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ORIGINAL PAPER

Enhancing Cream Cheese Nutritional Value and Shelf Life with the Incorporation of Free and Microencapsulated Onion Peel Extract

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Abstract

Onion production generates a substantial amount of waste rich in bioactive compounds with noteworthy biological properties. This study aimed to extract bioactive compounds from onion peels (OP) using a Soxhlet apparatus in a solid-liquid extraction. The extracted compounds were then encapsulated by applying the double emulsion technique and using ethyl cellulose (EC) as an encapsulating agent, followed by their incorporation into a cream cheese to extend its shelf life. Antioxidant capacity was determined using ABTS and DPPH assays, while total phenolic content (TPC) was quantified by the *Folin-Ciocalteu* method. The extract demonstrated strong antioxidant activity by scavenging ABTS and DPPH radicals, and the value of TPC was 396.7 $mg_{GAE} \cdot g_{extract}^{-1}$. Additionally, the extract inhibited the activity of α-amylase and β-glucosidase by 91.9 and 95.8%, respectively, demonstrating potential anti-diabetic properties. The microencapsulated OP extract revealed a production yield of 73.9% and an encapsulation efficiency of 71.2%. The microparticles exhibited a round-shaped morphology, with size variation indicated by a polydispersity index of 1.38 and showed greater thermal stability than the free OP extract. Evaluation of the extract and microparticles in cream cheese fortification demonstrated that the samples exhibited reduced syneresis, maintained stable moisture content, and offered extended protection against lipid oxidation compared to the negative control. In addition, both fortified samples also had higher TPC and increased antioxidant capacity. Therefore, incorporating free and microencapsulated OP extract into cream cheese offers a sustainable approach to valorising onion waste, enhancing product quality while promoting circular economy principles.

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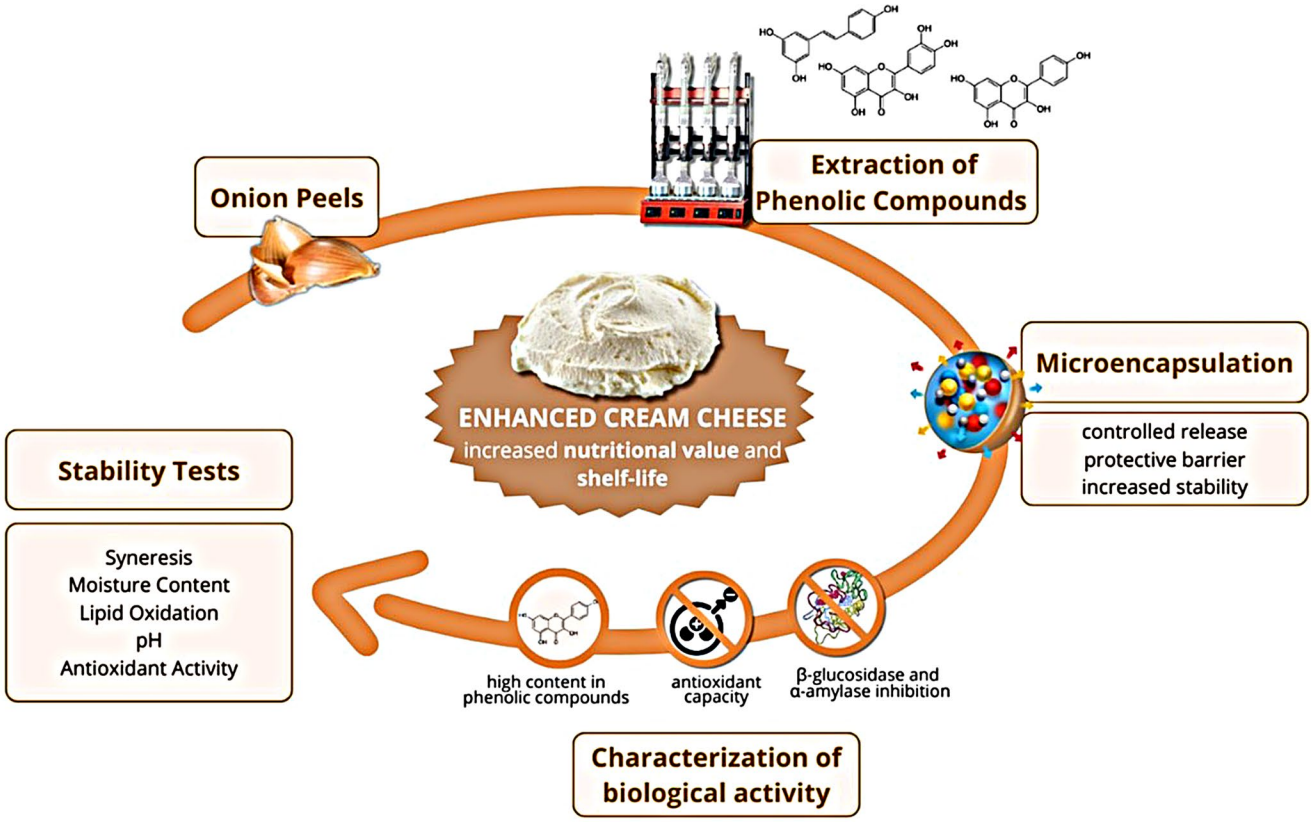
Graphical Abstract



Keywords Onion peels · Antioxidants · Cream Cheese · Microencapsulation · Shelf life · Circular economy

Introduction

Onions are the world's second most-produced vegetable crop, with an annual production of approximately 107 million tons. The industrial processing of onions generates a significant amount of waste, primarily in the form of peels. About 38% of the weight of processed onions is considered inedible and is disposed of as waste. Additionally, around 10% of harvested onions are deemed unfit for marketing and do not meet quality standards [1]. Consequently, more than 550 thousand tons of waste are generated by the onion production industry, making it crucial to properly utilise and dispose of these wastes to minimise their environmental impact [2]. Fortunately, onion by-products are rich in bioactive compounds (BACs), making them suitable for repurposing in other food products.

OP (onion peels) are particularly rich in phenolic compounds (PCs) known for their health benefits, representing the second most abundant group of secondary metabolites in plants [3]. Literature highlights the presence of flavonoids (quercetin, kaempferol and catechin), anthocyanins, tannins, and phenolic acids (vanillic and ferulic acid). It has been shown that, when compared to the bulb, OP exhibited higher concentrations of phenolic and flavonoids [4]. The main PCs present in onion are quercetin and resveratrol [5, 6]. These compounds have been gaining attention due to their potent antioxidant capacity that allows them to neutralise free radicals, reduce oxidative stress, and protect against cellular damage. This antioxidant potential leads to an ability to extend the shelf life of food products [7, 8]. Moreover, they have shown positive impacts on human health, mainly in the prevention of cardiovascular diseases and protection against chronic diseases, such as liver disease, obesity, diabetes, Alzheimer's, and Parkinson's. They have demonstrated anti-cancer effects by inhibiting cancer cell growth and promoting apoptosis [9, 10]. Moreover, PCs have also been gaining attention due to their potential anti-diabetic properties through the inhibition of enzymes, such as α-amylase and β-glucosidase. There are several mechanisms to inhibit these enzymes, for example, interactions of the PCs with the active site of the enzyme or competitive interaction between the PCs and the substrate [11]. In addition to their rich composition in BACs, OP are an easy-to-obtain by-product and require less pre-treatment

in comparison to other by-products, making them a more appealing raw material. Therefore, OP are an excellent source of PCs and can be used to extract them to incorporate in different applications.

