LWT - Food Science and Technology 155 (2022) 112976

Contents lists available at ScienceDirect

LWT

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

Fabrication and characterization of zein nanofibers integrated with gold nanospheres

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ARTICLE INFO

Keywords:
Electrospinning
Gold nanospheres
Surface coating
Microbial safety
Sea bream

ABSTRACT

The synthesis, characterization, and coating effect of zein nanofibers integrated with gold nanospheres were studied. Zein solution including ethanol/liquid gold nanospheres (80/20 v/v) was prepared to produce complex nanofibers. Dielectric constant (ε'), dielectric loss factor (ε''), and loss tangent ($\varepsilon''/\varepsilon'$) values of feed solutions were evaluated at 300 and 3000 MHz, and electrical conductivity was measured (808.67 ± 2.082 μs/cm). SEM image showed a bead free structure of the gold-zein complex. Zeta potential and DLS measurements (translational diffusion coefficient, hydrodynamic radii, polydispersity index, major axis) were done by dispersing nanofibers in ethanol or water. The stability of nanofiber dispersed in ethanol was higher (+41.73 mV) compared to that of dispersed in water (+5.1 mV). Molecular characterization by FTIR confirmed the formation of the zein-gold complex through a nitrogen-gold coordination bond and electrostatic interactions. The gold-zein nanofibers coating in sea bream fillets reduced 17.8% of total mesophilic bacteria (TMAB) compared to the uncoated group ($p < 0.05$) after 8 days of observation. The highest changes in ε' for uncoated (57.65%) and coated samples (14.84%) during storage showed the positive impact of nanofiber treatment. These results showed that zein nanofibers with gold nanospheres could be potentially used as antimicrobial layer for food products.

1. Introduction

The enrichment of food matrices with antioxidants, carotenoids, probiotics, bioactive compounds, and antimicrobial compounds has gaining interest in food industry. Spray-drying, spray-cooling, fluid-bed coating, extrusion, and emulsification followed by solvent evaporation are some of the example techniques that are available for the encapsulation of these compounds within a wall material (García-Moreno, Mendes, Jacobsen, & Chronakis, 2018; Mendes & Chronakis, 2019; Wei, Sun, Dai, Zhan, & Gao, 2018). Although spray drying is one of the most widely used process, it requires heating and the size of the encapsulates obtained in this process are large (10–100 μm). Emulsification-evaporation, emulsification followed by solvent evaporation, is another common technique for preparing nanoparticles <500 nm (Wei et al., 2018). Electrohydrodynamic techniques such as electrospinning and electrospraying are also emerging as alternative encapsulation techniques, since they are cost-effective and functional nanofibers, nanocapsules, and nanoparticles with very low size can be produced (García-Moreno et al., 2018; Mendes & Chronakis, 2019; Miguel et al., 2019). Therefore, nanotechnology applications related to food industry and food science are becoming people's next generation food applications. For instance, nanofibers that integrated with curcumin and nisin (Meral et al., 2019) or nano-probiotics (Ceylan, Meral, Karakaş, Dertli, & Yilmaz, 2018) successfully limited the microbial growth in fish meat and effectively improved the acceptability during cold storage. In the last decade, emulsification systems like nanoemulsions have been also utilized in order to inhibit pathogenic microorganisms, which could be host in fresh or processed fish fillets (Meral et al., 2019) Therefore, some coating treatments, could be called as nanocoating (<1000 nm) as well, have been newly applied for fish meat products.

Polymer-based nanomaterials have an important role in delaying microbiological spoilage, physical and chemical deterioration in foods. Zein, a plant protein and biodegradable polymer extracted from corn is

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https://doi.org/10.1016/j.lwt.2021.112976
Received 21 September 2021; Received in revised form 6 December 2021; Accepted 13 December 2021
Available online 16 December 2021

non-toxic, widely available, and biocompatible. Therefore, more researchers have been attracted to observe functionalities of zein-based nanofibers (Aman Mohammadi et al., 2021; Ansarifar & Moradinezhad, 2021; Turasan & Kokini, 2017). In addition to nanofibers, zein based nanoparticles has been also employed to encapsulate bioactive ingredients such as ω-3 fatty acids, antioxidants, and carotenoids (García-Moreno et al., 2018; Wei et al., 2018, 2020). For instance, the bioavailability of folic acid, vitamin D_3, curcumin, resveratrol, and beta-carotene can be improved using zein nanomaterials (Afonso et al., 2020; Kasaai, 2018; Silva, Torres-Giner, Vicente, & Cerqueira, 2021). Green tea catechins has been also effectively encapsulated in zein nanoparticles with a mean diameter of 157 nm by electrospraying. Bhushani, Kurrey, and Anandharamakrishnan (2017) indicated that the encapsulated green tea catechins were more stable under *in vitro* gastrointestinal stability and easier to permeate when compared with controls. Therefore, a similar concept can be applied to zein based nanofibers to improve the functionalities of nanomaterials.

Recently, the use of antimicrobial agents or nanoparticles to enhance the physical, chemical, antimicrobial properties of zein-based materials has received great interest (Aytac et al., 2020; Moradkhannejhad, Abdouss, Nikfarjam, Mazinani, & Heydari, 2018). Moreover, zein can be used to encapsulate metal nanoparticles as they act both as reducing as well as capping agents and provide chemical stability (Mahendia, Tomar, & Kumar, 2010). Zein-gold complex nanoparticles have been proved to improve the mechanical, functionality, and degradability of gold nanoparticles (Puthiyaveetil Yoosaf et al., 2019; Turasan, Cakmak, & Kokini, 2019).

Knowledge of a material's dielectric property can help determine its molecular structure. A change in molecular structure can lead to a change in the dielectric properties of material. A dielectric material (host material) containing filler particles with sizes of 10–100 nm in a well-dispersed state constitutes a nanodielectric system (Krishnaswamy & Orsat, 2015). Zein, is a dielectric material in which nanoparticles can be dispersed. Therefore, zein solution with nanoparticles can be considered as nanodielectric system. ε' is defined a material's ability to store electrical energy. ε'' is defined as an index of the energy dissipation characteristics (Icier & Baysal, 2004; Krishnaswamy & Orsat, 2015). The chemical composition especially water, salt contents and other minerals affects dielectric properties. Organic components make sample dielectrically inert (Sosa-Morales, Valerio-Junco, López-Malo, & García, 2010). Generally, gold nanospheres are obtained by reduction of gold salts by using reducing agents like polyvinylpyrrolidone. Therefore, it can be important to know ε'' values of the feeding solution. Another dielectric value is the $\varepsilon''/\varepsilon'$, which is related to the material's ability to penetrate and dissipate electrical energy as heat (Krishnaswamy & Orsat, 2015).

Maintaining the quality of seafood products is important during the storage period. As known, microbial growth of fish is related to the deterioration of chemicals in fish. Microbiological, chemical, physical, or sensory analyzes can be performed to predict fish deterioration. Since seafoods are dielectrically active, knowledge of dielectric properties, is also essential for the characterization of fish quality (Nguyen, Ahmad, & Jayanath, 2020). Dielectric properties can be considered a fast and non-invasive way to monitor different parameters of fishes (Franceschelli, Berardinelli, Dabbou, Ragni, & Tartagni, 2021). The dielectric properties of fish muscle may alter during the storage as tissue components degrade over time. In this respect, changes in the dielectric properties of food products can also be used for the evaluation of their spoilage degree (Çetinkaya, Altay, & Ceylan, 2021a; Sant'Ana, