC) and chitosan (CS) in

a controlled spray-drying process has been rarely addressed. Whey protein concentrate (WPC) was selected for its superior emulsifying and film-forming properties, enabling effective entrapment of lipophilic vitamin E. Chitosan (CS) complements WPC via electrostatic complexation, enhancing matrix strength and providing pH-responsive release in gastric conditions for hydrophilic vitamin C protection. The combined functionality of these wall materials in simultaneously protecting hydrophilic and lipophilic vitamins remains underexplored. Therefore, the present study aimed to design and optimize spray-dried microcapsules containing vitamins C and E by employing WPC and CS as wall materials. Specifically, the study investigated the influence of protein-to-polysaccharide ratios on encapsulation efficiency, moisture content, and release behavior under simulated oral and gastric conditions.

Materials and Method

Material

Vitamin E (α-tocopherol, 96% purity, w/w), vitamin C (L-ascorbic acid, 99% purity, w/w), pepsin (from porcine gastric mucosa), and medium molecular weight chitosan (CS, deacetylation degree ≈ 85%) were purchased from Sigma-Aldrich (St. Louis, MO, USA). Whey protein concentrate (WPC, 80% protein, w/w) was obtained from Fonterra (Auckland, New Zealand). Alpha-amylase and other analytical-grade reagents were purchased from Merck (Darmstadt, Germany)

Preparation of CS-WPC Complex Loaded with Vitamins C and E

Vitamin E/WPC Solution

Aqueous solutions of whey protein concentrate (WPC; 2, 3, and 4% w/w) were prepared by magnetic stirring at 1000 rpm for 2 h, followed by homogenization at 14,000 rpm for 3 min using a high-shear homogenizer (T25 digital ULTRA-TURRAX, IKA, Germany). An oil phase containing vitamin E (0.16% w/w), sunflower oil (4% w/w), and Tween 80 (1% w/w) was homogenized under identical conditions and subsequently incorporated into the WPC solution to obtain the vitamin E/WPC mixture (Luo, Zhang, Whent, Yu, & Wang, 2011; Wang *et al*., 2016).

Vitamin C/CS Solution

Chitosan (CS) solutions (0.25, 0.375, and 0.5% w/w) were prepared in 1% acetic acid, stirred at 1000 rpm for 10 min, and homogenized at 14,000 rpm for 3 min using the same homogenizer. Vitamin C (0.9 g/100 g) was then incorporated into the CS solution and homogenized under the same conditions (Desai & Park, 2006; Wang *et al.*, 2016). The CS–vitamin C solution was gradually added to the WPC–vitamin E solution while adjusting the pH to 4.3, followed by homogenization at 14,000 rpm for 2 min to ensure uniform dispersion.

Final Emulsion Characterization

Physicochemical properties (particle size, ζ-potential, viscosity, encapsulation efficiency) were measured according to established protocols and reported in our previous study (Mojtahedi, Mortazavian, & Varidi, 2023). The total solid content of the feed solution prior to spray drying was 3.5–5.5% (w/w), a critical parameter influencing drying efficiency, particle morphology, and microcapsule stability (Bazaria & Kumar, 2016; Fernandes, Borges, Botrel, & de Oliveira, 2014).

Spray Drying

Emulsions were spray-dried using a B-290 LabPlant spray dryer (Switzerland) under controlled conditions: feed rate 3 mL/min, compressed air flow 50 L/min, pump capacity 20%, aspirator speed 90%, inlet air temperature 165 °C, outlet temperature 99–100 °C. These operating conditions were optimized to balance encapsulation efficiency, particle morphology, and vitamin stability, ensuring reproducible production of stable microcapsules. These operating conditions were optimized to achieve a balance between encapsulation efficiency, particle morphology, and vitamin stability, thereby ensuring the production of stable microcapsules.

Encapsulation Efficiency

Encapsulation efficiency (EE) of vitamin C was determined using a modified method by (Desai & Park, 2006). A theoretical 90 mg of encapsulated vitamin C was dissolved in 10 mL of water, then was centrifuged at 4000×g for 30 min (Hettic, Universal 320, USA). The transparent part was passed through a 0.2 µm filter (Millipore, USA) and extracted by HCl (0.1 N) Its absorbance was then measured at 244 nm (ʎmax of vitamin C in HCL 0.1 N) after appropriate dilution using a UV spectrophotometer (Perkin Elmer Lambda-2 UV–Visible Spectrophotometer, Germany). The EE was calculated as:

$$EE(\%) = \frac{Total\ Vitamin\ C - free\ Vitamin\ C}{Total\ Vitamin\ C} \times 100 \quad (1)$$

Where Total Vitamin C indicate Theoretical vitamin C concentration in the emulsion and free vitamin C was obtained using a membrane separation method (Desai & Park, 2006).

For vitamin E, encapsulation efficiency was determined according to (Luo *et al.*, 2011). A theoretical 16 mg of vitamin E was dissolved in 10 mL of water, followed by centrifugation at 4000×g for 30 minutes (Hettic, Universal 320, USA). The transparent part was passed through a 0.2 µm filter and extracted by hexane. Then it was analyzed by measuring the absorbance at 290 nm with a UV/Vis spectrophotometer described in previous studies. Measurements were performed in triplicate for accuracy. EE was calculated using:

$$EE\ (\%) = \frac{Total\ Vitamin\ E - free\ Vitamin\ E}{Total\ Vitamin\ E} \times 100 \quad (2)$$

Moisture Content

The moisture content (MC) of the powdered sample was determined gravimetrically by drying in a forced-circulation oven (UFB 400, Memmert, Germany) at 105 °C until a constant weight was achieved, following the AOAC method (AOAC, 2005).

Moisture Absorption (Hygroscopicity)

The hygroscopicity of the produced powders was determined according to (Comunian *et al.*, 2013). Specifically, 0.2 g of sample was placed in a desiccator containing a saturated solution of Na_2SO_4 at 81% relative humidity. After a storage period of one week, hygroscopicity was expressed as the mass of water absorbed per 100 g of sample.

Cumulative Release

The release profile was examined under two simulated conditions: simulated mouth fluid (SMF) and simulated gastric fluid (SGF).

Release in Simulated Mouth Fluid (SMF) Condition

The *in vitro* release study was conducted in simulated mouth fluid (SMF), prepared with sodium bicarbonate, calcium chloride, potassium phosphates, and α-amylase, and adjusted to pH 6.8. The complex emulsion powder was dispersed in SMF and incubated at 37 °C with continuous stirring (100 rpm) for 10 min. Non-encapsulated vitamins were removed by centrifugation at 30,000×g, after which digestion was terminated by adjusting the pH to 7.5. Vitamin C and vitamin E were subsequently extracted using 0.1 M HCl and hexane, respectively. Their concentrations were quantified by UV–Vis spectrophotometry at 244 nm (vitamin C) and 290 nm (vitamin E) using calibration curves of free standards. Release profiles were obtained from measurements taken at 0, 5, and 10 min (Asprea, Leto, Bergonzi, & Bilia, 2017; Desai & Park, 2006; Luo *et al.*, 2011).

Vitamin Release in Simulated Gastric Fluid (SGF) Condition

Vitamin release was assessed in simulated gastric fluid (SGF; pH 2.0, containing 0.1 M HCl and 0.1% w/v pepsin) at 37 °C under continuous stirring for 2 h. The initial release kinetics were evaluated at 0, 60, and 120 min. Non-encapsulated vitamins were separated by centrifugation at 30,000×g, and digestion was terminated by adjusting the pH to 7.5. Vitamin C and vitamin E were subsequently extracted using 0.1 M HCl and hexane, respectively. Quantification was performed by UV–Vis spectrophotometry at 244 nm (vitamin C) and 290 nm (vitamin E), based on calibration curves of free standards (Desai & Park, 2006; Somchue, Sermsri, Shiowatana, & Siripinyanond, 2009).