Apart from constituting a great source of pollution, this agro-industrial by-product also represents an economic risk. However, utilising PCs extracted from OP addresses waste management concerns while providing a natural way to extend the shelf life of food products and enhance their nutritional value, while promoting a circular economy model. The integration of PCs into foods aligns with the trends of the functional food industry, which has experienced rapid growth in recent years and is expected to increase by 128 billion dollars by 2028 [1, 12]. Research has shown that PCs extracted from agri-food by-products have the potential to act as natural food preservatives [13, 14]. There has been growing interest in incorporating natural extracts into foods to enhance their nutritional value and as a strategy to replace commercial preservatives. Given the rich composition of OP in PCs and their biological properties, the opportunity to use this by-product to obtain phenolic-rich extracts to incorporate into foods arises.

Despite the characteristics that allow PCs to be so interesting, these are also responsible for their limitations, since most natural antioxidants are prone to oxidation, resulting in a loss of their bioactive properties. They can be easily degraded due to environmental factors such as light, oxygen, temperature, pH, and enzymatic activities. Their application to foods is limited as they can lead to unpleasant tastes and changes in product colour [15]. Microencapsulation is a commonly applied strategy to overcome this obstacle as it stabilises antioxidants, protects them from external factors, ensures stability, enhances their bioavailability, masks undesirable flavours and colour changes, and controls the release of substances into the matrix while preserving their beneficial properties. Literature reports that the food industry is currently leveraging this method to encapsulate various bioactive components, including flavours and vitamins [15, 16].

Cream cheese is a widely consumed dairy due to its high protein, fat, calcium, and vitamin content. Nonetheless, its high moisture content and pH may lead to the growth of pathogenic and spoilage microorganisms. Moreover, given its high-fat content, it is prone to oxidation, resulting in unappealing odours and flavours and a decrease in nutritional value. These have negative repercussions regarding the preparation, packaging, and sale of this type of product. The shelf life of cream cheese relies on its ability to resist oxidation and spoilage, which can be prevented using commercial preservatives; however, some of these may be harmful to consumers or may decrease the nutritional value of the final product [17, 18]. As an alternative, PCs from by-products, such as OP, can be used as preservatives, aligning with the latest food industry trends. These extracts can be used as food preservatives and nutritional supplements, offering benefits to human health, and can be incorporated in free and microencapsulated form. The research on cheese fortification through the incorporation of phenolic extracts and extracts rich in antioxidants has been increasing, as observed in the literature, revealing the relevance of this topic [16, 19, 20]. Most studies focus on the fortification of ripped cheese (e.g. cheddar) and the use of phenolic extracts from plants as fortification agents. Overall, it is noticeable that the incorporation of antioxidants of natural origin increases the antioxidant properties of the cheese and the phenolic content. Nonetheless, these extracts influence the physical characteristics of the product, mainly viscosity and colour, enhancing the need for microencapsulation [21–23]. The literature on fortifying cheese with microparticles loaded with phenolic-rich extracts is limited, highlighting the need for further research in this area [24].

To the best of the authors' knowledge, no prior research has explored the fortification of cream cheese with free and encapsulated phenolic-rich extracts from onion peels, highlighting the novelty of this study. Therefore, the current research is designed to investigate the antioxidant and antimicrobial properties of onion peel (*Allium cepa* L.) extracts, evaluate the effectiveness of their microencapsulation, and examine how the addition of both free and microencapsulated extracts affects the sensory, physicochemical, antioxidant, and textural qualities of cream cheese throughout its shelf life.

Materials and Methods

Samples and Reagents

Onion peels (OP) were collected from Braga, Portugal. The solvents used in this study ethanol, ethyl acetate, and hydrochloric acid were acquired from VWR (Radnor, PA, USA), while chloroform and methanol were purchased from Carlo Erba (Barcelona, Spain), and acetic acid and isooctane were acquired from Sigma Aldrich (St. Louis, MO, USA). The *Folin–Ciocalteu* reagent, the 2,2-diphenyl-1-picrylhydrazyl (DPPH), and the 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) were purchased from Sigma-Aldrich (St. Louis, MO, USA) and sodium carbonate from Merck (Darmstadt, Germany). For the microencapsulation, ethyl cellulose and poly(vinyl) alcohol were purchased from Sigma Aldrich (St. Louis, MO, USA). For the lipid oxidation tests, barium chloride dihydrate, iron chloride III, iron sulfate (II) heptahydrate and *p*-anisidine were obtained from Sigma Aldrich (St. Louis, MO, USA), and ammonium

thiocyanate was acquired from Alfa Aesar (Haverhill, MA, USA).

Methods

Extraction of the Phenolic Compounds from the Onion Peels

The phenolic-rich extract from OP was obtained through solid-liquid extraction utilising a Soxhlet apparatus. The OP were first dried in an oven at 65 °C for 24 h and ground to a diameter of 50 mesh. Ethanol was used as the extraction solvent, due to its low polarity, relatively low boiling point, and GRAS (Generally Recognised as Safe) status, making it suitable for food applications. The mass-to-volume ratio of the sample to solvent was 1:20 (m/V), with an extraction temperature of 70 °C for 2 h. The solvent was then removed using a rotary evaporator and completely evaporated with a nitrogen stream.

Characterisation of the Onion Peel Extract

The Total Phenolic Content (TPC) of the extract was determined according to the literature (Ferreira & Santos, 2023). In a 2 mL cuvette, 20 µL of the sample solution (1 $g{\cdot}L^{-1}$ of extract in ethanol), 100 µL of the *Folin-Ciocalteu* reagent, and 1580 µL of distilled water were added. Posteriorly, 300 µL of the sodium carbonate solution (333.3 $g{\cdot}L^{-1}$) were added. The mixture was incubated for 2 h in a dark environment at room temperature. After that, the absorbance was measured at 750 nm using a spectrophotometer (UV-6300PC, VWR). Using a calibration curve, a correlation between gallic acid and absorbance was used to determine the milligrams of gallic acid equivalents (GAE) per gram of onion peel extract (OPE). ($Abs = 0.0748 \times C_{gallic\ acid}$) [25].

In order to determine the OPE's antioxidant capacity, two methods with different radicals were employed: an assay with 2,2-diphenyl-1-picrylhydrazyl (DPPH) and an assay with 2,2-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS). This assessment was performed according to protocols in the literature [25]. The results for these assays were presented in two forms, as IC_{50} and as Trolox Equivalent Antioxidant Capacity (TEAC).

The antidiabetic properties were evaluated using inhibitory assays of enzyme activity of a-amylase and b-glucosidase. The a-amylase assay was conducted as described in previous studies, with some modifications [26, 27]. To determine the effects of the extract on inhibiting β-glucosidase, a Sigma-Aldrich β-glucosidase Activity Assay Kit was utilised. The activity of β-glucosidase was determined according to Eq. 1. The percentage of inhibition from the enzyme activity was determined through the difference between the enzyme activity without the inhibitor and the enzyme activity with the inhibitory agent.

$$\beta\text{-glucosidase activity (Units/L)} = \frac{Abs_{final} - Abs_{initial}}{Abs_{calibrator} - Abs_{water}} \quad (1)$$

