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Research article

Development of edible nanoemulsions containing vitamin E using a low-energy method: Evaluation of particle size and physicochemical properties for food and beverage applications

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ABSTRACT

Pasta, a globally popular dish, serves as a complete meal around the world. This research aims to improve the nutritional value of pasta by enriching it with vitamin E. Firstly, vitamin E and sesame oil were mixed in different ratios (1:10, 1:5, 10:10) and dissolved in an aqueous medium at 50 °C with different concentrations of Tween 80 (10 %, 20 %, 30 %). Coarse emulsions were formed by gradual addition of the oil phase to the aqueous phase, followed by equilibration using an Ultratrax mixer at 15,000 rpm for 5 min. The target nanoemulsions were then produced using an ultrasonic system. After 30 days of storage, the most stable nanoemulsions containing 10 % Tween 80 and a 1:10 ratio of vitamin E to sesame oil showed minimal changes. In addition, nanoemulsions with 10 % Tween 80 and a 10:10 ratio of vitamin E to sesame oil showed less turbidity than those with 20 % and 30 % Tween 80. Evaluation of enriched pasta for physical, chemical and sensory properties compared to non-enriched samples showed no significant differences in properties such as pH, ash, total solids, texture and colour characteristics ($P < 0.05$). Enriched pasta samples showed an increase in moisture content of 0.94 % and a decrease in weight loss of 2.13 % compared to the control, with improved brightness (L) and yellowness (b) due to the addition of nanoemulsion. Sensory evaluation showed higher scores for pasta samples enriched with nanoemulsions containing vitamin E compared to control samples. This pioneering study introduces nanoemulsion technology to improve the nutritional profile of pasta by enriching it with vitamin E. The research demonstrates the successful formulation of stable nanoemulsions and their positive effects on pasta properties, suggesting promising avenues for improving public health through innovative pasta enrichment methods.

1. Introduction

In recent years, numerous studies have evaluated the absorption of vitamins through different food matrices [1] According to the World Health Organisation (WHO), the majority of people worldwide do not receive adequate amounts of vitamins due to an inadequate daily diet [2]. Among cereal products, pasta is one of the most popular and efforts have been made to enrich it with nutraceutical compounds such as vitamins [3]. Heating, whether drying or baking, is the most effective processing method for reducing vitamins and other nutritional compounds [4]. To increase the chemical stability of natural ingredients, new approaches called micro-

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and nano-encapsulation have been developed [5].

Nanoemulsions are a type of emulsion with droplets of small diameter, typically in the range of 20–200 nm, due to their small diameter their appearance is transparent or bluish translucent [6]. The preparation of nano-emulsions can be divided into two types: low energy and high energy [7]. High-energy methods use mechanical equipment such as microfluidizers and high-speed homogenisers or ultrasound systems [8]. The main low-energy methods include spontaneous emulsification, emulsion inversion point method, phase inversion temperature and phase inversion composition [9]. Sometimes a combination of low and high energy methods can produce a nano-emulsion with higher physical stability and bioavailability [10].

Spontaneous emulsification is used to form nano-emulsions, which require small molecule surfactants that can quickly adsorb onto the surface of oil droplets and form a special arrangement where the hydrophobic tail of the surfactant interacts with the hydrophobic tail of the oil molecules (fatty acids) and the hydrophilic tail interacts with the hydrophilic part of the oil molecules (glycerol) [11].

The nanoemulsions have a very low interfacial tension and considerable oil in the interfacial water region. The nanoemulsion has greater sustainability aspects than basic micellar solutions, and its thermodynamic resilience gives it an advantage over unstable dispersions such as suspensions and emulsions, as it is made with much less energy (heat or mixing) and has a long shelf life [12].

Sesame oil, extracted from sesame seeds (*Sesamum indicum* L.), is known for its nutritional and therapeutic value, with sesamolin and console as its main components. These compounds have antioxidant activity and can increase plasma gamma-tocopherol, thereby enhancing vitamin E activity to prevent cancer and heart disease [13]. Vitamin E, which is soluble in oil, provides antioxidant activity, strengthens the immune system and prevents coronary heart disease. It has several isomers, including α-, β-, γ- and δ-tocopherols and tocotrienols, with different bioavailabilities. Among these, α-tocopherol has the highest biological activity [14].

Nano-based dispersions offer promising ways to overcome the problems associated with low solubility and bioavailability of bioactive and fat-soluble compounds, especially vitamins [15]. Recently, nano-emulsions containing bioactive and food-grade lipophilic nutraceuticals have been developed for the production of beverages, liquid foods and hydrogels [16].

Research has shown that vitamin E-enriched nanoemulsions, particularly those made from food-grade ingredients, can significantly improve the nutritional value of various foods, including pasta, sauces, dressings, baked goods and dairy products [17]. These nanoemulsions are stable and have positive effects on the properties of pasta, suggesting their potential for scalable production and improving overall food quality and consumer appeal [18]. They can also be used in the food industry for the encapsulation and delivery of bioactive lipophilic compounds such as vitamins and nutraceuticals [19]. Although promising, the produced nanoemulsions may present challenges such as potential stability issues during prolonged storage or when exposed to varying environmental conditions.

This research pioneers the use of nanoemulsion technology to fortify pasta with vitamin E, addressing a critical gap in current pasta production methods. Despite efforts to fortify pasta, significant vitamin and mineral losses occur during production and cooking. By recognising the health benefits of vitamin E and using innovative nanoemulsion techniques, this study aims to improve the safety and nutritional value of pasta while addressing nutrient deficiencies. The research demonstrates the successful preservation of vitamin E in pasta, marking a significant advance in nutrient retention during production and cooking. Conducted in an industrial setting, the study demonstrates the practical feasibility and scalability of this approach. The novelty of using nano-methods to fortify pasta further highlights the potential impact on public health and dietary practices. Overall, the aim is to fortify pasta with vitamin E using nanoemulsion technology and to evaluate its impact on pasta properties and consumer acceptance in order to improve overall health outcomes.

2. Material and methods

2.1. Materials

All chemicals used in this study were of analytical grade and were purchased from Sigma-Aldrich (Spain). Vitamin E was purchased from Zahravi CO. (Tehran, Iran) and sesame oil was purchased from Behshahr Co. Tehran (Iran).