Spectroscopic Characterization

The spectroscopic characterization of optimum sample, carriers, and vitamins was conducted using Fourier transform infrared (FTIR) spectroscopy, following the methodology described by (Desai & Park, 2005). Briefly, the powdered samples were compressed into KBr disks and analyzed with an FTIR spectrophotometer (Bruker Vector22, USA) within a wave number range of 4000–400 cm^{-1}.

Scanning Electron Microscopy

The microstructural characterization of the optimized powdered complex emulsion was performed using scanning electron microscopy (SEM, EVO 18 Special Edition, Zeiss, Germany), following a slightly modified method based on (Desai & Park, 2006). To examine the morphological properties, the powdered sample was fixed onto metallic adhesive tapes attached to metallic stubs, which were subsequently coated with a thin layer of gold using an evaporator. Imaging was conducted in secondary electron mode under an acceleration voltage of 15 kV.

Statistical Analysis

Response Surface Methodology (RSM) was applied using Design-Expert software (version 13.0, Stat-Ease Inc., Minneapolis, MN, USA) with a central composite design (CCD) to evaluate the effects of whey protein concentrate (2–4% w/w) and chitosan (0.25–0.5% w/w) on encapsulation efficiency (EE), moisture content (MC), and hygroscopicity. The optimization goals, specifying that the study aimed to maximize encapsulation efficiency while minimizing hygroscopicity and moisture content. Model adequacy was verified through analysis of variance (ANOVA), and coefficient of determination (R^2). Statistical comparisons were performed using Duncan's multiple range test in SPSS software (version 22.0), with significance defined at $p < 0.05$. Confirmatory experiments under the predicted optimal conditions showed close agreement between experimental and predicted values, confirming the reliability of the optimization model

Results and Discussion

Encapsulation Efficiency

The encapsulation efficiency (EE) of vitamins C and E was significantly influenced by the concentrations of whey protein concentrate (WPC) and chitosan (CS) via spray drying ($P \leq 0.05$). whereas the encapsulation efficiency of vitamin E was not significantly affected by these factors ($P > 0.05$). As shown in Table 1, the ANOVA results demonstrated that all examined factors had a significant effect on vitamin C encapsulation efficiency after drying ($p \leq 0.05$).

The model is statistically significant ($p < 0.05$), indicating that the fitted model for vitamin C encapsulation efficiency adequately describes the response within the experimental range. Among them, chitosan concentration had the greatest impact, based on the F-value. Increasing WPC or CS concentration reduced vitamin C encapsulation efficiency, ranging from 93.6% to 77.33% (Fig. 1-a), while vitamin E maintained consistently high efficiency (93.36%–93.83%) (Fig. 1-b) with minimal variation.

Table 1- ANOVA of the effect of chitosan and WPC concentration on the efficiency of vitamin C encapsulation

Source	Sum of Squares	df	Mean Square	F Value	p-value
Model	180.70	3	60.23	7.95	0.0238
WPC (g/100g)	55.05	1	55.05	7.27	0.0430
Chitosan (g/100g)	112.09	1	112.09	14.79	0.0120
WPC* Chitosan	13.57	1	13.57	1.79	0.2385
Residual	37.88	5	7.58		

This difference highlights the structural and physicochemical sensitivities of vitamin C compared to the superior stability of vitamin E under diverse formulation environments (Desai & Park, 2005; Hu *et al.*, 2020; Oliveira *et al.*, 2023). Equation 3 presents the regression model used to predict vitamin C encapsulation

efficiency (EE) based on the actual levels of the parameters (R^2 = 0.7904).

$$Vit\ C - EE(\%) = 96.2705 + 2.4961 * WPC\left(\frac{g}{100g}\right) + 9.6222 * Chitosan\left(\frac{g}{100g}\right) - 14.7333 * WPC\left(\frac{g}{100g}\right) * Chitosan(\frac{g}{100g}) \quad (3)$$

Increasing WPC concentration reduced EE due to protein aggregation, which created voids within the microcapsules and promoted surface deposition of the core compounds. This aggregation altered emulsion viscosity, diminished encapsulation uniformity, and reduced compound retention. The decline in EE may be related to larger particle sizes, which could lead to increased heterogeneity and reduced emulsion stability, ultimately compromising capsule integrity (De Queiroz *et al.*, 2018; Hinnenkamp, Reineccius, & Ismail, 2021; Mojtahedi *et al*., 2023).

Similarly, higher CS concentrations also reduced EE. As a polycation, CS interacts with WPC through electrostatic associations, steric crowding, and hydrogen bonding. When the WPC-to-CS ratio is imbalanced, excessive polymer entanglement decreases system stability and promotes leakage of hydrophilic compounds such as vitamin C.

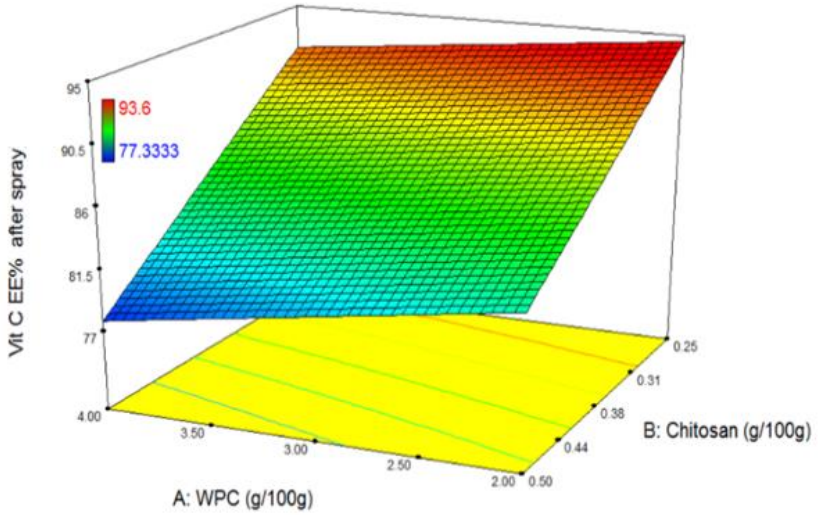

Fig. 1. a–Surface diagram of the effect of WPC and chitosan concentration on EE vitamin C

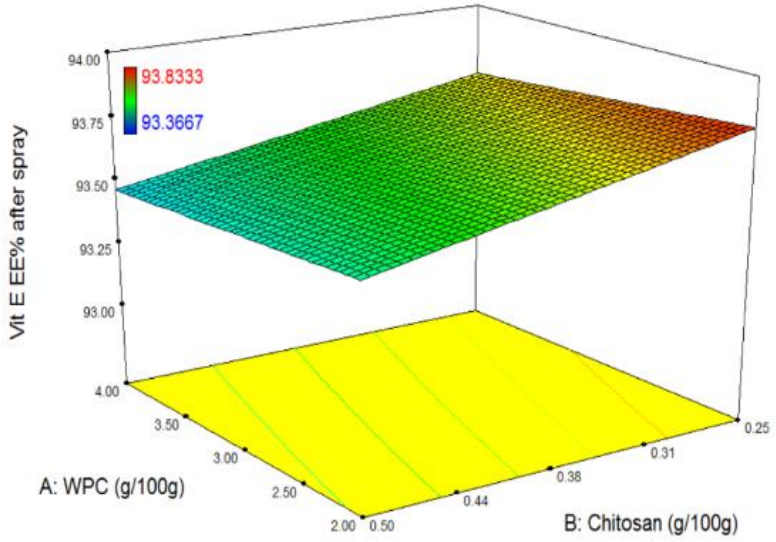

Fig. 1. b- Surface plot of the effect of WPC and chitosan concentration on EE vitamin E