Microencapsulation Using the Double Emulsion Technique

The OP extract microparticles were produced using the double emulsion technique according to the literature [28]. This technique was selected as it can be used to encapsulate both hydrophilic and hydrophobic material, offering high encapsulation efficiency. Ethyl cellulose was selected as the encapsulating agent since it is a non-toxic and cheap material, insoluble in water, extremely stable, and it is odourless and tasteless [29, 30]. It is also accepted as a food additive in different sectors and has GRAS status [31]. Initially, the oil phase was prepared by mixing 200 mg of EC with 10 mL of ethyl acetate with the use of a vortex for 15 min. Afterwards, the extract was dissolved in ultrapure water (UPW) with a theoretical loading of 15% to create the first aqueous phase (w_1). To form a stable primary emulsion (w_1/o), 1 mL of the w_1 mixture was added to the o phase and mixed for 5 min using a vortex. The primary emulsion was hydrophilically emulsified in a room temperature solution of PVA (poly(vinyl alcohol)) (1% w/V) (w_2), previously heated and stirred at 120 °C until fully dissolved, resulting in the formation of a $w_1/o/w_2$ double emulsion. The double emulsion was homogenised using a high-performance homogeniser (IKA T18 Digital ULTRA-TURRAX) for 5 min at a rotation speed of 3000 rpm. The $w_1/o/w_2$ double emulsion was stirred at 700 rpm on a stirring plate within a fume hood for 3 h at room temperature to remove the solvent and solidify the microparticles. Afterwards, the microparticles were filtered through a 0.2 µm filter and washed with UPW to eliminate any PVA residue. Finally, the microparticles were frozen at -20 °C for 24 h before being freeze-dried in a VirTis freeze-dryer at -90.2 °C for an additional 24 h.

Characterisation of the Microparticles

Determination of the Production Yield and the Onion Peel Extract Encapsulation Efficiency The total amount of microparticles recovered can be determined through the production yield, in Eq. 2.

$$PY\ (\%) = \frac{m_{MP}}{m_I + m_{polymer}} \times 100 \quad (2)$$

where m_{MP} is the mass of the freeze-dried OPE-loaded microparticles, m_I is the mass of the OPE initially added for

the encapsulation and $m_{Polymer}$ is the mass of the polymer used.

To determine the amount of encapsulated OPE, the microparticles were disrupted, and the TPC was measured using the *Folin-Ciocalteu* method previously described. The disruption involved adding 10 mg of microparticles to a 4 mL mixture of n-hexane and ethanol (1:1) and placing it in a 35 °C water bath for 20 min. This was followed by vortexing for 1 min and sonicating for 5 min, repeated 3 times. The mixture was then centrifuged (Rotofix 32 A, Hettich) and filtered before evaporating the solvents with a nitrogen stream. The remaining content was diluted in ethanol and the TPC was measured. The OPE encapsulation efficiency (EE) was calculated using Eq. 3.

$$EE\ (\%) = \frac{m_{BAC,encaps}}{m_I} \times 100 \quad (3)$$

where $m_{BAC,encaps}$ corresponds to the mass of OPE used for the microencapsulation.

Morphological Analysis and Particle Size Distribution The microparticles' external morphology was assessed using scanning electron microscopy (SEM). The desired morphological characteristics were analysed using different levels of magnification (2000x and 200x) using an accelerating voltage of 40 keV. Additionally, the microparticles' size distribution was assessed using the Coulter Counter-LS 230 Particle Size Analyzer (Miami, FL, USA). The results obtained were calculated using the average values, and Eq. 4 was employed to calculate the polydispersity index (PDI).

$$PDI = \left(\frac{D_{v,90} - D_{v,10}}{D_{v,50}}\right) \quad (4)$$

where $D_{v,90}$ represents the maximum diameter that contains 90% of the sample volume, $D_{v,10}$ indicates the maximum diameter that contains 10% of the sample volume, and $D_{v,50}$ displays the maximum diameter containing 50% of the sample volume.

Thermal Stability of the Microparticles To analyse the thermal properties of the microparticles, a Hitachi STA300 instrument was used to perform thermogravimetric analysis (TGA). Samples weighing 10 mg were heated in an inert environment, with temperatures ranging from 20 to 600 °C at a heating rate of 10 °C·min^{-1}.

Fourier-Transform Infrared Spectroscopy The Fourier-transform Infrared Spectroscopy (FTIR) spectra were collected using an FTIR-ATR spectrometer (Spectrum Two, Perkin Elmer, Waltham, Massachusetts, USA). The samples were measured in attenuated total reflectance (ATR) mode with a Platinum ATR (A225/Q) with a diamond crystal with a single reflection accessory. The infrared spectra were captured by scanning from 4000 to 400 cm^{-1}, with a resolution of 4 cm^{-1}.

Table 1 Different additives used in the production of the three cream cheeses

Additive (g·L$_{milk}^{-1}$)	Cheese 1 (NC)	Cheese 2	Cheese 3
OP Extract	-	1.0	-
Microencapsulated OP Extract	-	-	1.0

NC: Negative Control; OP: Onion Peel; - no additive

Cream Cheese Production

The cream cheese was produced using 1 L of UHT semi-skimmed milk, which was heated to 90 °C. The choice of milk was guided by the requirement to use pasteurised milk for this type of cheese production. Additionally, semi-skimmed milk was selected to lower the initial fat content of the cheese, thereby minimising the risk of lipid oxidation and enhancing the product's stability. Three different cream cheeses were produced with slight differences in the additives as described in Table 1. The amount of each additive was added to the respective formulation and mixed to ensure homogeneity. Then 40 mL of previously squeezed lemon juice were added to lower the pH of the milk, leading to the destabilisation and aggregation of casein proteins to achieve coagulation. After resting for 30 min, the mixture was filtered and blended. To achieve the desired consistency and flavour, 45 mL of whey and 2.5 g of salt were added. Subsequently, the obtained product was placed in a recipient and stored in a fridge at 4 °C. Stability tests for each sample were performed at three different time points: t_0, t_1 and t_2, for two weeks.

Physico-Chemical Characterisation of the Cream Cheese Samples

The determination of the pH value of each sample at each timepoint was performed by adding 9 mL of water to 1 g of sample into a beaker. An agitation plate was used to homogenise the mixture. The pH was measured using a digital pH meter (XS pH 50+).

In order to assess syneresis, 2 g of each sample were weighed and placed in a centrifuge at 1510 G for 20 min, after which the weight of the supernatant was measured. The percentage of syneresis can be calculated using Eq. 5.

$$Syneresis\ (\%) = \frac{m_{supernatant}}{m_{cheese}} \times\ 100 \tag{5}$$

where $m_{supernatant}$ and m_{cheese} correspond to the mass of the supernatant and the mass of the cheese used, respectively.

The moisture content was determined by weighing 2 g of each sample and drying them at 105 ºC in an oven until constant weight. The total water content of the sample was then calculated using Eq. 6.

$$Moisture\ Content\ (\%) = \frac{m_{initial\ cheese} - m_{final\ cheese}}{m_{initial\ cheese}} \times\ 100 \tag{6}$$

where $m_{final\ cheese}$ and $m_{initial\ cheese}$ correspond to the mass of the cheese before and after total drying, respectively.

Finally, the lipid oxidation of the samples was determined. For this, 2 g of the cheese samples were weighed and placed in an oven at 50 °C for 24 h. The supernatant was removed to perform the oxidation tests. The primary oxidation of the samples was determined by the peroxide value (PV), and the secondary oxidation was determined by the *p*-anisidine value (*p*-AV), following the literature protocols described in the Supplementary Information [17]. Lastly, the total oxidation value (Totox), was determined by using Eq. 7.

$$Totox = \ 2\mathrm{PV} + p - \mathrm{AV} \tag{7}$$

Evaluation of the Antioxidant Properties of the Cream Cheese Samples

In order to evaluate the antioxidant capacity and determine the TPC of each cream cheese sample, PCs were extracted combining 2 g of each sample with 4 mL of the ethanol. The mixture was vortexed (VWR VV3) for 1 min, and placed in an ultrasonic bath (VWR, United States) for 5 min, repeating this process twice. Afterwards, the mixture was centrifuged (Rotofix 32 A Centrifuge, Hettich, Germany) at 1510 G for 10 min. The supernatant was collected to a different tube. Additionally, 4 mL of ethanol were added to the initial tube and the process was repeated. The supernatant obtained was combined with the previously collected one and centrifuged once more. The resulting solution was stored in the dark at 4 °C for further analysis of TPC and antioxidant capacity assays, performed according to the methods previously described.