2.2. Preparation of nano emulsions containing vitamin E

This involved mixing vitamin E with sesame oil at different ratios [1 (vitamin E):10 (sesame oil), 1:5 and 10:10], heating to 50 °C and adding different concentrations of Tween 80 (10 %, 20 % and 30 %). The resulting mixture was gradually added to distilled water to form a coarse emulsion, which was stirred to equilibrium. The coarse emulsion was further homogenised using an Ultra-Turrax homogeniser at 15,000 rpm for 5 min to improve droplet size and stability. Finally, the nanoemulsion was prepared by ultrasonication [20], which includes preparation of the disperse phase, formation of the initial coarse emulsion, homogenisation and preparation of the nanoemulsion [21].

2.3. Characterization nano emulsions

2.3.1. Particle size

The particle size distribution of the nanoemulsion sample was assessed using a Nanosizer dynamic light scattering instrument (Malvern, USA). Samples were diluted 1:100 in phosphate buffer solution and injected into the instrument at room temperature [22]. Mean volume particle diameter and distribution were determined from the data collected [23]. Droplet diameter was calculated from the mean volume diameter (D43) and reported as the final result [24].

The following equation was used to calculate the droplet dispersion index:

Span = D (90%) – D (10%)/D (50%) (1)

Where D (90 %) is a diameter smaller than 90 % of the total volume of droplets in the system, D (10 %) a diameter of less than 10 % of the total volume of droplets in the system and D (50 %) the diameter of a droplet smaller than 50 % of the total volume of droplets in the system [25].

2.3.2. Stability index

To assess the stability of the emulsion against gravitational separation, aggregation, coalescence, oscillation and Ostwald ripening, droplet diameters were measured at intervals of 1, 7, 14, 21 and 30 days with samples stored at a controlled 20 °C. Monitoring the droplet size distribution over time allows the ability of the emulsion to resist destabilisation and maintain stability to be assessed [26].

2.3.3. Physical stability

To assess stability, 15 mL of each sample was left undisturbed in a test tube at room temperature for 90 days. The supernatant was then measured and subtracted from the initial 15 mL to calculate the volume of the oil layer. This value was divided by the initial sample volume to determine the percentage of retained oil [27]. This method provides an indication of the ability of the sample to resist separation and maintain stability over time without agitation.

2.3.4. pH and turbidity

A digital pH meter (model PL 600, ezdo, Taiwan) was used to measure the pH of the nanoemulsion samples at room temperature according to the method described by Leal-Calderon et al. [28]. After 30 days, the nanoemulsion samples were transferred to a cuvette and their absorbance was measured at 600 nm using a UV–visible spectrophotometer (model 2100 A, Unico, USA). Distilled water (twice distilled) was used as a control in this experiment, as described by Lee and McClements [29]. By comparing the pH and absorbance values of the nanoemulsion samples with those of the control, we can gain insight into the stability and characteristics of the nanoemulsion.

2.3.5. Rheological properties

Rheological tests were carried out on the nanoemulsion using a Physical Anton Paar Model 301 MCR Rheometer equipped with a thermal circulator. Nanoemulsion samples were tested at 4 ± 0.1 °C and shear stress and viscosity were measured at shear rates ranging from $2s^{-1}$ to 100s-1 over a 10 min interval. Using the power law model described by Bouchemal et al. [30], the flow index and consistency coefficient were determined. These measurements provide insight into the behaviour of the nanoemulsion and its suitability for various applications.

2.4. Physiochemical properties of pasta

2.4.1. - Moisture and ash determination

Total ash was determined by the AOAC 923.04 17th Ed method as described by Hooper et al. [31]. These well-established analytical techniques provide valuable insight into the composition and quality of pasta to ensure compliance with nutritional standards and suitability for culinary use.

2.4.2. -Weight loss

Pasta quality was assessed by weight loss measurements according to AACC 66–50.01 [32]. This standard defines cooking conditions and procedures to ensure data reliability and consistency [33]. Adherence to these guidelines allows comparison with industry standards and provides valuable insight into the cooking behaviour of pasta. This knowledge is used in the development of new pasta products to meet consumer needs and preferences.

2.4.3. - Total solid content in water after cooking

The total solid content of the pasta samples was determined using the methods described in the 10th edition of the AACC (American Association of Cereal Chemists) Approved Methods [34]. This method involves the complete removal of moisture from the sample by oven drying and subsequent measurement of the remaining solids content.

2.4.4. -pH and crude protein

The pH of cooked pasta strands was determined using a Metrohm 827 pH meter. Samples were prepared by homogenising 5 g of pasta strands with 50 mL of deionised water for 5 min. After standing for 30 min, the suspension was filtered before pH measurement [35]. The crude protein content of pasta samples was measured using AACC method 46–13, according to Pagnussatt et al. [36]. This standardised approach ensures reliable determination of pasta protein content, which is critical in food production. These rigorous methods ensure the accuracy and consistency of data that can be used to make decisions about pasta quality and nutritional value.

2.4.5. -Texture profile analysis (TPA)

A Brookfield CT_3 texture analyser with Texture Expert software was used to assess pasta texture. Pasta samples enriched with nanoemulsions were subjected to compression tests. Parameters such as hardness, stickiness, cohesiveness, chewiness and elasticity were obtained by analysing stress curves versus deformation. The compression rate was set at 60 % with a test speed of 2 mm s^{-1} [37]. Texture profile analysis (TPA) is widely used to evaluate the mechanical properties of foods. The application of this method provides valuable insight into the texture of pasta. By using a standardised approach and analysing different textural parameters, the effect of nanoemulsion enrichment on pasta texture can be evaluated and used to guide the development of new and improved pasta products.

2.4.6. -Color measurement

To measure the colour of uncooked pasta samples fortified with Nano Emulsion Vitamin E (E.P.E), a Koolertron 5 MP 20-300X USB Digital Microscope Magnifier was used to measure the L*, a* and b* values. The L* value measures the black to white colour of the pasta, while the a* value measures the redness and greenness and the b* value measures the yellowness and blueness. A minimum of six measurements were taken for each sample to ensure accuracy and consistency of results [36,38]. This rigorous approach to colour measurement allowed us to obtain accurate and reliable data on the colour properties of the pasta samples, which can be used to inform the development of new and improved pasta products.