This electrostatic-driven mechanism explains why balanced formulations maintained higher EE, whereas excessive wall material concentrations reduced efficiency (Liu, Jing, Han, Zhang, & Tian, 2019). In contrast, encapsulation efficiency for vitamin E remained relatively stable, owing to its lipophilic nature and lower sensitivity to variations in wall material concentration (Barbosa, Borsarelli, & Mercadante, 2005). This highlights the distinct physicochemical behavior of hydrophilic versus lipophilic compounds in encapsulation systems, where electrostatic interactions play a dominant role for hydrophilic molecules but less so for lipophilic ones. An increase in particle size combined with a decrease in zeta potential directly compromised colloidal stability and thereby reduced EE. Larger particles exhibited lower surface area-to-volume ratios and a higher tendency to aggregate, which led to void formation and heterogeneity in the capsule wall. At the same time, reduced zeta potential indicated weaker electrostatic repulsion between particles, facilitating aggregation and leakage of encapsulated compounds. This phenomenon was often accompanied by higher PDI values, confirming reduced uniformity and stability of the emulsion system.

These findings are consistent with previous studies, supporting and corroborating the results obtained in the present work. Mojtahedi *et al.* (2023) demonstrated that increased particle size and reduced zeta potential in protein-based complexes resulted in lower emulsion stability and decreased EE. Likewise, Hinnenkamp *et al*. (2021) and De Queiroz *et al*. (2018) showed that protein aggregation and diminished surface charge promoted particle heterogeneity and reduced encapsulation efficiency of hydrophilic compounds. Xu, Lv, Mu, Zhou, & Yang, (2021) reported an EE of 86.3% for α-tocopherol in WPC–CS complexes, while Agustinisari, Mulia, & Nasikin, (2020) demonstrated enhanced emulsion stability and improved EE for eugenol. Wang *et al. (2021)* highlighted superior efficiency (60.70%) in WPI nanocomplexes compared to ACNs, CS, and CMC formulations. De Queiroz *et al*. (2018) achieved remarkably high efficiencies (98.5%) for CS–WPC microcapsules, and Lekshmi *et al*. (2019) documented an EE of 75.40% for squalene encapsulated with WPC and CS. These studies reinforce the conclusion that balanced wall material systems are critical for achieving high encapsulation efficiency.

Moisture Content

The moisture content of microcapsules prepared with varying concentrations of WPC and CS was systematically analyzed. Results indicated a significant inverse relationship between wall material concentration and moisture content ($P < 0.05$), as illustrated in Table 2 and Fig. 2.

Table 2- ANOVA of the effect of chitosan and WPC concentration on the efficiency of moisture content

Source	Sum of Squares	df	Mean Square	F Value	p-value
Model	0.16	3	0.053	52.05	0.0003
WPC (g/100g)	0.15	1	0.15	148.9	0.0001
Chitosan (g/100g)	0.00735	1	0.00735	7.18	0.0439
WPC* Chitosan	0.00007	1	0.00007	0.068	0.8050
Residual	0.00512	5	0.00104		

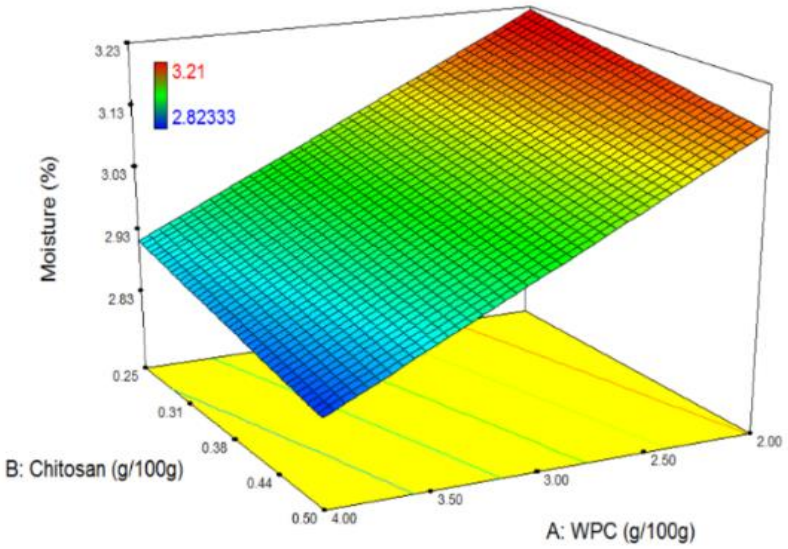

Fig. 2. Surface plot of the effect of WPC and chitosan concentration on moisture content

The model was found to be statistically significant ($p < 0.05$), suggesting that it provides an adequate representation of the response across the experimental range. Higher concentrations of WPC and CS reduced moisture content (2.82% at 4% WPC, 0.5% CS), whereas lower concentrations resulted in greater retention (3.21%). This reduction highlights the role of wall materials in regulating water evaporation and underscores final moisture content as a key quality control parameter affecting stability and product quality (Díaz-Montes, 2023). Equation 4 presents the regression model used to predict the moisture content of microcapsules based on the actual levels of the parameters (R^2 = 0.9690).

$$Moisture(\%) = 3.5795 - 0.1469 * WPC\left(\frac{g}{100g}\right) - 0.18 * Chitosan\left(\frac{g}{100g}\right) - 0.0333 * WPC\left(\frac{g}{100g}\right) * Chitosan\left(\frac{g}{100g}\right) \quad (4)$$

The functional groups in WPC (–NH_2, –COOH, –OH) and CS (–OH, –NH_2) facilitate stronger interactions with water molecules, promoting faster evaporation and thereby reducing residual moisture in the final powder. At higher concentrations, these groups facilitate more efficient water binding during atomization and promote faster evaporation during spray-drying, thereby reducing residual moisture. This functional group-driven mechanism explains the observed decrease in moisture content with increasing wall material concentration, consistent with previous studies (Chen *et al.*, 2018; Pinto *et al.*, 2021).

Similar findings have been reported for other wall materials such as gum Arabic (Frascareli, Silva, Tonon, & Hubinger, 2012) and UDP-coated WPC (Choi & Chang, 2018), where functional groups enhanced water interactions and reduced moisture retention. Maintaining moisture levels within the range of 3–10% has been shown to improve oxidative stability of microencapsulated oils (Klaypradit & Huang, 2008), while a final moisture content of ~5% is considered optimal for structural stability and retention of bioactive compounds (Stabrauskiene, Pudziuvelyte, & Bernatoniene, 2024).

Moisture Absorption (Hygroscopicity)

The encapsulation system's hygroscopicity was significantly influenced by WPC concentration ($P \leq 0.05$), while its interaction with CS was not significant ($P > 0.05$). As shown in Table 3, the ANOVA results demonstrated that all factors had a significant effect on hygroscopicity ($p \leq 0.05$).