Table 2 Characterisation of the extract obtained from onion peels regarding TPC, antioxidant capacity, and enzyme Inhibition

TPC ($mg_{GAE}{\cdot}g_{extract}^{-1}$)		396.7±4.2
ABTS	IC_{50} ($mg{\cdot}L^{-1}$)	3.62±0.16
	TEAC ($mg_{trolox}{\cdot}g_{extract}^{-1}$)	551.8±31.1
DPPH	IC_{50} ($mg{\cdot}L^{-1}$)	36.1±1.7
	TEAC ($mg_{trolox}{\cdot}g_{extract}^{-1}$)	33.4±9.5
Enzyme Inhibition (%)	a-amylase	91.9±1.2
	b-glucosidase	95.8±4.2

GAE: gallic acid equivalents; IC_{50}: concentration of extract needed to inhibit 50% of the free radicals; TE: Trolox equivalent; TEAC: Trolox equivalent antioxidant capacity; TPC: total phenolic content. The results are expressed as means±standard deviations of three independent measurements obtained from the same sample

Statistical Analysis

In order to identify any significant differences in the findings, a two-way ANOVA was performed with the Tukey multiple comparisons test using *GraphPad*. Results with p-values above 0.05 (indicating a 95% confidence interval) suggest that there is no significant difference between the groups being compared.

Results and Discussion

Onion Peel Extract Characterisation

The PCs were extracted from OP using a solid-liquid extraction method with a Soxhlet apparatus, achieving an average extraction yield of 10.9±2.2%. Other studies present in the literature reported extraction yields of 4.5% and 8.3% for onion peel using 70% ethanol (V/V) and hot water extraction, respectively [32]. In another study, the phenolic-rich extract from onion waste (including peels and bulbs) was obtained using a mixture of ethanol: water (50:50 V/V), achieving yields of 20% [33]. The achieved value is within the literature range for extractions of these compounds using similar conditions. Various factors, including extraction technique, solvent, extraction time, and temperature, among others, can influence extraction yield and may account for the differences in obtained values. Afterwards, the phenolic-rich extract was characterised in terms of its antioxidant and anti-diabetic properties, and TPC, with the results presented in Table 2.

The OPE revealed a TPC mean value of 396.7 $mg_{GAE}{\cdot}g^{-1}$, which aligns with the range of 377–585 $mg_{GAE}{\cdot}g^{-1}$, observed in previous studies performed by the authors, and tends towards the higher end [25, 34]. A study with a focus on the characterisation of extracts from red onion skin demonstrated a TPC of 384 $mg_{GAE}{\cdot}g^{-1}$ [35]. In another study, different extracts were obtained from red onion skin, using different solvents, and results revealed that the TPC was in the range of 64–208 $mg_{GAE}{\cdot}g^{-1}$, with the extracts obtained with ethanol exhibiting superior values [36]. Moreover, a study of extracts obtained from 15 different onion cultivars demonstrated a TPC in the range 15–289 $mg_{GAE}{\cdot}g^{-1}$ [4]. This result indicates that the extract displays high phenolic content. Indeed, in another study by the authors, it was

observed through HPLC analysis that the OPE exhibited a variety of PCs, including quercetin, resveratrol, epicatechin, and catechin, among others, contributing to the high TPC value. Regarding the differences in the literature, these can result from the aforementioned reasons.

For both the DPPH and ABTS methods, the IC_{50} (required concentration of extract to inhibit 50% of a radical) and TEAC (expressed in Trolox Equivalents (TE) per gram of dried extract) values were measured to determine the antioxidant capacity and to enable a better comparison with the literature. A lower IC_{50} value implies a stronger antioxidant capacity, meaning less extract is needed to achieve the same effect. Considering the values obtained for the IC_{50}, one can note that the concentration of OPE needed to inhibit ABTS and DPPH radicals is considerably reduced. The achieved values of IC_{50} for the assays with the ABTS and DPPH radicals were, respectively, 3.6 and 36 $mg{\cdot}L^{-1}$; these values are inferior to 50 $mg{\cdot}L^{-1}$, which proves that the extract can be considered a very strong antioxidant [37]. Regarding DPPH inhibition, the literature reveals that the IC_{50} values are in the range of 16–31 $mg{\cdot}L^{-1}$, demonstrating that the achieved value is close to the ones in the literature [25, 29, 34]. Considering the ABTS radical, the literature is slightly more scarce regarding IC_{50} values; however, an IC_{50} of 13.6 $mg{\cdot}L^{-1}$ has been reported, demonstrating that the achieved value is superior to the literature one [29]. Considering the TEAC, it can be observed that the extract exhibits superior values towards the ABTS radical than towards the DPPH radical. Indeed, different studies have demonstrated that TEAC of OP extract towards the ABTS radical can be in the range of 6-413 $mg_{trolox}{\cdot}g_{extract}^{-1}$, while for the DPPH radical, the values are within 2-330 $mg_{trolox}{\cdot}g_{extract}^{-1}$, demonstrating that the extract is more efficient in the inhibition of ABTS [25, 29, 34, 38]. The discrepancies between the values obtained for TPC and antioxidant capacity and those reported in other studies may be attributed to several factors, such as differences in extraction methods or parameters, variations in onion variety and growth conditions. Even though a better result was obtained for the ABTS assay, the results underline the extract's great antioxidant capacity towards both radicals.

The α-amylase and β-glucosidase assays provide information on the ability of the extract to inhibit these enzymes, which are responsible for breaking down carbohydrates. The inhibition of these two enzymes is particularly useful in managing diseases related to the breakdown of glycosidic bonds, such as diabetes. By slowing down their activity, inhibitors can aid in controlling these conditions [10]. The results revealed that the extract could inhibit both enzymes, being slightly more effective towards β-glucosidase. Upon comparison with the literature, it was found that the OPE in the study exhibited superior values to other studies (60–65%) [39, 40]. To the authors' knowledge, no previous research has been conducted on the effects of OPE on inhibiting the β-glucosidase. However, the results demonstrate a significant outcome. The high values of inhibition could be due to the presence of flavonoids, which are known to possess α-amylase and glucosidase inhibitory properties [41]. The presence of these compounds in the extract composition corroborates the fact that flavonoids display anti-diabetic properties.

The results indicate that OPE may function as an antioxidant agent and has the potential to inhibit α-amylase and β-glucosidase, making it a promising anti-diabetic agent. These biological properties make OPE a valuable ingredient for enhancing the nutritional value of foods and creating value-added products.