2.5. Vitamin E

The antioxidant activity of the fortified pasta samples was assessed by their ability to scavenge the free radical DPPH, which is indicative of preserved vitamin E [39]. The samples were reacted with stable DPPH radical in ethanol solution. Specifically, 0.5 mL of sample was mixed with 3 mL of absolute ethanol and 0.3 mL of 0.5 mM DPPH radical solution [40]. Reduction of the DPPH radical by antioxidants resulted in a colour change from purple to yellow, measured as absorbance (Abs) at 517 nm after 100 min using a UV-VIS spectrophotometer (Spectra ax Gemini XS: Molecular Devices, Sunnyvale, CA). Background effects were accounted for using blank (ethanol and sample) and control (ethanol and DPPH radical) solutions. The percentage of antioxidant activity was calculated from the absorbance values of sample and control [41].

2.6. Nano emulsion efficiency

To determine the efficiency of the nano-emulsions in retaining vitamin E in the pasta after the extrusion and drying process, the initial amount of vitamin E in the nano-emulsions was measured and compared with the amount of vitamin E retained in the pasta [39,42]. The vitamin E assay was performed using a UV-VIS spectrophotometer (model UV2100, UNICCO, USA) according to the method described by Garcia et al. [41].

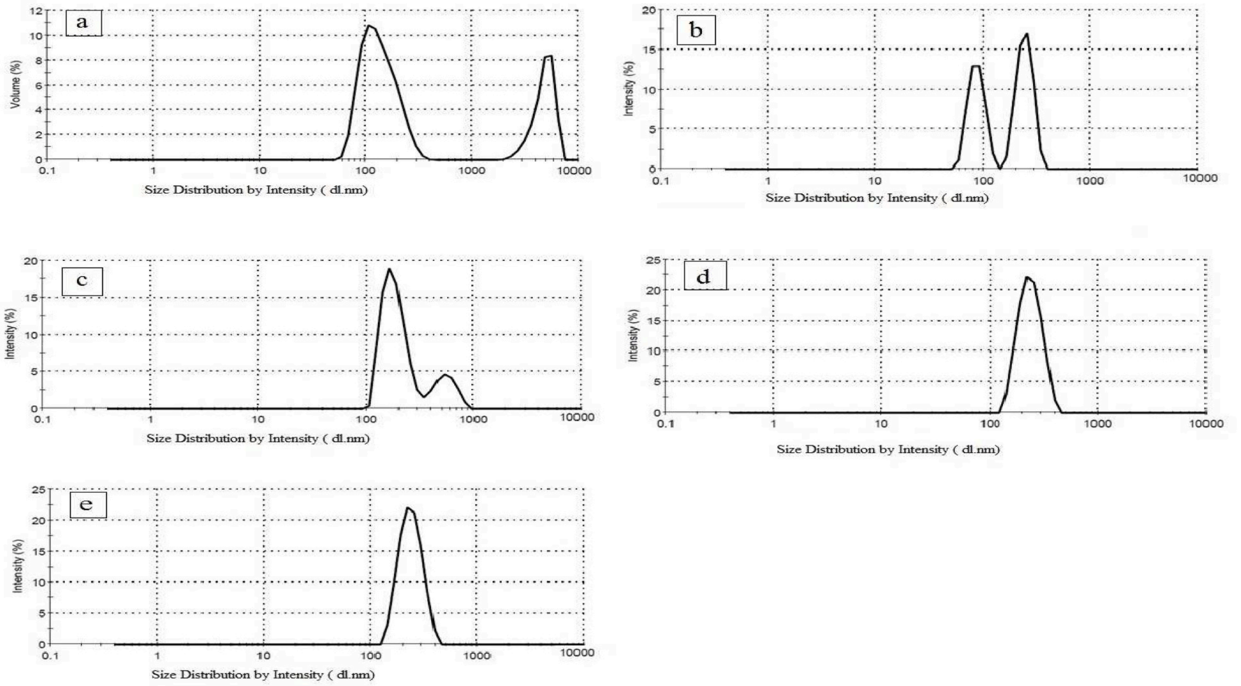

Fig. 1. Medium diameter emulsion nanoparticle containing 10 % tween with l g of vitamin E to 10 g of sesame oil during 1 (a), 7 (b), 14 (C), 21 (d), and 30 (e) day of storage time.

2.7. Sensory evaluation

Sensory analysis of pasta samples fortified with nano-emulsion vitamin E (E.P.E.) was carried out using a consumer panel of 20 members (11 women and 9 men, aged 25–40 years) according to ISO standard 6658:2005E. Panelists rated samples based on smell, taste, texture and overall quality using a five-point hedonic scale. Samples were presented in random order to reduce bias. This method provides valuable insight into the acceptability of the fortified pasta product and areas for sensory improvement [43].

2.8. Statistical analysis

Experiments were performed in triplicate and statistical analysis was performed using SAS statistical software (version 9.8) with a randomised complete block design. Duncan's test was used to determine significance, with p values < 0.05 indicating significance. Conducting experiments in triplicate and using statistical analysis increases the reliability of results and ensures that observed effects aren't due to chance.

3. Results and discussion

3.1. Physiochemical properties of nano emulsion

3.1.1. -Particle size of nano emulsions of vitamin E

The study investigated the influence of different concentrations of Tween 80 and vitamin E on the particle size of nanoemulsions formulated with sesame oil. The results showed that the lowest mean particle diameter of 160.78 nm was obtained on day 1 for nanoemulsions prepared with 10 % Tween 80 and a 1:10 (w/w %) ratio of vitamin E to sesame oil (treatment A) (Fig. 1a). Conversely, the highest mean particle diameter of 336.87 nm was observed for nanoemulsions formulated with 30 % Tween 80 and a 10:10 (w/w %) ratio of vitamin E to sesame oil (treatment E). The mean particle diameter for all treatments was 214.2 nm with a mean dispersion index of 0.216. Notably, increasing the Tween 80 content from 10 % to 30 % at a constant ratio of vitamin E to sesame oil (1:10) resulted in a significant difference in nanoparticle size ($p < 0.05$), with a slight decrease in size observed when Tween 80 was increased to 30 %. Consequently, based on these results, 10 % w/w Tween 80 was selected for future studies. Unexpectedly, increasing the Tween 80 concentration to 20 % SER (surfactant to emulsion ratio) resulted in an increase in droplet size and dispersion. This phenomenon is consistent with previous research [44]. The increase in droplet size at higher Tween 80 concentrations could be attributed to the formation of viscous liquid crystal structures at the interface of the oil, hindering surfactant migration into the aqueous phase and delaying nanoemulsion formation [45]. In addition, beyond a certain threshold, escalating surfactant concentration can lead to the formation of a crystalline phase with increased viscosity, which hinders spontaneous oil wall breakage [46]. In addition, increased surfactant concentration can promote the formation of surfactant micelles, increase the local osmotic pressure and induce droplet flocculation [47]. The instability of nanoemulsions at higher surfactant concentrations could also be due to the depletion mechanism of flocculation of adsorbed surfactant micelles [48].