Table 3- ANOVA of the effect of chitosan and WPC concentration on the efficiency of moisture absorption

Source	Sum of Squares	df	Mean Square	F Value	p-value
Model	8.97	3	2.99	82.01	0.0001
WPC (g/100g)	8.09	1	8.09	221.84	0.0001
Chitosan (g/100g)	0.88	1	0.88	24.18	0.0044
WPC* Chitosan	0.00027	1	0.00027	0.0076	0.9338
Residual	0.18	5	0.036		

The model is statistically significant ($p < 0.05$), indicating that the fitted model adequately describes the response within the experimental range. Equation 5 presents the regression model used to predict the moisture absorption of microcapsules based on the actual levels of the parameters ($R^2 = 0.9801$).

$$Moisture(\%) = 9.775 + 1.1361 * WPC\left(\frac{g}{100g}\right) + 2.8666 * Chitosan\left(\frac{g}{100g}\right) + 0.0666 * WPC\left(\frac{g}{100g}\right) * Chitosan\left(\frac{g}{100g}\right) \quad (5)$$

Increasing WPC and CS levels raised hygroscopicity up to 15.8%, indicating their role in water retention and microcapsule stability (Fig. 3). The hydrophilic functional groups of chitosan (–OH, $–NH_2$) and WPC promote moisture uptake (Carvalho-Silva, 2013), which contributes to controlled swelling and the formation of a gel-like matrix. This matrix acts as a physical barrier, limiting oxygen and light penetration and thereby enhancing the stability of encapsulated bioactive compounds (Chen, Mao, Hou, Yuan, & Gao, 2020). Moisture absorption also improves the physical integrity of spray-dried microcapsules by softening the wall structure and reducing cracking or rupture (Li *et al.*, 2022). When the dried powders are exposed to ambient humidity, the hydrophilic functional groups act as active sites for water sorption. Their abundance in CS and WPC increases the powder's affinity for atmospheric moisture, leading to higher hygroscopicity. From a biological perspective, enhanced hydration increases biocompatibility and supports controlled release in gastrointestinal environments (Pateiro *et al.*, 2021).

However, excessive moisture absorption may compromise wall integrity, accelerate compound release, and promote microbial growth, underscoring the need for optimized wall composition and humidity control. Comparisons with previous studies confirm the hygroscopic nature of WPC-based formulations. Beetroot spray-dried microparticles retained 11.13–15.59% moisture (Bazaria & Kumar, 2016), while WPC–inulin blends encapsulating rosemary essential oil absorbed 15.7–17.1% moisture (Fernandes *et al.*, 2014). These findings highlight the importance of balancing WPC and CS concentrations to achieve structural stability while minimizing adverse effects of excessive hydration.

Release of Encapsulated Vitamins C and E under Simulated Mouth Fluids

The release behaviors of vitamins C and E under SMF conditions at 0, 5, and 10 minutes are shown in Table 4, Statistical analysis confirmed that neither chitosan nor WPC concentration had a significant effect on the release of vitamins E and C under simulated mouth conditions ($p > 0.05$). Vitamin E showed no measurable release throughout the 10-minute testing period, indicating strong retention within the encapsulation matrix under neutral pH conditions. In contrast, vitamin C exhibited only a minimal release (~0.1%), likely due to electrostatic interactions between WPC and CS at neutral pH, which restrict diffusion and solubility. Such interactions play a key role in controlling release behavior.

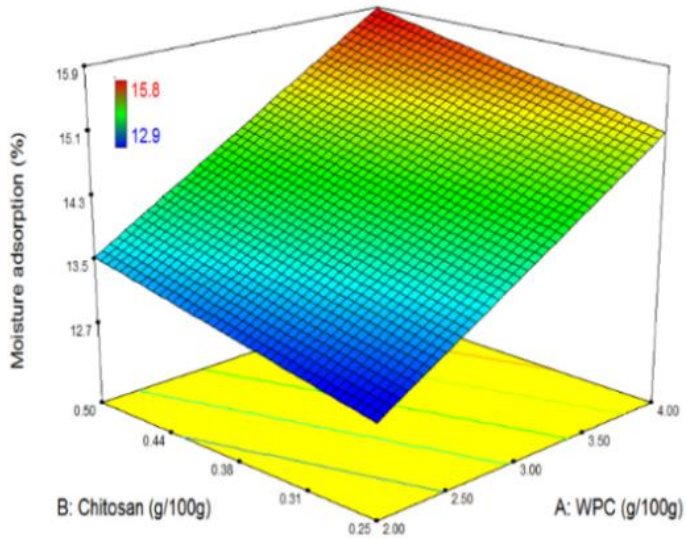

Fig. 3. Surface plot of the effect of WPC and chitosan concentration on moisture absorption

Table 4- Comparison of average results of vitamin E and C release studies in simulated mouth fluid

Row	WPC (%)	Chitosan (%)	Time (Min)	Vitamin C release (%)	Vitamin E release (%)
1	2	0.25	0	0.06[a]	0[a]
2	2	0.25	5	0.07[a]	0[a]
3	2	0.25	10	0.07[a]	0[a]

Different letters in each column indicate significant differences (P<0.05).

The remarkably low release observed under SMF conditions has important practical implications. By preventing premature diffusion of bioactive compounds in the oral cavity, the encapsulation system effectively avoids undesirable taste perception and enhances consumer acceptability. This functional control ensures that sensitive compounds remain protected until reaching the stomach, where targeted release and absorption can occur.

Encapsulation plays a pivotal role in controlling the release of bioactive compounds within target environments. The strong retention of vitamins in neutral conditions (e.g., SMF) ensures that premature release is minimized. This functional control must be maintained across different stages of digestion to guarantee that bioactive compounds are only released in the intended site, such as the stomach, where absorption can occur effectively. The controlled release of vitamins provides an advantage by reducing nitrite accessibility through enhanced interaction with encapsulated antioxidants. This targeted release not only improves product stability but also strengthens food safety by limiting nitrosamine formation (Rot, Kovačević, Habschied, & Mastanjević, 2025).

Previous studies have highlighted similar applications of encapsulation systems. For instance, Mohammadi, Ehsani, & Bakhoda, (2018) demonstrated that alginate-based microcapsules modulated caffeine release, while De Queiroz *et al*. (2018), reported high retention efficiencies in CS–WPC complexes. Similar findings have been reported in probiotic encapsulation studies, where controlled release ensured survival through the oral phase and targeted delivery in the intestine (Anal & Singh, 2007).

Release of Encapsulated Vitamins C and E under Simulated Gastric Fluid

Table 5 provides a comparative overview of the release patterns of vitamins C and E under simulated gastric fluid (SGF) conditions at 0, 60, and 120 minutes, using optimized samples obtained through response surface

methodology (RSM). Both vitamins C and E exhibited a clear time-dependent increase in release under simulated gastric conditions. Statistical analysis confirmed significant differences among the three sampling times ($p < 0.05$).

Table 5- Comparison of average results of vitamin E and C release studies in simulated gastric floid

Row	WPC (%)	Chitosan (%)	Time (Min)	Vitamin C Release (%)	Vitamin E Release (%)
1	2	0.25	0	56.85[a]	0.93[a]
2	2	0.25	60	69.44[b]	9.58[b]
3	2	0.25	120	85.74[c]	31.45[c]

Different letters in each column indicate significant differences (P<0.05).

Vitamin E exhibited minimal release at the beginning (0.93%), which gradually increased to 31.45% after 120 minutes. In contrast, vitamin C showed a markedly higher release, with 56.85% released initially and a cumulative release of 85.74% after 120 minutes. This behavior can be explained by several mechanistic factors. First, under SGF conditions, the release mechanism is strongly influenced by gastric acidity and enzymatic activity. The acidic pH (pH ≈ 1.2) leads to protonation of CS, which reduces its electrostatic binding capacity and accelerates vitamin C diffusion. Simultaneously, pepsin-induced denaturation of whey protein disrupts the protein matrix, further enhancing release. In contrast, vitamin E release remains limited because of its hydrophobic nature and stronger entrapment within the polymeric wall, which resists enzymatic degradation and proton-mediated diffusion (Tan *et al.*, 2020; Xu, Tang, Yang, Wang, & Zhou, 2020). Consistent with these observations, Hu *et al.* (2020) reported curcumin release values between 20.3% and 24.8% from whey protein–CS microcapsules after two hours in SGF, reinforcing the influence of wall material composition, electrostatic interactions, and gastric acidity on controlled release. Similarly, Pudjiastuti *et al.* (2020) emphasized that release is governed not only by solubility but also by polymer structure and capsule integrity. Xu *et al.* (2021) reported accelerated release of hydrophilic bioactive in acidic environments due to protonation-induced matrix loosening, while Barbosa *et al.* (2005), emphasized the stability of lipophilic compounds in protein–polysaccharide complexes. These findings emphasize that nutrient release is not solely dependent on solubility but is governed by a complex interplay of polymer structure, electrostatic binding, enzymatic degradation, and matrix hydrophobicity, all of which must be considered in designing targeted encapsulation strategies for enhanced bioavailability.

Scanning Electron Microscopy

Scanning electron microscopy (SEM) images revealed notable variations in particle size and surface morphology among the encapsulated vitamins C and E, indicating a polydisperse nature of the microcapsules (Fig. 4). This observation is consistent with the inherent characteristics of spray-drying processes, where atomization typically generates particles with non-uniform sizes due to rapid droplet formation and drying kinetics (Lekshmi *et al.*, 2019). Such polydispersity is a well-documented phenomenon in spray-dried systems and is often influenced by nozzle type, feed composition, and drying temperature (Schmitt, Baumann, & Morgen, 2022).

The wrinkled surface morphology observed in the SEM micrographs is likely a result of premature solidification of the wall material prior to complete internal vapor expansion. This leads to surface collapse and structural deformation, as previously reported in systems encapsulating linoleic acid with maltodextrin and WPC (Choi, Ryu, Kwak, & Ko, 2010). Similar morphological features have been described in recent studies, where wall material composition and drying conditions were shown to significantly affect particle shape and integrity (Díaz-Montes, 2023; Wang & Mutukumira, 2022).

Moreover, the sample containing 2% WPC and 0.25% chitosan exhibited smaller particle size and higher dispersion, suggesting improved emulsification and stabilization during atomization. This aligns with findings by (Suwannasang *et al.*, 2022), who demonstrated that optimized protein–polysaccharide combinations enhance colloidal stability and reduce particle aggregation in spray-dried formulations. Overall, the observed polydispersity and morphological variations underscore the importance of wall material selection and process optimization in achieving consistent particle characteristics.

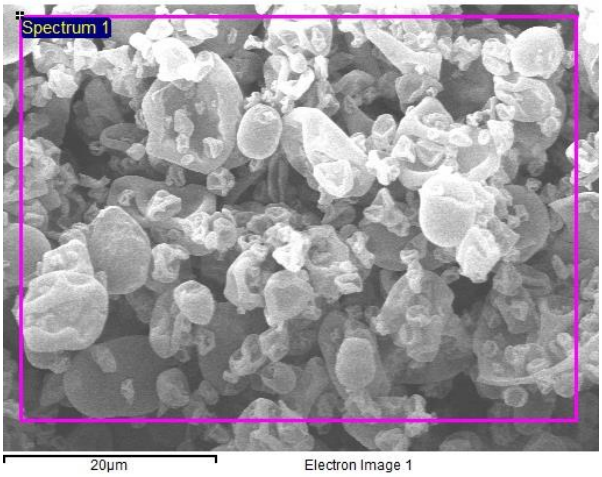

Fig. 4. Scanning electron microscope images of sample9

FT-IR Analysis

The FT-IR analysis can be applied to the intermolecular and structural characterization of complex materials. The FTIR spectrums of encapsulated vitamin C and vitamin E in samples (9) are shown in Fig. 5. Vitamin C shows property bands in the number of waves that vary from 2924–3989 cm^{-1} owing to CH3 stretching. Vitamin E exhibits symmetric bending and methyl asymmetric peaks emerge at 1403 cm^{-1} and 1442 cm^{-1}, respectively. The ether group emerges at 1075 cm^{-1}. The phenol C–O stretching emerges at 1245 cm^{-1} while the C–H stretching absorption peak appears at 2924 cm^{-1} and 2966 cm^{-1}.

FTIR analysis revealed characteristic peaks for vitamin C (2911–3024 cm^{-1}) and vitamin E (1378 and 1466 cm^{-1}), along with functional groups such as ether (1093 cm^{-1}), phenol (1260 cm^{-1}), and C-H stretching (2847 and 2918 cm^{-1}), that are consistent with Pang *et al.* (2017). WPC showed distinctive bands between 1651 and 1547 cm^{-1} corresponding to amide I and II, in line with Dickinson, Radford, & Golding, (2003), who reported peaks in this range. An amide III band at 1245 cm^{-1}, attributed to N-H deformation and C=N stretching, was also observed. Chitosan exhibited peaks at 1075 cm^{-1} (C-O-C), 3289 cm^{-1} (-OH and NH2 stretching), 1651 cm^{-1} (amide I), and 2966 cm^{-1} (CH stretching), as reported by Lekshmi *et al.* (2019). All characteristic peaks of the wall materials and vitamins appeared similarly in the spectra of encapsulated samples, indicating no structural interaction between core and wall materials. Muley & Singhal (2020) confirmed conjugation between chitosan and WPI through bands at 1546.09 and 1639.58 cm^{-1}. Finally, in our study, the conjugation confirmation between CS and WPC was seen from bands at 1547 and 1651 cm^{-1} that showed the Schiff base formation. It was formed between the groups of amino (CS and WPC) and the groups of carboxylic and/or hydroxyl of acetic acid. The observed alterations in the structural fractions might result from the conjugation of WPC with CS.

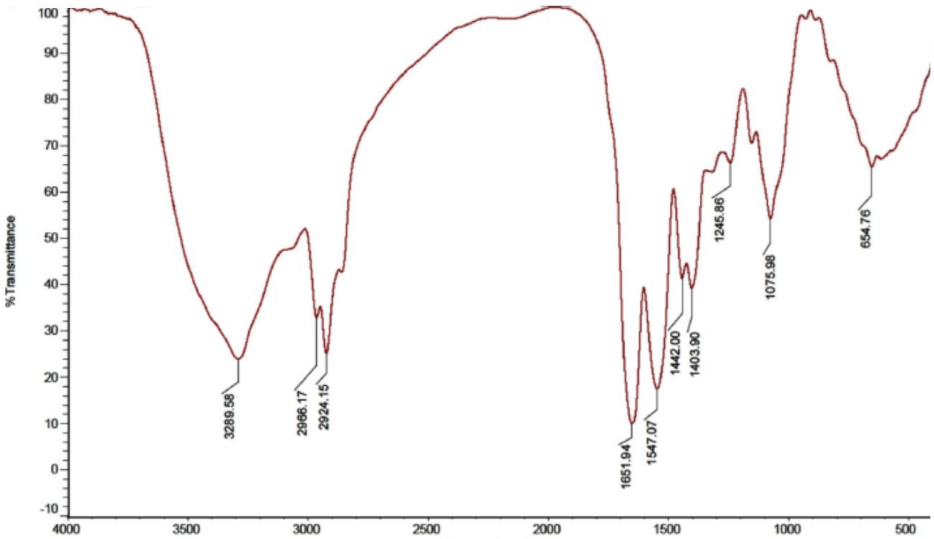

Fig. 5. FTIR spectra of sample 9

Conclusion

A new set of wall materials was used to prepare microcapsules. CS-WPC was confirmed as an effective delivery system for vitamins C and E, showing high encapsulation efficiency and optimal moisture content and water absorption capacity. The optimal formulation, with WPC at 2.0%, CS at 0.25%, was selected. The microcapsules were mostly spherical with rugged surfaces. SEM images revealed that the particle size was fine and well-scattered. Schiff base formation between CS and WPC was confirmed by bands at 1547 and 1651 cm^{-1}, indicating a reaction between acetic acid's carboxyl groups and amino groups in both polymers. These structural changes are likely due to the conjugation process. The results showed that the encapsulated vitamins remained stable in SMF but were released in SGF. Overall, encapsulating vitamins C and E in CS-WPC via spray drying appears to be a promising approach for developing stable encapsulates for functional foods.

Author Contribution

Pooneh Mojtahedi: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Software, Validation, Visualization, Writing – original draft. **Mehdi Varidi:** Supervision, Project administration, Resources, Funding acquisition, Validation, Writing– review and editing. **Amir M. Mortazavian:** Supervision, Project administration, Resources, Funding acquisition, Validation, Writing – review and editing.

Founding Source

The authors declare that they have no competing interests and no funding source.

References

1. Agudelo-Chaparro, J., Ciro-Velásquez, H.J., Sepúlveda-Valencia, J.U., & Pérez-Monterroza, E.J. (2022). Microencapsulation of *Lactobacillus rhamnosus* ATCC 7469 by spray drying using maltodextrin, whey protein concentrate and trehalose. *Food Science and Technology International*, *28*(6), 476-488. https://doi.org/10.1177/10820132211020621
2. Agustinisari, I., Mulia, K., & Nasikin, M. (2020). Physical and chemical properties of encapsulated eugenol using whey protein-maltodextrin mixed solution and chitosan as

emulsifier and wall materials. *AIP Conference Proceedings, 2255*(1), 1-7. https://doi.org/10.1063/5.0013758

3. Anal, A.K., & Singh, H. (2007). Recent advances in microencapsulation of probiotics for industrial applications and targeted delivery. *Trends in Food Science & Technology*, *18*(5), 240-251. https://doi.org/10.1016/j.tifs.2007.01.004
4. AOAC. (2005). Determination of moisture, ash ,protein and fat. Official method of analysis of the association of analytical chemists. In: AOAC Washington DC.
5. Askari Vaselabadi, S., Gharibzahedi, S.M.T., Greiner, R., Vale, J.M., Ovenseri, A.C., Rashidinejad, A., & Roohinejad, S. (2025). Advancements in spray-drying for the microencapsulation of fat-soluble vitamins: Stability, bioavailability, and applications. *Journal of Food Biochemistry*, *2025*(1), 9974476. https://doi.org/10.1155/jfbc/9974476
6. Asprea, M., Leto, I., Bergonzi, M.C., & Bilia, A.R. (2017). Thyme essential oil loaded in nanocochleates: Encapsulation efficiency, in vitro release study and antioxidant activity. *LWT*, *77*, 497-502. https://doi.org/10.1016/j.lwt.2016.12.006
7. Barbosa, M., Borsarelli, C., & Mercadante, A. (2005). Light stability of spray-dried bixin encapsulated with different edible polysaccharide preparations. *Food Research International*, *38*(8-9), 989-994. https://doi.org/10.1016/j.foodres.2005.02.018
8. Bazaria, B., & Kumar, P. (2016). Effect of whey protein concentrate as drying aid and drying parameters on physicochemical and functional properties of spray dried beetroot juice concentrate. *Food Bioscience*, *14*, 21-27. https://doi.org/10.1016/j.fbio.2015.11.002
9. Budinčić, J.M., Petrović, L., Đekić, L., Fraj, J., Bučko, S., Katona, J., & Spasojević, L. (2021). Study of vitamin E microencapsulation and controlled release from chitosan/sodium lauryl ether sulfate microcapsules. *Carbohydrate polymers*, *251*, 116988. https://doi.org/10.1016/J.CARBPOL.2020.116988
10. Carvalho-Silva, L.B.D. (2013). Physico-chemical properties of milk whey protein agglomerates for use in oral nutritional therapy *Food and Nutrition Sciences*, *4*(9), 69-78. https://doi.org/10.4236/fns.2013.49a210
11. Chang, C., & Nickerson, M.T. (2018). Encapsulation of omega 3-6-9 fatty acids-rich oils using protein-based emulsions with spray drying. *Journal of Food Science and Technology*, *55*(8), 2850-2861. https://doi.org/10.1007/s13197-018-3257-0
12. Chen, H., Mao, L., Hou, Z., Yuan, F., & Gao, Y. (2020). Roles of additional emulsifiers in the structures of emulsion gels and stability of vitamin E. *Food Hydrocolloids*, *99*, 105372. https://doi.org/10.1016/j.foodhyd.2019.105372
13. Chen, X., McClements, D.J., Wang, J., Zou, L., Deng, S., Liu, W., Yan, C., Zhu, Y., Cheng, C., & Liu, C. (2018). Coencapsulation of (−)-Epigallocatechin-3-gallate and quercetin in particle-stabilized W/O/W emulsion gels: Controlled release and bioaccessibility. *Journal of Agricultural and Food Chemistry*, *66*(14), 3691-3699. https://doi.org/10.1021/ACS.JAFC.7B05161
14. Chen, Y., Zhu, C., Zhao, Y., Zhang, S., & Wang, W. (2022). Transcriptomics integrated with changes in cell wall material of chestnut (*Castanea mollissima* Blume) during storage provides a new insight into the "calcification" process. *Foods*, *11*(8), 1136. https://doi.org/10.3390/foods11081136
15. Choi, K.-O., Ryu, J., Kwak, H.-S., & Ko, S. (2010). Spray-dried conjugated linoleic acid encapsulated with Maillard reaction products of whey proteins and maltodextrin. *Food Science and Biotechnology*, *19*(4), 957-965. https://doi.org/10.1007/S10068-010-0134-7
16. Choi, Y.-R., & Chang, Y.H. (2018). Microencapsulation of gallic acid through the complex of whey protein concentrate-pectic polysaccharide extracted from *Ulmus davidiana*. *Food Hydrocolloids*, *85*, 222-228. https://doi.org/10.1016/j.foodhyd.2018.07.022.

17. 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
18. De Queiroz, J.L.C., Costa, R.O.D.A., Matias, L.L.R., De Medeiros, A.F., Gomes, A.F.T., Pais, T.D.S., Passos, T.S., Maciel, B.L.L., Dos Santos, E.A., & Morais, A.H.D.A. (2018). Chitosan-whey protein nanoparticles improve encapsulation efficiency and stability of a trypsin inhibitor isolated from *Tamarindus indica* L. *Food Hydrocolloids*, *84*, 247-256. https://doi.org/10.1016/j.foodhyd.2018.06.010
19. Delaporte, A., Duchemin, B., Grisel, M., & Gore, E. (2024). Impact of wall material-to-active ratio in the stability of spray-dried ascorbic acid using maltodextrin and gum arabic. *Molecules*, *29*(15), 3587. https://doi.org/10.3390/molecules29153587
20. Desai, K., & Park, H. (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
21. Desai, K., & Park, H.J. (2006). Effect of manufacturing parameters on the characteristics of vitamin C encapsulated tripolyphosphate-chitosan microspheres prepared by spray-drying. *Journal of Microencapsulation*, *23*(1), 91-103. https://doi.org/10.1080/02652040500435436
22. Díaz-Montes, E. (2023). Wall materials for encapsulating bioactive compounds via spray-drying: A review. *Polymers*, *15*(12), 2659. https://doi.org/10.3390/polym15122659
23. Dickinson, E., Radford, S.J., & Golding, M. (2003). Stability and rheology of emulsions containing sodium caseinate: combined effects of ionic calcium and non-ionic surfactant. *Food Hydrocolloids*, *17*(2), 211-220. https://doi.org/10.1016/S0268-005X(02)00055-3
24. Fernandes, R.V.D.B., Borges, S.V., Botrel, D.A., & de Oliveira, C.R. (2014). Physical and chemical properties of encapsulated rosemary essential oil by spray drying using whey protein–inulin blends as carriers. *International Journal of Food Science and Technology*, *49*(6), 1522-1529. https://doi.org/10.1111/ijfs.12449
25. Frascareli, E., Silva, V., Tonon, R., & Hubinger, M. (2012). Effect of process conditions on the microencapsulation of coffee oil by spray drying. *Food and Bioproducts Processing*, *90*(3), 413-424. https://doi.org/10.1016/j.fbp.2011.12.002
26. Guldiken, B., Linke, A., Capanoglu, E., Boyacioglu, D., Kohlus, R., Weiss, J., & Gibis, M. (2019). Formation and characterization of spray dried coated and uncoated liposomes with encapsulated black carrot extract. *Journal of Food Engineering*, *246*, 42-50. https://doi.org/10.1016/j.jfoodeng.2018.10.025
27. Hinnenkamp, C., Reineccius, G., & Ismail, B.P. (2021). Efficient encapsulation of fish oil: Capitalizing on the unique inherent characteristics of whey cream and hydrolyzed whey protein. *Journal of Dairy Science*, *104*(6), 6472-6486. https://doi.org/10.3168/jds.2020-19880
28. Hu, Y., He, C., Jiang, C., Liao, Y., Xiong, H., & Zhao, Q. (2020). Complexation with whey protein fibrils and chitosan: A potential vehicle for curcumin with improved aqueous dispersion stability and enhanced antioxidant activity. *Food Hydrocolloids*, *104*, 105729. https://doi.org/10.1016/j.foodhyd.2020.105729
29. Katouzian, I., & Jafari, S.M. (2016). Nano-encapsulation as a promising approach for targeted delivery and controlled release of vitamins. *Trends in Food Science & Technology*, *53*, 34-48. https://doi.org/10.1016/j.tifs.2016.05.002
30. Klaypradit, W., & Huang, Y.-W. (2008). Fish oil encapsulation with chitosan using ultrasonic atomizer. *LWT-Food Science and Technology*, *41*(6), 1133-1139. https://doi.org/10.1016/j.lwt.2007.06.014
31. Lan, Y., Ohm, J.-B., Chen, B., & Rao, J. (2021). Microencapsulation of hemp seed oil by pea protein isolate −sugar beet pectin complex coacervation: Influence of coacervation pH and

wall/core ratio. *Food Hydrocolloids*, *113*, 106423. https://doi.org/10.1016/j.foodhyd.2020.106423

32. Lekshmi, R.K., Rahima, M., Chatterjee, N., Tejpal, C., Anas, K., Vishnu, K., Sarika, K., Asha, K., Anandan, R., & Suseela, M. (2019). Chitosan–Whey protein as efficient delivery system for squalene: Characterization and functional food application. *International Journal of Biological Macromolecules*, *135*, 855-863. https://doi.org/10.1016/j.ijbiomac.2019.05.153
33. Li, K.-Y., Zhou, Y., Huang, G.-Q., Li, X.-D., & Xiao, J.-X .(2022). Preparation of powdered oil by spray drying the Pickering emulsion stabilized by ovalbumin–gum Arabic polyelectrolyte complex. *Food Chemistry*, *391*, 133223. https://doi.org/10.1016/J.FOODCHEM.2022.133223
34. Liu, Q., Jing, Y., Han, C., Zhang, H., & Tian, Y. (2019). Encapsulation of curcumin in zein/caseinate/sodium alginate nanoparticles with improved physicochemical and controlled release properties. *Food Hydrocolloids*, *93*, 432-442. https://doi.org/10.1016/j.foodhyd.2019.02.003
35. 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
36. Mohammadi, N., Ehsani, M.R., & Bakhoda, H. (2018). Design and evaluation of the release characteristics of caffeine-loaded microcapsules in a medicated chewing gum formulation. *Food Biophysics*, *13*(3), 240-249. https://doi.org/10.1007/s11483-018-9530-y
37. Mojtahedi, P., Mortazavian, S.A.M., & Varidi, M. (2023). Encapsulation of vitamins E and C in whey protein concentrate-chitosan emulation: Physicochemical properties. *Management Strategies and Engineering Sciences*, *5*(4), 136-146. https://msesj.com/index.php/mses/article/view/183
38. Muley, A.B., & Singhal, R.S. (2020). Extension of postharvest shelf life of strawberries (*Fragaria ananassa*) using a coating of chitosan-whey protein isolate conjugate. *Food Chemistry*, *329*, 12-72. https://doi.org/10.1016/j.foodchem.2020.127213
39. Oliveira, N.L., Espinal-Ruiz, M., Neves, I.C.O., Silva, S.H., de Resende, J.V., & Rogers, M.A. (2023). Evaluation of α-tocopherol microencapsulation stability with either coconut oil or canola oil cores in Greek yogurt and butter. *Food Chemistry Advances*, *2*, 100277. https://doi.org/10.1016/j.focha.2023.100277
40. Pang, Y., Qin, A., Lin, X., Yang, L., Wang, Q., Wang, Z., Shan, Z., Li, S., Wang, J., & Fan, S. (2017). Biodegradable and biocompatible high elastic chitosan scaffold is cell-friendly both in vitro and in vivo. *Oncotarget*, *8*(22), 35583. https://doi:10.18632/oncotarget.14709
41. Pateiro, M., Gómez, B., Munekata, P.E., Barba, F.J., Putnik, P., Kovačević, D.B., & Lorenzo, J.M. (2021). Nanoencapsulation of promising bioactive compounds to improve their absorption, stability, functionality and the appearance of the final food products. *Molecules*, *26*(6), 1547. https://doi.org/10.3390/molecules26061547
42. Pinto, J.T., Faulhammer, E., Dieplinger, J., Dekner, M., Makert, C., Nieder, M., & Paudel, A. (2021). Progress in spray-drying of protein pharmaceuticals: Literature analysis of trends in formulation and process attributes. *Drying Technology*, *39*(11), 1415-1446. https://doi.org/10.1080/07373937.2021.1903032
43. Pudjiastuti, P., Wafiroh, S., Hendradi, E., Darmokoesoemo, H., Harsini, M., Fauzi, M.A.R.D., Nahar, L., & Sarker, S.D. (2020). Disintegration, in vitro dissolution, and drug release kinetics profiles of k-carrageenan-based nutraceutical hard-shell capsules containing salicylamide. *Open Chemistry*, *18*(1), 226-231. https://doi.org/10.1515/chem-2020-0028
44. Rabkin, B., Tirosh, O., & Kanner, J. (2022). Reactivity of vitamin E as an antioxidant in red meat and the stomach medium. *Journal of Agricultural and Food Chemistry*, *70*(38), 12172-12179. https://doi.org/10.1021/ACS.JAFC.2C03674

45. Rot, T., Kovačević, D., Habschied, K., & Mastanjević, K. (2025). N-nitrosamines in meat products: formation, detection and regulatory challenges. *Processes*, *13*(5), 1555. https://doi.org/10.3390/pr13051555
46. Schmitt, J.M., Baumann, J.M., & Morgen, M.M. (2022). Predicting spray dried dispersion particle size via machine learning regression methods: Schmitt, Baumann and Morgen. *Pharmaceutical Research*, *39*(12), 3223-3239. https://doi.org/10.1007/s11095-022-03370-3
47. Somchue, W., Sermsri, W., Shiowatana, 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
48. Stabrauskiene, J., Pudziuvelyte, L., & Bernatoniene, J. (2024). Optimizing encapsulation: Comparative analysis of spray-drying and freeze–drying for sustainable recovery of bioactive compounds from *Citrus x paradisi* L. peels. *Pharmaceuticals*, *17*(5), 596. https://doi.org/10.3390/ph17050596
49. Suwannasang, S., Zhong, Q., Thumthanaruk, B., Uttapap, D., Puttanlek, C., Vatanyoopaisarn, S., & Rungsardthong, V. (2022). Optimization of wall material composition for production of spray-dried Sacha inchi oil microcapsules with desirable physicochemical properties. *Food and Bioprocess Technology*, *15*(11), 2499-2514. https://doi.org/10.1007/s11947-022-02893-2
50. Tan, Y., Li, R., Liu, C., Mundo, J.M., Zhou, H., Liu, J., & McClements, D.J. (2020). Chitosan reduces vitamin D bioaccessibility in food emulsions by binding to mixed micelles. *Food & Function*, *11*(1), 187-199. https://doi.org/10.1039/C9FO02164G
51. Wang, F., & Mutukumira, A.N. (2022). Microencapsulation of *Limosilactobacillus reuteri* DPC16 by spray drying using different encapsulation wall materials. *Journal of Food Processing and Preservation*, *46*(10), e16880. https://doi.org/10.1111/jfpp.16880
52. Wang, L., Gao, Y., Li, J., Subirade, M., Song, Y., & Liang, L. (2016). Effect of resveratrol or ascorbic acid on the stability of α-tocopherol in O/W emulsions stabilized by whey protein isolate: Simultaneous encapsulation of the vitamin and the protective antioxidant. *Food Chemistry*, *196*, 466-474. https://doi.org/10.1016/j.foodchem.2015.09.071
53. Wang, S., Ye, X., Sun, Y., Liang, J., Yue, P., & Gao, X. (2021). Nanocomplexes derived from chitosan and whey protein isolate enhance the thermal stability and slow the release of anthocyanins in simulated digestion and prepared instant coffee. *Food Chemistry*, *336*, 127707. https://doi.org/10.1016/j.foodchem.2020.127707
54. Xu, W., Lv, K., Mu, W., Zhou, S., & Yang, Y. (2021). Encapsulation of α-tocopherol in whey protein isolate/chitosan particles using oil-in-water emulsion with optimal stability and bioaccessibility. *LWT*, *148*, 111724. https://doi.org/10.1016/j.lwt.2021.111724
55. Xu, W., Tang, Y., Yang, Y., Wang, G., & Zhou, S. (2020). Establishment of a stable complex formed from whey protein isolate and chitosan and its stability under environmental stresses. *International Journal of Biological Macromolecules*, *165*, 2823-2833. https://doi.org/10.1016/j.ijbiomac.2020.10.130
56. Yousefi, S., Rajaei, P., Nateghi, L., Nodeh, H.R., & Rashidi, L. (2023). Encapsulation of sesamol and retinol using alginate and chitosan-coated W/O/W multiple emulsions containing Tween 80 and Span 80. *International Journal of Biological Macromolecules*, *242*, 124766. https://doi.org/10.1016/j.ijbiomac.2023.124766

نشریه پژوهشهای علوم و صنایع غذایی ایران
https://ifstrj.um.ac.ir

مقاله پژوهشی

جلد ۲۲، شماره ۱، فروردین- اردیبهشت ۱۴۰۵، ص. ۳۴-۱۷

کپسوله‌سازی ویتامین‌های E و C با استفاده از کنسانتره پروتئین آب‌پنیر-کیتوزان از طریق خشک‌کردن پاششی: بهینه‌سازی، ویژگی‌های فیزیکوشیمیایی و تحلیل نرخ رهایش

پونه مجتهدی[۱]- مهدی وریدی[۱*]- امیرمحمد مرتضویان[۲]

تاریخ دریافت: ۱۴۰۴/۰۷/۳۰
تاریخ پذیرش: ۱۴۰۴/۱۱/۰۸

چکیده

نگرانی‌های فزاینده در مورد سلامت انسان، تقاضا برای ترکیبات زیست‌فعال عملکردی مانند ویتامین‌های E و C را افزایش داده است. با این حال، ناپایداری ذاتی و حساسیت آنها به شرایط محیطی، کاربرد مستقیم آنها را در سیستم‌های غذایی محدود کرده است. استراتژی‌های کپسوله‌سازی، رویکردی مؤثر برای محافظت از این ویتامین‌ها در برابر تخریب و آزادسازی کنترل‌شده ارائه می‌دهند. این مطالعه با هدف توسعه یک سیستم کپسوله‌سازی پایدار برای ویتامین‌های E و C با استفاده از کمپلکس کنسانتره پروتئین آب پنیر-کیتوزان (WPC-CS) از طریق خشک کردن پاششی انجام شد که امکان تحویل همزمان ویتامین‌های آب‌دوست و لیپوفیل را فراهم می‌کند. روش‌شناسی سطح پاسخ (RSM) برای بهینه‌سازی فرمولاسیون با هدف به حداکثر رساندن راندمان کپسوله‌سازی و در عین حال به حداقل رساندن میزان رطوبت و رطوبت‌پذیری به کار گرفته شد. میکروکپسول بهینه، راندمان کپسوله‌سازی ۹۳.۸۳٪ و ۹۳.۶٪ را برای ویتامین‌های E و C به‌ترتیب با رطوبت کم (۳.۱۷٪) و رطوبت‌پذیری کنترل‌شده (۱۳.۱۸٪) نشان داد. مطالعات آزادسازی، رفتار وابسته به فاز را نشان دادند: ویتامین C آزادسازی ناچیزی در مایع شبیه‌سازی شده دهان (۰.۰۷٪ پس از ۱۰ دقیقه) نشان داد، اما آزادسازی تجمعی قابل توجهی در مایع شبیه‌سازی شده معده (۸۵.۷۴٪ پس از ۱۲۰ دقیقه) داشت. در مقابل، ویتامین E پروفایل آزادسازی آهسته‌تر و پایدارتری را نشان داد، به‌طوری‌که ۳۱.۴۵٪ پس از ۱۲۰ دقیقه در مایع شبیه‌سازی شده معده آزاد شد و هیچ آزادسازی قابل تشخیصی در شرایط دهان مشاهده نشد. آنالیز میکروسکوپ الکترونی روبشی (SEM) نشان داد که میکروکپسول‌ها دارای مورفولوژی عمدتاً کروی با سطح چروکیده هستند که مشخصه ماتریس‌های پروتئین-پلی‌ساکارید تشکیل شده در طول خشک کردن سریع است. طیف‌سنجی مادون قرمز تبدیل فوریه (FTIR) کپسوله کردن موفقیت‌آمیز هر دو ویتامین را از طریق برهمکنش‌های بین WPC و CS تأیید کرد. این یافته‌ها نشان می‌دهد که کمپلکس‌های WPC-CS تولید شده از طریق خشک کردن پاششی، یک سیستم کپسوله کردن همزمان و آزادسازی کنترل‌شده مؤثر را فراهم می‌کنند که پایداری ویتامین را افزایش داده و کاربردهای امیدوارکننده‌ای را در غذاهای کاربردی و فرمولاسیون‌های دارویی ارائه می‌دهند.

واژه‌های کلیدی: بهینه‌سازی، خشک‌کن پاششی، رهایش کنترل‌شده، میکروکپسول

۱- گروه علوم و صنایع غذایی، دانشکده کشاورزی، دانشگاه فردوسی مشهد، مشهد، ایران
(*- نویسنده مسئول: Email: m.varidi@um.ac.ir)
۲- گروه علوم و صنایع غذایی، دانشکده علوم تغذیه و صنایع غذایی، انستیتو تحقیقات تغذیه‌ای و صنایع غذایی کشور، دانشگاه علوم پزشکی شهید بهشتی، تهران، ایران
https://doi.org/10.22067/ifstrj.2026.96109.1497