Characterisation of the Microencapsulation of the OPE

Microencapsulation was performed to improve the stability and performance of the OPE. The double emulsion solvent evaporation technique was selected due to being commonly applied to hydrophilic molecules, since their release in external aqueous phases in single emulsions is considerably swift, leading to a reduced encapsulation efficiency [42]. The microparticles containing extract were produced using EC as core material since this is a weakly polar and biocompatible polymer, making it suitable for the desired application in the present study. A major benefit of microencapsulation is the controlled release of the BACs, in this case OP extract, from the particle to the medium, meaning that the release can be adjusted over time, instead of being immediate, which leads to the degradation of the compounds. The sustained release of the compounds extends their biological properties during storage. To characterise the microencapsulation technique, the process's production yield (PY) and encapsulation efficiency (EE) were evaluated, resulting in values of 73.9 and 71.2%, respectively. These two parameters are crucial in assessing the microencapsulation. While the production yield of the process is important to assess its cost-effectiveness, the EE indicates how effectively the core material is encapsulated within the microparticles. In this instance, it shows that 73.9% of the EC and OPE used in microencapsulation was retained in the resulting microparticles, revealing a high value of PY. The achieved result is within the literature range, for this encapsulation methodology, of 67–85% [43]. One of the reasons for being in the lower range can be associated with the loading content, since the amount of EC present might be insufficient to encapsulate the extract. The EE revealed a promising result, with approximately 71% of the extract successfully encapsulated. Literature studies report the use

of a single emulsion to encapsulate OPE, achieving EE values in the range of 82–93% [14, 44]. A study employing the same technique to encapsulate hydroxytyrosol revealed EE within 79–88% [28]. The achieved results are slightly lower than the reported ones. As previously mentioned, the loading content might be the cause of these values, since an increase in the loading content results in a decrease in the EE [45]. This can be the result of the significant diffusion of the PCs (due to an increase in the loading) to the external aqueous phase, as a result of higher flux at high phenolic content during the development of the MP [46, 47].

The microparticles were also characterised by their morphology. Therefore, a morphological analysis was performed by SEM, which allowed to obtain the photomicrographs present in Fig. 1.

Analysing Fig. 1, it is evident that the produced microparticles (A) exhibit irregular sizes and a round shape with some pores. The literature states that freeze-drying can affect particle size as a result of the low temperatures used in this method and the absence of forces to break the freeze-dried liquid into particles. The presence of pores on the surface of the microparticles seems to indicate that the controlled release of the content inside the microparticles might occur through diffusion [48, 49]. Additionally, in sample A, some microparticles appear to have initiated their release process, showing a deformed shape. These observations are consistent with literature findings using the same microencapsulation technique, which reported microparticles with a smooth surface and low porosity [28].

Apart from morphological analysis, the particle size distribution was also assessed, and the result obtained is presented in Figure S.1, in the Supplementary Information, with an average size of 18.73 ± 9.55 µm. In another study by the authors, molecular inclusion was applied to encapsulate OPE, where the mean diameter was between 16 and 32 µm [34]. The result is within the desired range, since it is possible to prevent the development of nanoparticles, as they can infiltrate the bloodstream. The mean particle size can be influenced by various factors, such as type and concentration of the core material, homogenisation velocity, and the polymer's molecular weight [50]. The polydispersity index (PDI) was determined, resulting in a value of 1.38, indicating significant heterogeneity in the particle size distribution; the sample is considered polydisperse according to ISO 22412:2017. Results indicate that aggregates might have been formed during the production of the microparticles. However, Figure S.1. shows the PDI as a monomodal curve. These results align with those reported in the authors' previous study, using the same homogenisation parameters of a rotation speed of 3000 rpm for 5 min, in which the particle size distribution exhibited similar behaviour, with a mean particle size of 16.23 µm and a higher PDI of 2.25 [34].

The Fourier Transform Infrared Spectroscopy (FTIR-ATR) was used to assess the inclusion of the OPE in the polymeric systems and to confirm the absence of specific functional groups of the extract in the MP. Figure 2(A) displays the FTIR spectra of the OPE, the microencapsulated extract and EC.

The FTIR analysis is of great importance since it allows to know the structure of a sample. Analysing the spectrum obtained for the OPE, there are peaks at 3300 cm^{-1} and 1645 cm^{-1} resulting from stretching of O-H and C=C bonds present in its structure. At 1050 cm^{-1} and 2950 cm^{-1} there are other peaks characteristic of an O-C stretching of an anhydroglucose ring and of C-H bonds, respectively. It is noticeable that at 3300 cm^{-1}, the peak of the OPE has greater intensity, probably because of its higher moisture content, which evidences many O-H bonds. This greater intensity can also be due to the content of PCs in the OPE. The absence or decrease in intensity of this peak in the microparticles spectrum can indicate that the PCs are encapsulated within the coating material, EC. Also, the resemblance of the EC and microparticle spectra further indicated that the extract was successfully microencapsulated.

Results for the thermal stability of the microparticles, along with results of the thermal stability of the OPE and EC are presented in Fig. 2(B) and Table S.1., in the

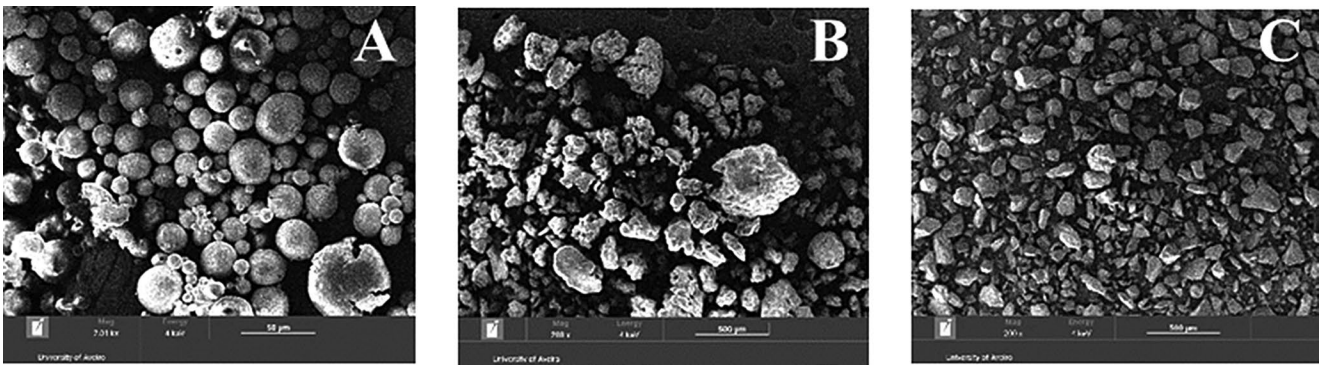


Fig. 1 Microphotographs of the OPE microparticles (**A**), Ethyl Cellulose (**B**) and PVA (**C**) analysed by scanning electron microscopy (SEM)

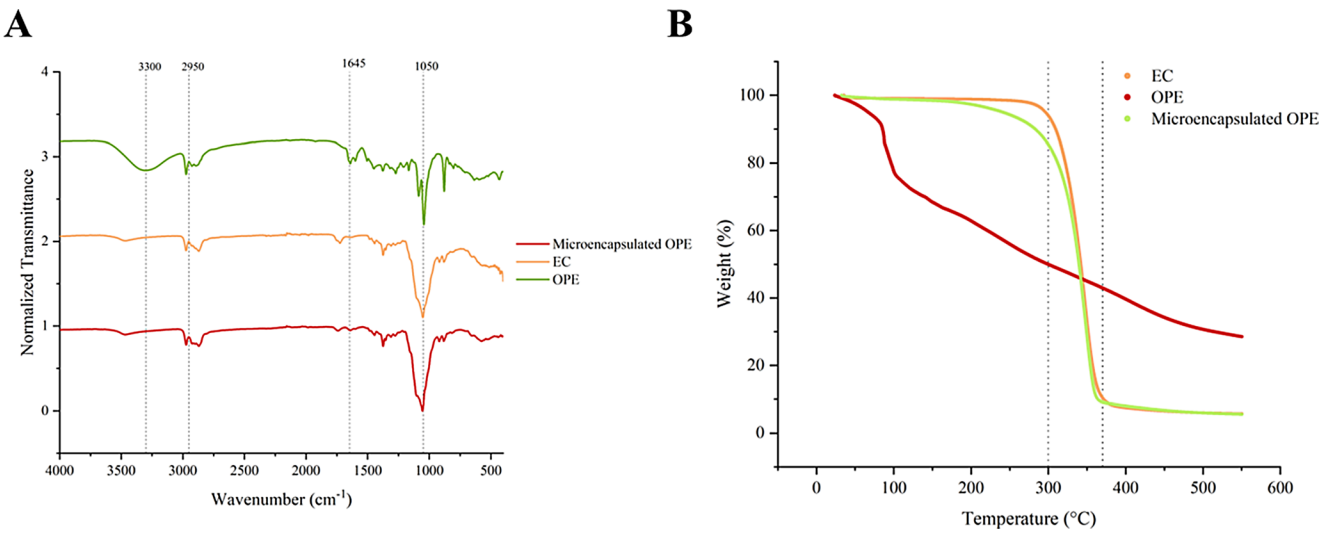


Fig. 2 (**A**) FTIR spectra of the unloaded microparticles, the microencapsulated OPE, ethyl cellulose and the OPE; (**B**) Thermogravimetric analysis (TGA) curves for ethyl cellulose, OPE and microencapsulated OPE

Supplementary Information. The TGA results for EC and microencapsulated OPE revealed three distinct thermal events where the mass loss occurred, showing similar thermal behaviour, contrary to the OPE sample. At lower temperatures, initial weight loss is due to the evaporation of water and volatile compounds, with OPE losing weight up to about 105 °C. Being a dry powder, EC showed minimal weight change in this range [51]. During thermal decomposition, a sharp weight decrease is typical. The primary breakdown of polymer structures occurred above 300 °C, with the most significant deterioration between 300 and 370 °C. This was expected since the literature reports that the composition of EC starts at about 280 °C and that the degradation of the polymer is concluded in one step, as can be observed [52, 53]. In this range, EC and microencapsulated OPE samples showed weight losses of 83.57 and 76.30%, respectively, as seen in Table S.1. For the microparticles, the decomposition of EC's functional groups in this temperature range exposed and decomposed the BACs. In contrast, the non-encapsulated OPE had a weight loss of over 50% at 300 °C, demonstrating the protective capability of the wall material, EC, and microencapsulation against heat. Overall, the microparticles exhibited significantly higher thermal stability, proving their ability to protect the extract up to 200 °C. The TGA analysis is crucial as cheese-making involves heating to 90 °C. The findings indicate that cheese incorporating microparticles will undergo less deterioration compared to cheese with free OPE, highlighting the enhanced thermal stability of the microparticles and their potential to protect BACs in food applications.

Characterisation of the Enhanced Cream Cheeses

The ability of the free and microencapsulated OPE to enhance cream cheese's nutritional value and increase its shelf life was assessed. Three cheese formulations were produced: C1 – negative control cream cheese; C2 – cream cheese incorporated with OPE; C3 – cream cheese incorporated with OPE-loaded microparticles. Figure S.2, in the Supplementary Information, presents a photograph of the three cheese samples on the day of production. Upon direct observation, it is noticeable that the cheese samples display different appearances. There is a significant colour change in the case of C2 due to the addition of the extract, resulting in a reddish colour. Observing sample C3, and comparing it to C1, it is evident that the incorporation of microparticles effectively masks the colour change, resulting in minimal alteration to the original colour. These findings suggest that incorporating microencapsulated OPE does not compromise the visual aspect of the product and proves effective in overcoming the colour limitation caused by the inclusion of free OPE. Afterwards, each sample was evaluated regarding its physical and chemical stability throughout a testing period of two weeks, at 3 different time points. The values for pH, syneresis and moisture content are presented in Fig. 3.

The pH of a cream cheese is a parameter that should be verified frequently due to its susceptibility to spoilage and proneness for microbial growth, due to its ideal pH, thus affecting its shelf life [54]. This parameter also plays an important role in the flavour of the product. Overall, it is evident that the pH of the cream cheeses remained relatively stable throughout the testing period. C1 showed a slight decrease, while C2 and C3 exhibited a slight increase, although the increase was not significantly different.

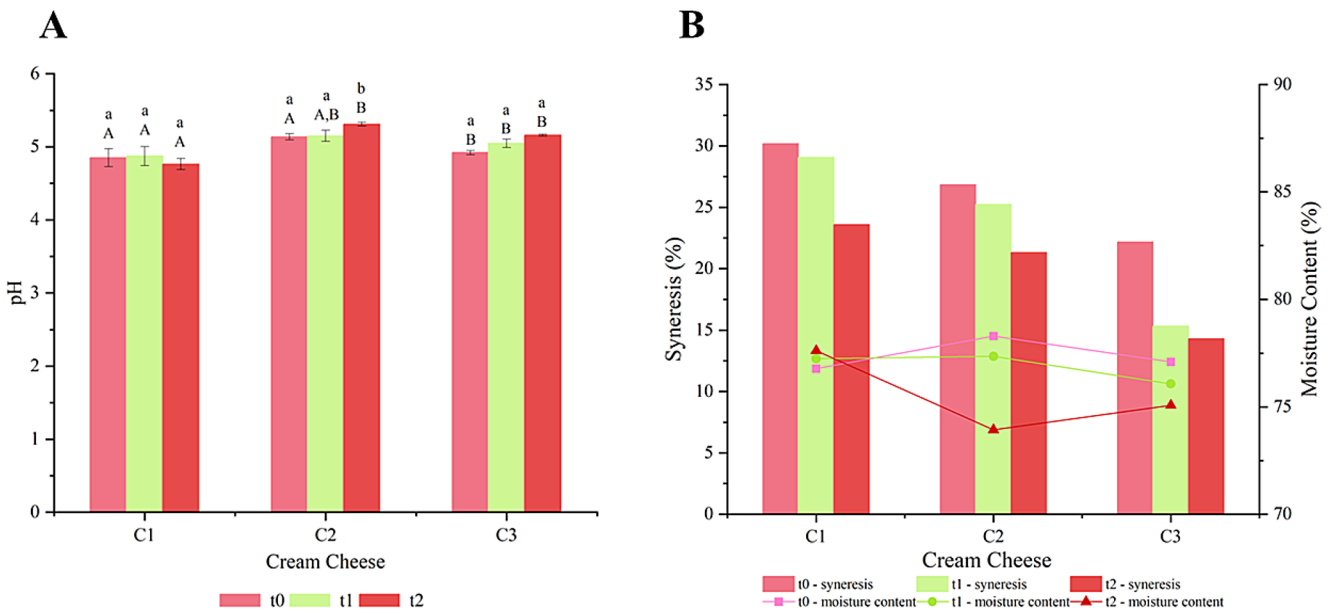


Fig. 3 (**A**) Variation of pH values for each cream cheese over time; (**B**) Variation of syneresis (represented by the bars) and moisture content (represented by the lines) of the cream cheeses over time. Negative control cream cheese (C1), cream cheese with OPE (C2), and cream cheese with OPE Microparticles (C3). t0 – week of production; t1 – one week after production; t2 – two weeks after production. Different lowercase letters represent a significant statistical difference between results in each sample over time; Different uppercase letters represent a significant statistical difference between samples at the same time

Analysing Fig. 3A, one can observe that all samples presented pH values within a range of 4.9–5.1 at the time of production. The addition of OPE and microencapsulated OPE led to a slight increase in pH for samples C2 and C3 when compared to C1 (negative control). Other literature studies revealed a slight increase in pH value when OP powder was added, supporting the results obtained [55]. Additionally, it is noticeable that the increase in the pH is significantly different between samples C1 and C3, meaning that the incorporation of the encapsulated extract influences this parameter. This fact can result from the core material used in the production of the microparticles. Indeed, EC has a neutral pH, and its incorporation into cream cheese might have led to an increase in the pH. To overcome this situation, different core materials should be evaluated to maintain this important parameter of the product.