Vitamin E can act as a cofactor in the presence of Tween 80, meaning that when vitamin E molecules are sandwiched between surfactant molecules, they can achieve their desired curvature [49]. This ability of vitamin E is attributed to the presence of functional groups in its structure. Vitamin E contains the hydrophilic carbonyl group (hydroxychromium) which is capable of forming hydrogen

Table 1
Comparison of the average vitamin E Nano-emulsion treatments.

Treatments	Nano-emulsion treatments (nm)					pH	Physical stability (%)	Turbidity (%)
	1 day	7 day	14 day	21 day	30 day			
Tween (10 %), vitamin E (1 g) sesame oil (10 g)	160.56 ± 20.15^{h}	181 ± 15.36^{g}	197.87 ± 21.5^{f}	210 ± 22.35^{i}	221 ± 10.23^{i}	5.605 ± 0.56^{a}	8.5 ± 0.93^{b}	0.2845 ± 0.01^{a}
Tween (10 %), vitamin E (1 g) sesame oil (5 g)	180 ± 22.51^{e}	188.82 ± 18.6^{f}	197.64 ± 25.3^{f}	258.95 ± 15.3^{f}	324 ± 9.36^{f}	5.485 ± 0.36^{b}	$9\pm\pm0.89^{b}$	0.265 ± 0.00^{c}
Tween (10 %), vitamin E (10 g) sesame oil (10 g)	24 ± 19.253^{b}	279.6 ± 16.36^{c}	319 ± 25.22^{c}	391.7 ± 14.3^{c}	465 ± 17.3^{c}	5.395 ± 0.53^{c}	9 ± 1.0^{b}	0.224 ± 0.02^{e}
Tween (20 %), vitamin E (1 g) sesame oil (10 g)	165.08 ± 18.23^{f}	180.55 ± 20.55^{h}	198 ± 18.3^{f}	216.55 ± 11.2^{h}	234.1 ± 17.5^{h}	4.96 ± 0.61^{d}	8.5 ± 0.56^{b}	0.2735 ± 0.01^{b}
Tween (20 %), vitamin E (1 g) sesame oil (5 g)	195.46 ± 15.9^{d}	209.65 ± 25.33^{e}	223 ± 15.3^{e}	305.89 ± 10.2^{e}	390 ± 23.6^{e}	4.805 ± 0.56^{e}	10.5 ± 0.89^{a}	0.254 ± 0.02^{d}
Tween (20 %), vitamin E (10 g) sesame oil (10 g)	240.64 ± 23.5^{c}	296 ± 23.3^{b}	352 ± 12.3^{b}	466 ± 22.3^{b}	580 ± 26.3^{b}	4.75 ± 0.36^{f}	10.5 ± 1.1^{a}	0.2245 ± 0.01^{e}
Tween (30 %), vitamin E (1 g) sesame oil (10 g)	163.12 ± 16.33^{g}	180.1 ± 18.33^{h}	195.43 ± 15.3^{g}	222.1 ± 16.3^{g}	247 ± 21.4^{g}	4.485 ± 0.41^{g}	9.5 ± 0.83^{ab}	0.2635 ± 0.03^{c}
Tween (30 %), vitamin E (1 g) sesame oil (5 g)	243.12 ± 21.33^{b}	268.8 ± 17.3^{d}	293 ± 19.3^{d}	361 ± 14.2^{d}	432.54 ± 19.3^{d}	4.22 ± 0.62^{h}	10.5 ± 0.53^{a}	0.2615 ± 0.01^{c}
Tween (30 %), vitamin E (10 g) sesame oil (10 g)	337.23 ± 24.33^{a}	347 ± 25.33^{a}	363 ± 21.3^{a}	496.3 ± 12.3^{a}	629 ± 12.3^{a}	4.085 ± 0.15^{i}	10.5 ± 0.65^{a}	0.2235 ± 0.02^{e}

The treatments with a letter in common do not differ statistically significantly at the probability level of 5 %.

bonds with water molecules [50]. On the other hand, the long carbon chains in its structure are hydrophobic and help to dissolve the vitamin in the oil phase. Vitamin E therefore has an amphiphilic structure. It is likely that vitamin E has a synergistic behaviour with Tween 80 [51].

In our study we observed a remarkable increase in the size of nanoemulsion droplets when the concentration of vitamin E in the oil phase was increased to more than 50 % (Table 1). This finding highlights the importance of considering the composition of the oil phase in nanoemulsion formulation. Low-energy methods, such as spontaneous emulsification, exploit the physicochemical properties of both oil and aqueous phases, including viscosity, to produce nanoparticles with smaller droplet sizes [52]. Our results are consistent with previous research by Ref. [53] who investigated the effect of oil composition on the particle size of vitamin E nanoemulsions and found an average increase in particle diameter of over 60 % with increasing viscosity of vitamin E in the oil phase [54].

Analysis of the treatments on day 7 showed a significant difference in the particle size of the vitamin E nanoemulsions produced (Fig. 1b). As shown in Table 1, the nanoemulsion with the largest particle size (348.5 nm) was formulated with 30 % Tween 80 and a 10:10 (w/w%) ratio of vitamin E to sesame oil (treatment E), whereas the smallest particle size (179.7 nm) was obtained with 30 % Tween 80 and a 1:10 (w/w%) ratio of vitamin E to sesame oil (treatment K). The mean particle diameter of the nanoemulsion cohort was 236.83 nm, reflecting a mean change of 22.62 nm, accompanied by a mean dispersion index of 0.223. Results from previous studies indicate that both the surfactant to emulsion ratio (SER) and the surfactant to oil phase ratio (SOR) have a significant influence ($p < 0.05$) on the particle size distribution of nanoemulsions [55]. These findings highlight the multifaceted nature of nanoemulsion formulation, where subtle variations in surfactant and oil phase ratios can result in significant differences in particle size. Understanding and optimising these ratios is critical to tailoring nanoemulsions for specific applications and ensuring that desired properties such as stability and bioavailability are achieved.