Regarding the syneresis and moisture content, these are critical aspects when considering a product's performance. Syneresis refers to the separation of the liquid whey from the cheese, typically visible on the surface of the product, resulting from structural changes, such as the breaking and reformation of internal and external bonds of casein micelles. It is important to monitor this parameter during the processing and storage of the cheese [56]. Moisture content is crucial for the quality control of the product, as it can affect its texture and shelf life [57]. Cheeses with lower moisture content provide less water for microorganisms to thrive. From Fig. 3B, the syneresis revealed a trend to decrease over time in all cheese samples, which is favourable, showing the ability of the samples to maintain their casein network structure and retain the whey. Sample C3 revealed a lower syneresis value in the second week of storage, followed by C2 and then C1. This means that the addition of OPE and microencapsulated OPE allows the reduction of the syneresis phenomena and, therefore, for the cheese sample to remain stable regarding this parameter. Nonetheless, it is noticeable that in all the formulations, there is a trend regarding syneresis decrease, meaning that there is an increase in the water holding capacity. This ability might stem from the interaction between the PCs, present in the free and microencapsulated OPE, with the casein network, forming a stabilised complex and thus, preventing the rearrangement of the casein network and creating a denser gel network. Also, PCs can form hydrogen bonds with water, increasing the ability to retain whey [13]. Regarding moisture content, at the time of production, it ranged from 76.8 to 78.3% among samples, revealing a small difference. It revealed a tendency to increase in the negative control (sample C1), and decrease in the samples with OPE and microencapsulated OPE. This drop in moisture for samples C2 and C3 reveals that the OPE conferred the cheese the ability to better retain its water content. The observed decrease was less pronounced compared to C2 (a decrease of 5.6%), suggesting that OPE microparticles may aid in retaining more moisture compared to OPE alone. This may be due to the presence of EC, since nonphenolic material might be capable of binding water, and consequently,

the moisture content [58]. Results found in the literature found that the higher the concentration of extract added to the product, the higher the moisture content, and that moisture can increase when the extract is added [58]. This can explain the initial difference in moisture content between the control cheese and the fortified cheeses.

The total oxidation value for each cream cheese sample was also evaluated one and two weeks after production. The results can be observed in Fig. 4. This value accounts for both primary and secondary oxidation of the product and can be an indicator of its stability [59].

The results obtained reveal an increase in oxidation markers over time for all samples. The negative control sample exhibited higher values of oxidation, followed by the cheese with free OPE and the sample with microencapsulated OPE. It is observable that the three samples exhibited the same behaviour, with the oxidation increasing with time, which was anticipated. Nonetheless, it is observable that the incorporation of free and encapsulated OPE slowed down the oxidation of the product when compared to the negative control. It is noteworthy that the encapsulated extract exhibits considerably superior results. Indeed, the incorporation of the extract in the form of microparticles enables the antioxidants to be released gradually into the cream cheese. This means that the initial amount of extract added is released over time, contrary to when the extract is in its free form, where the antioxidants are available all at once. Therefore, the findings suggest that the controlled release of PCs from microparticles during storage allows for the protection of the antioxidants from their oxidation while helping to delay cheese oxidation.

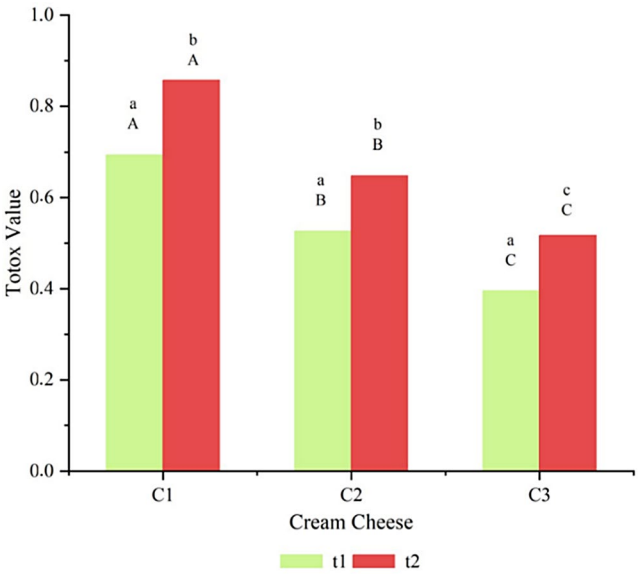


Fig. 4 Variation of the total lipid oxidation of the cream cheese samples over the testing period, where the green bars represent t1 – one week after production – and the red bars represent t2 – two weeks after production. Negative control cream cheese - C1, cream cheese with OPE - C2, cream cheese with OPE microparticles - C3. Totox value: Total Oxidation Value. Different lowercase letters represent a significant statistical difference between results in each sample over time; Different uppercase letters represent a significant statistical difference between samples at the same time

The results reveal the potential of incorporating OPE, particularly in microencapsulated form, to increase stability, reduce syneresis, stabilise its moisture content and prevent oxidation. Even though the cheese incorporated with OPE revealed promising results, the sample with microparticles revealed further increased stability. Moreover, the achieved results indicate that this approach could enhance the oxidative stability of other dairy products, including butter and yoghurt that display a high-fat content, and other foods, including emulsions such as mayonnaise and salad dressings. This strategy may improve the shelf life of the products while decreasing the need for synthetic/commercial preservatives. It is noteworthy to mention some limitations of this study, including the short storage period and potential differences in the OP extract composition. To overcome these limitations, it would be interesting to conduct a longer study and evaluate extracts from various onion varieties and mixtures of onion peels, to reach broader conclusions.

Antioxidant Capacity of the Enhanced Cream Cheeses

The different cream cheese samples were evaluated regarding their TPC and antioxidant activity over the testing period. The results obtained are displayed in Fig. 5.

Regarding the TPC of each sample, for all time points, C1 showed to be the sample with the lowest TPC, followed by C3 and lastly C2. It is expected that C2 has a higher TPC since the extract's effect is immediate, whereas, in the case of C3, there is a controlled release of the PCs from the microparticles, which explains the lower value of TPC in this case. The TPC value obtained for C1 can be explained by the breakdown of amino acid chains, caused by microbial activity, forming products with a structure that resembles PCs, which can react with the *Folin-Ciocalteu* reagent, leading to the obtained results [60]. Comparing the evolution of TPC for each sample over time, in the case of C1 and C3, it remained relatively stable. C1 values revealed a tendency to increase, whereas C3 also increases from t1 to t2, which can result from the controlled release previously mentioned, and its stability due to the protective barrier provided by the wall material of the microparticles. Regarding C2, the TPC value seems to decrease over time, probably due to the exposure of the PCs to light and oxygen, leading to oxidation. Literature reports that the low stability of PCs can influence their activity, leading to a decrease in their biological properties. Additionally, PCs have restricted bioavailability, and

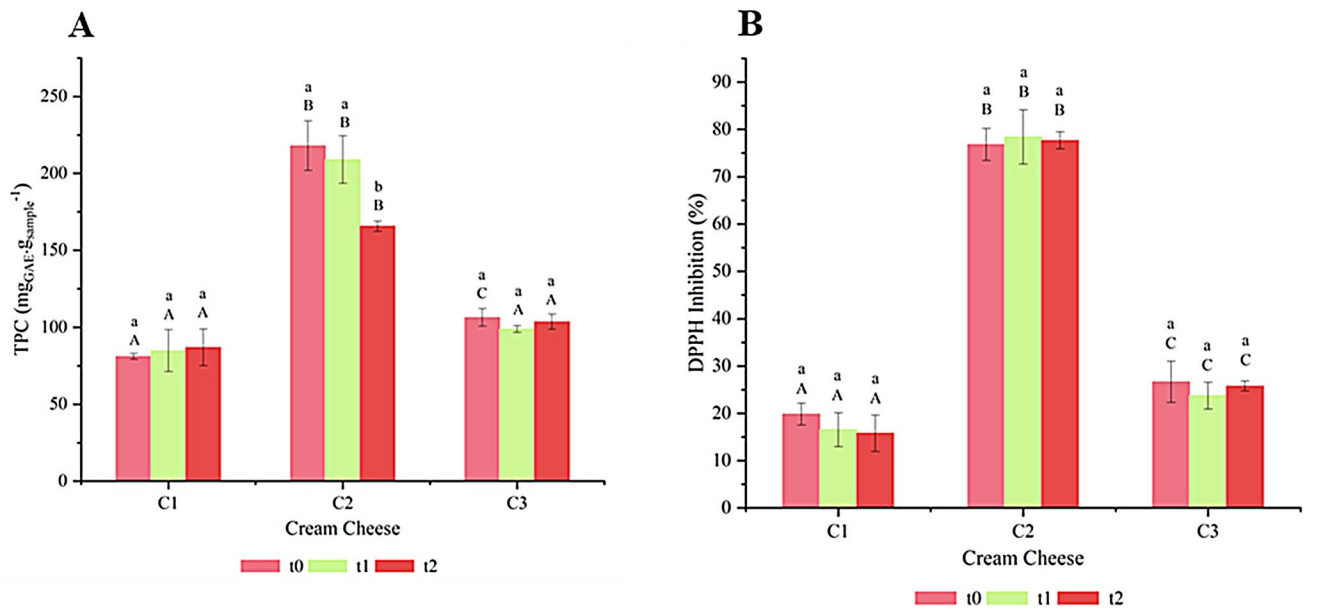


Fig. 5 TPC (**A**) and DPPH inhibition (**B**) of the cream cheeses. Negative control cream cheese (C1), cream cheese with OPE (C2), and cream cheese with OPE Microparticles (C3). t0 – week of production; t1 – one week after production; t2 – two weeks after production. Different lowercase letters in each column represent a significant statistical difference between results in each sample over time; Different uppercase letters in each line represent a significant statistical difference between samples at the same time

some of the compounds of the extract display low solubility in water, which limits their activity in certain products [29]. Therefore, the observed decrease in TPC in sample C2 may be related to the stability and availability of the free PCs present in the extract. It is noticeable that the inclusion of the extract within the polymer reduces the loss of PCs, leading to stable TPC, which demonstrates the efficacy of microencapsulation in the protection of these compounds.

The DPPH inhibition results for the different samples were revealed to follow the tendency of the TPC results. The observed decrease in inhibition capacity for sample C1 is likely due to the breakdown of amino acids, which influenced the TPC measurements. PCs are known for their antioxidant properties, which contribute to inhibition capacity. However, when amino acids break down, they can interfere with the TPC results, leading to an overestimation of the actual phenolic content in the sample. Consequently, the measured TPC does not accurately reflect the true concentration of PCs. This discrepancy results in a lower DPPH inhibition, as the actual amount of PCs is inferior to the one suggested by the TPC measurement. Regarding sample C2, it shows higher antioxidant capacity, which can be explained by the higher TPC result. PCs can act as great donors of hydrogen atoms, helping stabilise free reactive radicals [61]. Finally, C3 displayed higher antioxidant capacity when compared to C1, which can be explained by the incorporation of the microparticles in the samples; however, due to the controlled release of the PCs, this capacity is significantly lower than that of C2. It is noteworthy that the antioxidant capacity displayed in sample C1 might result from the presence of peptides, fatty acids, and vitamins (such as A, C and D) that have this property. Incorporating free and encapsulated OPE allowed a significant improvement in this property, enhancing the antioxidant content of the cream cheese. Moreover, results indicate that the controlled release of the phenolic extract from the microparticles into the cheese may be relatively low, affecting the antioxidant properties and TPC of sample C2 when compared to sample C1. This can be associated with the particle size; indeed, it has been reported that smaller particles increase the amount of BACs released, increasing the phenolic content and antioxidant capacity, since the surface area increases. In this case, even though the microparticles were within the micrometre range, the sample was considered polydisperse, which can lead to the presence of bigger particles that delay the release of the compounds [62]. Additionally, despite the porosity exhibited in the SEM analysis, it may be insufficient for the full release of the compounds from the microparticles, decreasing the amount in the medium. A possible strategy to overcome this problem would be to evaluate different core materials to produce the microparticles and select the most suitable one for the incorporation of a complex matrix, such as cream cheese.

The results demonstrated that the incorporation of phenolic-rich extracts from OP increases the antioxidant properties and TPC of cream cheese, indicating that it is possible to develop value-added products with increased shelf life and nutraceutical properties. The incorporation of

the encapsulated extract revealed positive outcomes mostly regarding syneresis, moisture content and antioxidant properties (in this case, not as positive as the free extract). Moreover, microencapsulation of the extract effectively masked undesirable changes in the cheese, such as alterations in colour and odour, while enhancing the physical and chemical stability of the OPE during storage. The observed reduction in syneresis and delay in oxidation are particularly advantageous for the dairy industry. Maintaining product quality over time without affecting its appearance or composition contributes to extended shelf life, reducing both product loss and distribution costs. Overall, the incorporation of microencapsulated phenolic extracts into food products shows promising potential. However, optimising encapsulation parameters is essential to ensure maximum efficacy without compromising product quality.

Conclusion

Onion peels (OP) are a main agricultural by-product with negative environmental impacts. To address this issue, phenolic compounds found in onion peels, which possess antioxidant properties, can be extracted and added to foods. To prevent their degradation and oxidation, microencapsulation can be used to prolong their stability. The present study aimed to evaluate the potential of using free and encapsulated OP extract as a natural preservative in cream cheese. The phenolic-rich extract was obtained through solid-liquid extraction using a Soxhlet apparatus with absolute ethanol, revealing a total phenolic content of 397 $mg_{GAE} \cdot g_{extract}^{-1}$ and interesting antioxidant properties. The extract demonstrated significant inhibition of α-amylase and β-glucosidase activities of 91.9% and 95.8%, respectively, indicating potential anti-diabetic benefits. The double emulsion solvent evaporation technique revealed a successful entrapment of the extract, with a production yield of 73.9%, a diameter of 18.7 μm, and with improved thermal stability in comparison to the free extract. The incorporation of the extract into the cheese revealed that extract-fortified samples had superior water retention and reduced syneresis, which is beneficial for the product. The encapsulation of the extract allowed the cheese to maintain a stable total phenolic content and antioxidant capacity, in comparison to the free extract, suggesting the controlled release of the extract from the microparticles. The sample with microparticles displayed the best oxidative stability. The findings indicate that the incorporation of OP extracts, especially microencapsulated, might be an effective strategy to enhance product stability and shelf life. The study highlighted the potential to valorise agricultural by-products, such as OP, through the obtainment of phenolic-rich extracts and their incorporation into foods, providing a strategy for waste management and reduction of their environmental impacts. Moreover, it contributes to the field of sustainable food innovation since it allows the development of foods enriched with natural compounds that improve their nutritional value and shelf life.

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Declarations

Conflict of interest The authors declare no conflict of interest.

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