Synthesis of vitamin E nanoemulsions revealed a marked influence of tween 80 concentration on particle size, with a gradual decrease with increasing surfactant concentration (Table 1) [46,56], an observation consistent with previous findings in the literature. The unique chemical structure of tween 80, characterised by an oleate ester double bond in its hydrophobic chain, makes it more mobile than other saturated chain tweens such as tween 20 [51]. Conversely, high molecular weight tweens, such as Tween 85, exhibit reduced emulsifying activity due to their slower rate of movement from the oil phase to the aqueous phase [57].

The reduction in droplet size with increasing surfactant concentration can be attributed to the role of Tween 80 in increasing the adsorption of surfactant molecules onto the oil surface, thereby reducing the surface tension [58]. In addition, the increased concentration of Tween 80 facilitates the diffusion of more surfactant molecules from the oil phase into the aqueous phase, thereby promoting the formation of smaller emulsion droplets [59]. This phenomenon is consistent with the findings of Hategekimana et al., who also observed a decrease in droplet diameter with increasing surfactant concentration [60]. On day 7, the evolving trend in nanoemulsion particle size mirrored that observed on day 1, highlighting the significant influence of the ratio of vitamin E to sesame oil on particle size ($p < 0.05$) (Table 1).

On day 14, significant variations in the particle size of the nanoemulsions were observed (Fig. 1c). The highest particle size of 362.9 nm was recorded for the nanoemulsion formulated with 30 % Tween 80 and a ratio of vitamin E to sesame oil of 10:10 (w/w%). Conversely, the smallest particle size of 197.25 nm was obtained with a formulation containing 20 % Tween 80 and a ratio of vitamin E to sesame oil of 1:10 (w/w%). Interestingly, increasing the Tween 80 concentration from 10 % to 30 % had no significant effect on the particle size of the nanoemulsion samples ($p > 0.05$). The mean diameter of the nanoparticles was 259.88 nm, with a mean variation of 23.04 nm and a mean dispersion index of 0.234. In addition, an increase in the ratio of vitamin E to sesame oil corresponded to an increase in nanoparticle diameter. This phenomenon could be attributed to the high ratio of sesame oil to vitamin E and the complete dissolution of the oil composition in the aqueous phase, facilitated by the good coverage of vitamin E by the carrier oil [61]. The functional and stability properties of nanoemulsions are often dependent on the physicochemical properties of the oil phase, including polarity, water solubility, surface tension, refractive index, viscosity, phase behaviour and stability [62]. While certain edible oils are conducive to the production of nanoparticles with fine particle sizes, the resulting emulsions often exhibit low stability [63]. Essential oils, characterised by relatively high polarity, interfacial tension and low viscosity, are well suited to the production of nanoemulsions using low energy methods. Most of these oils consist mainly of long-chain triacylglycerols (LCTs). On the other hand, oils such as medium-chain (MCT) and short-chain (SCT) triacylglycerols, which are commonly used in the food industry, have high viscosity, low polarity and reduced interfacial traction, making the preparation of nanoemulsions challenging [64]. Nanoemulsions formulated with LCT and MCT oils typically exhibit high physical stability. In low energy methods such as spontaneous emulsification, MCT oils such as coconut oil and sesame oil are more effective than LCT oils in producing nanoparticles with smaller particle sizes [65].

On day 21, significant differences in nanoparticle size were observed between samples with different formulations ($p < 0.05$) (Fig. 1d). The nanoemulsion formulated with 30 % Tween 80 and a 10:10 (w/w%) ratio of vitamin E to sesame oil (treatment E) exhibited the highest particle size (496.1 nm), whereas the lowest particle size (209.8 nm) was observed for the nanoemulsion containing 10 % Tween 80 and a 1:10 (w/w%) ratio of vitamin E to sesame oil (treatment A). Increasing the concentration of Tween 80 from 10 % to 30 % had a significant effect on the particle size ($p < 0.05$) [66]. Specifically, the nanoparticle diameter increased from 209.8 to 225.55 nm with increasing amount of Tween 80 surfactant. On day 21, the mean nanoparticle diameter was 325.38 nm, reflecting a mean change of 65.50 nm, with a mean dispersion index of 0.274 (Table 1). Notably, the nanoparticle size increased with increasing ratio of vitamin E to sesame oil (0.1, 0.2 and 1), ranging from 209.8 to 391.6 nm. Overall, it can be concluded that the droplet size of the nanoemulsion remained relatively stable over the shelf life due to droplet stability and surfactant composition. Over time, the surface tension and interactions between the surfactant and the aqueous phase would reach equilibrium, leading to significant changes in nanoparticle diameter [67].

On day 30, significant differences in nanoparticle size were observed between samples with different compositions (Fig. 1e). The nanoemulsion formulated with 30 % Tween 80 and a 10:10 (w/w%) ratio of vitamin E to sesame oil (treatment E) exhibited the highest particle size (629.05 nm), whereas the lowest particle size (222.55 nm) was observed for the sample containing 10 % Tween 80 and a 1:10 (w/w%) ratio of vitamin E to sesame oil (treatment A). Increasing the Tween 80 content from 10 % to 30 % at a constant ratio of vitamin E to sesame oil (1:10) significantly changed the nanoparticle diameter. Specifically, nanoparticle size increased from 221.25 to 247.26 nm with increasing amount of Tween 80. On day 30, the mean diameter was 391.4 nm, with a mean change of 66.01 nm and a dispersion index of 0.254.

3.2. *pH*

Significant differences ($p < 0.05$) were observed in the pH values of the samples (Table 1). The nanoemulsion containing 30 % Tween 80 and a 10:10 (w/w%) ratio of vitamin E to sesame oil had the lowest pH (4.08), while the sample containing 10 % Tween 80 and a 1:10 (w/w%) ratio of vitamin E to sesame oil had the highest pH (5.6). These results indicate that the pH decreased with increasing concentration of Tween 80. Furthermore, samples with the same concentration of Tween 80 showed variations in pH depending on the ratio of vitamin E to sesame oil, with the sample containing a 1:10 ratio of vitamin E to sesame oil having the highest pH and the sample containing a 10:10 ratio of vitamin E to sesame oil having the lowest pH. These findings are consistent with previous research suggesting that the pH of the nanoemulsion system becomes more alkaline as the droplet diameter decreases and more acidic as the droplet diameter increases [68]. pH variations in nanoemulsions can result from interactions between formulation components such as surfactants, oils and water, as well as changes in droplet size and surface area to volume ratio.

3.3. *Physical stability*

The physical stability of the nanoemulsions was evaluated over a storage period of 90 days to determine the optimum emulsion for macaroni production. The data obtained from the physical stability test (Table 1) showed significant differences ($p < 0.05$) in the physical stability values among the nanoemulsions. The highest instability (10.5 %) was observed in samples containing 20 % and 30 % Tween 80 and vitamin E to sesame oil ratios of 1:5 and 10:10 (w/w%). Conversely, the sample containing 10 % Tween 80 and a ratio of vitamin E to sesame oil of 1:10 (w/w%) showed the lowest instability (8.5 %) of the samples. Emulsion stability, whether synthetic or thermodynamic, is a critical property, particularly from an appearance point of view. Stability is closely related to the adsorption of surfactant molecules at the interface of two phases, which reduces the surface interfacial tension [49,56,69]. The instability of the nanoemulsions was directly correlated with the Tween 80 content; as the Tween content increased, the tendency to instability also increased. According to Stokes' law, the physical stability of the emulsion is inversely proportional to the size of the droplet diameter. As the diameter of the nanoemulsion particles decreased, the physical stability increased. The droplet size and dispersion distribution (polydispersity index) of nanoemulsions in colloidal systems are crucial in determining their physicochemical properties. The stability of these parameters over time indicates the stability of the system. The creaming rate of a lipid droplet increases with increasing droplet diameter [70]. Therefore, over a given storage period, emulsions containing larger droplets tend to become creamier than those containing smaller droplets [71]. As a result, nanoemulsions containing larger droplets or with an increase in mean droplet

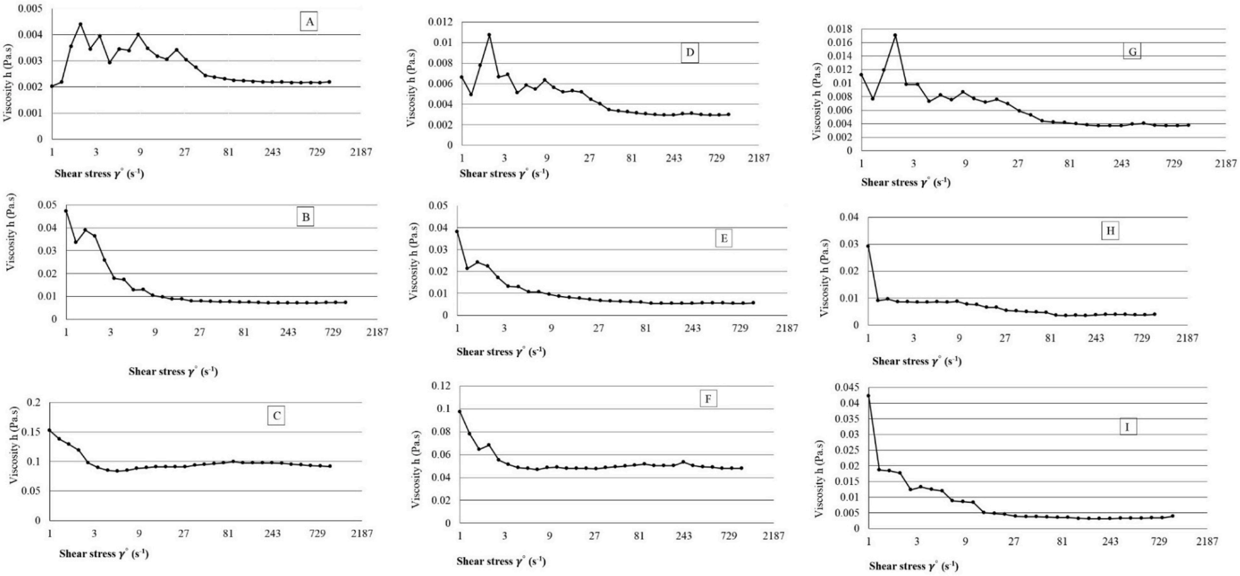

Fig. 2. Viscosity - Shear rate of Nano emulsion. Formulation A (10 % tween, vitamin E to sesame oil (1: 10)), B (10 % Tween, Vitamin E to Sesame Oil (1:5)), C (10 % Tween, Vitamin E to Sesame Oil (10:10) D (20 % Tween, vitamin E to sesame oil (1: 10)), E (20 % Tween, Vitamin E in Sesame Oil (1:5), F (20 % Tween, Vitamin E in Sesame Oil (10:10)) G (30 % tween, vitamin E to sesame oil (1: 10)), H (30 % Tween, Vitamin E in Sesame Oil (1:5)) & I (30 % Tween, Vitamin E in Sesame Oil (10:10)).

diameter over time may become two-phase and creamy. Furthermore, the physical stability of the nanoemulsions decreased as the ratio of vitamin E to sesame oil increased. As the oil phase increased, the interaction between the surfactant and the nanoparticles weakened over time, leading to a loss of particle stability and subsequent separation of the dispersed phase from the continuous phase [72].

3.4. Turbidity

The turbidity values of the nanoemulsions showed significant differences ($p < 0.05$), as shown in Table 1. An emulsion is considered transparent when its turbidity is less than 0.05 cm-1. Our results showed a negative correlation between turbidity and the amount of Tween 80. Specifically, higher surfactant content resulted in brighter nanoemulsions, whereas lower surfactant content resulted in higher turbidity. This observation is consistent with a study by Wang et al., in 2014, where the turbidity of nanoemulsions was inversely correlated with surfactant concentration [73]. The highest turbidity was observed in nanoemulsions containing a 1:10 ratio of vitamin E to sesame oil with a constant concentration of Tween 80 (10 %, 20 % and 30 %). This finding is consistent with the results of a study by Lee et al., in 2010, which showed that the turbidity of nanoemulsions increased with increasing oil concentration [63]. In our study, the optimum turbidity was determined by evaluating the surfactant/oil phase ratio (SOR concentration) between 0 and 0.1 % oil content, aiming for a turbidity of less than 0.05 cm^{-1}. The optimum formulation was achieved at a 16 % SOR concentration with the lowest surfactant consumption, in agreement with the results reported by Lee and McClements in 2010 [29].

3.5. Rheological properties

In this study, the relationship between shear stress and viscosity was investigated to elucidate the flow behaviour of nanoemulsions at different shear rates ranging from 1 to 729 s-1 over 10 min intervals. The flow behaviour of three nanoemulsions containing a constant amount of 10 % Tween 80 and varying ratios of vitamin E to sesame oil (1:10, 1:5 and 10:10) was analysed and presented in Fig. 2. The rheology of emulsions is influenced by numerous factors such as the volume fraction of the dispersed phase, the constituent phases, droplet size and distribution, inter-droplet interactions, droplet flocculation, the type and thickness of the surfactant layer and the ionic strength of the continuous phase [74].

The study showed that the ratio of vitamin E to sesame oil significantly affected the viscosity of the nanoemulsion. At low shear rates of 3.29 s^{-1}, the viscosity showed a sharp decrease due to droplet deformation and the breaking of weak hydrogen bonds within the bulk droplets. Conversely, at higher shear rates the viscosity became independent of shear rate and remained constant over all rates applied (Fig. 2). However, a clear trend was observed for nanoemulsions containing a 1:10 ratio of vitamin E to sesame oil. The viscosity of these emulsions showed a dependence on shear rate up to 57.4 s^{-1}, exhibiting dilatant or thickening flow behaviour. This increase in viscosity at low shear rates (1.27 s^{-1}) was probably due to droplet coagulation, which can increase viscosity [75].

As shown in Fig. 2, nano-emulsions prepared with 20 % Tween showed similar flow behaviour to those prepared with 10 % Tween80. At low shear rates (1.61 s^{-1}) the viscosity decreased abruptly and remained constant with increasing shear rate. The viscosity was not dependent on the shear rate and increasing the shear rate had little effect on the viscosity of the nanoemulsion produced. However, the flow behaviour of the sample prepared with a 1:10 ratio of vitamin E to sesame oil was different. Initially the viscosity of the nanoemulsions was 0.006 Pa and up to a shear rate of 2.04 s^{-1} the viscosity was dependent on the shear rate, indicating a dilatant or thickening behaviour with shear. The increase in viscosity at low shear rates (1.27 s^{-1}) was probably due to the smaller size of the nanoemulsion droplets, as a smaller particle size (at constant volume fraction) results in a higher viscosity of the system [76].

At low shear rates (1.27 s^{-1}), the viscosity of the prepared nanoemulsion showed a sharp decrease and then remained constant with increasing shear rate, indicating a shear thinning behaviour. However, a distinct flow behaviour was observed for the nanoemulsion containing vitamin E and sesame oil at a ratio of 10:1. The initial viscosity of the nanoemulsion was 0.011 Pa-s^{-1} and was found to depend on the shear rate up to 2.04 s^{-1}, indicating a dilatant or thickening behaviour. The viscosity of the oil phase plays a crucial role

Table 2

Comparison of average pasta tests containing vitamin E Nano-emulsions and controls.

Treatments			Pasta	E.P.E
pH			6.02^{a}	6.01^{a}
Moisture		(%)	10.51^{b}	11.45^{a}
Ash		(%)	0.31^{a}	0.35^{a}
Weight		(%)	76.86^{a}	74.13^{b}
Total solids in water		(%)	4.9^{a}	4.97^{a}
Crude protein		(%)	10.66^{a}	10.57^{b}
Texture		(%)	24.71^{a}	20.86^{a}
Vitamin E in	Dough pasta	(ppm)	0.125^{b}	5.75^{a}
	Remaining in product	(ppm)	0.074^{b}	4.08^{a}
Nano-emulsion efficiency	Extruder step	(ppm)	0.125^{b}	5.75^{a}
	After process	(ppm)	0.074^{b}	4.08^{b}
Color features		L	64.89^{b}	67.68^{a}
		a	6.02^{a}	6.01^{a}
		b	5.277^{a}	4.341^{b}

The treatments with a letter in common do not differ statistically significantly at the probability level of 5 %.

in determining the rate at which non-ionic surfactant molecules migrate from the organic phase to the aqueous phase, and a decrease in oil viscosity can accelerate the movement of surfactant molecules, leading to the formation of smaller droplets [74]. However, a decrease in viscosity was observed for the nanoemulsion over time, with occasional slight fluctuations [77]. Based on the results of the nanoparticle diameter, turbidity, pH, rheological properties and physical stability tests, the nanoemulsion containing 10 % Tween, 1 g vitamin E and 10 g sesame oil was initially selected for E.P.E. However, taking into account the experience of pasta production lines in industrial plants and the suggestions of the authorities, the nanoemulsion containing 10 % Tween 80 and a 10:10 ratio of vitamin E to sesame oil was finally selected. It was observed that the changes in diameter of the nanoemulsion with 10 % Tween 80 were lower than those produced with 20 % and 30 % Tween.

3.6. Physiochemical attributes of E.P.E

3.6.1. Moisture

Based on the results presented in Table 2, a significant difference ($p < 0.05$) in moisture content was observed between the control pasta and the pasta enriched with vitamin E nanoemulsion (E.P.E.). The E.P.E. sample had a higher moisture content (11.45 %) than the control sample (10.51 %). Elevated moisture content can adversely affect the appearance of pasta and lead to surface cracking [78]. In addition, exceeding a certain moisture threshold can promote microbial growth. The robust gluten-starch-carrageenan network formed during the drying process at high temperatures plays a key role in achieving low moisture content in pasta. These hydrophilic compounds wrap tightly around the dried pasta strands, reducing the surface area available for water absorption during cooking. This idea is supported by the firmer texture observed in pasta dried at 88 °C compared to pasta dried at 60 °C [79]. The gluten-nanoemulsion network formed in the E.P.E. pasta was apparently strong enough to withstand higher moisture contents while maintaining the desired texture.

3.6.2. Weight analysis after baking pasta

The weight after cooking of the E.P.E. pasta was significantly lower ($p < 0.05$) compared to the control sample, with mean values of 74.13 % and 76.86 %, respectively (Table 2). This reduction in weight after cooking in the pasta enriched with the vitamin E nanoemulsion can be attributed to the rheological properties of the nanoemulsion used (containing 10 % Tween 80, 10 g vitamin E and 10 g sesame oil). The decreasing behaviour of the nanoemulsion could have influenced the texture of the pasta, possibly leading to a compression of its structure and consequently a decrease in weight after cooking. However, the incorporation of the nanoemulsion (10 % Tween 80, 10 g vitamin E and 10 g sesame oil) into the semolina dough could have contributed to a firmer and more cohesive texture. This improvement in texture could be explained by the strengthening and increased elasticity of the semolina network due to its interaction with the nanoemulsion. In particular, semolina flour has a granular structure that exhibits elasticity upon hydration, and a firmer texture is often desirable in pasta production [80].

3.6.3. Protein and vitamins of pasta

The addition of nano-emulsion significantly affected the protein and vitamin E content of E.P.E. ($p < 0.05$) (Table 2). The pasta produced with nanoemulsion had a protein content of 10.57 %, which was slightly lower than that of the control pasta (10.66 %) [81]. However, the amount of vitamin E in E.P.E. was significantly higher than in the control sample, with 5.75 ppm of vitamin E compared to 0.125 ppm ($p < 0.05$). The vitamin E content in the nano-emulsion fortified uncooked pasta was 6 ppm, which is within the maximum limit of 2.5 ppm for mineral and vitamin supplements in 35 g of uncooked pasta set by the Australian and New Zealand Standards Organisation [82].

3.6.4. Vitamin E content of baked pasta

Based on our results, the pasta enriched with the vitamin E nanoemulsion (E.P.E) retained a significantly higher amount of vitamin E after processing compared to the control pasta (4.08 ppm vs. 0.074 ppm) ($p < 0.05$). It's worth noting that pasta flour (semolina) naturally contains trace amounts of vitamin E, which are further reduced during thermal processing and baking by the consumer. However, the addition of the nanoemulsion containing vitamin E to the pasta dough seemed to help preserve the vitamin E content [83].

3.6.5. Nano emulsion efficiency

Based on the Australian and New Zealand Standards Organisation's maximum level for 100 kg of pasta, 7.15 g of vitamin E was used to produce the nanoemulsion. The efficiency of the nanoemulsion was found to be 84.2 % after the extrusion process and 71.6 % after the drying process. Consequently, the amount of vitamin E lost during the extrusion process was calculated to be 1.4 ppm (for 100 kg of pasta), while for the pasta after the extrusion step it was 1.67 ppm. The loss of vitamin E relative to the initial amount was determined to be 2.84 ppm. Finally, the vitamin E content was calculated to be 6 ppm in the extruded pasta and 4.31 ppm in the baked pasta.

3.6.6. Color

The L* value of E.P.E. was found to be superior to that of the control sample (Table 2). The acceptable colour of pasta is amber yellow and it should be free from black spots caused by brown and light bran products without the addition of external colouring agents. The evaluation of the sample prepared with the nano-emulsion containing vitamin E showed that it was lighter in colour than the control sample. This was attributed to the presence of vitamin E, which has an amber colour. The main colour of pasta is also clear yellow [84]. The superior lightness-darkness of the produced pasta sample was observed over the control sample [81,83]. The b* parameter of the pasta showed that the nano-emulsion containing vitamin E had a lower blue-yellow value than the control sample. This lower value of the yellowness factor could be due to the turbidity of the nano-emulsion [79].

3.6.7. Sensorial analysis

It's important to stress that sensory evaluation plays a key role in food product development as it reflects consumer preferences and expectations. In this study, a panel of trained judges conducted sensory evaluations, rating samples based on appearance, aroma, texture and taste. The results of this evaluation provide valuable insights for product improvement and guide future research directions. Based on the sensory evaluation, the E.P.E. sample was deemed acceptable and received high scores in three categories (Fig. 3). However, the control sample received a low score due to its chewing gum-like texture, while the sample containing the vitamin E nanoemulsion received a lower score for taste.

4. Conclusion

The successful use of vitamin E nanoemulsions in the development of new pasta formulations represents a significant advance in the field. The identified optimal nanoemulsion, characterised by a 10 % Tween concentration and a 1:10 ratio of vitamin E to sesame oil, exhibited exceptional physical stability over a 30-day storage period. This nanoemulsion not only enriched the pasta with essential nutrients, but also resulted in increased moisture content and reduced weight compared to the control, indicating potential improvements in both nutritional value and product characteristics. In addition, the addition of the vitamin E nanoemulsion enhanced the visual appeal of the pasta by improving its colour, potentially increasing consumer interest and acceptance. Given the growing market interest in food products enriched with nanoemulsions containing bioactive substances and the promising research results, there is a clear opportunity to introduce nanoemulsion-enriched pasta to the public. By enriching pasta with essential vitamins such as vitamin E through nanoemulsion methods, we can address nutritional deficiencies and promote health, while offering consumers a convenient and enjoyable dietary option. In conclusion, the successful application of nanoemulsion technology to enrich pasta holds great promise for meeting the nutritional needs of individuals and improving overall health. With further development and commercialisation efforts, fortified pasta could become a valuable addition to the market, contributing to the well-being of consumers worldwide.

Ethics statement

Our research paper maintains the highest ethical standards. We obtained informed consent from human participants. Data collection and analysis were transparent and in line with guidelines. We have disclosed funding sources and declare no conflicts of interest. Our research upholds academic integrity, acknowledging all contributions appropriately.

Data availability statement

Data will be made available on request.

CRediT authorship contribution statement

Wrya Mohamadyan: Writing – original draft, Methodology. **Shima Yousefi:** Writing – review & editing. **Weria Weisany:** Writing – review & editing, Writing – original draft, Supervision, Project administration.

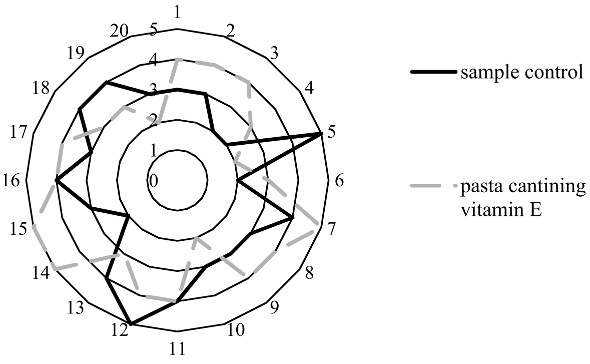

Fig. 3. The sensory evaluation of pasta produced with Vitamin E and the control sample was conducted by 20 judges. The scoring was based on taste, aroma, and the pleasure of chewing after cooking the pasta without salt, oil, and sauce. Scores ranged from 1 (lowest) to 5 (highest). Three judges who rated the produced sample lower in the test did so based on taste and flavor.

Declaration of competing interest

The authors declare that there are no known financial conflicts of interest or personal relationships that could have potentially influenced or biased the outcomes reported in this paper.

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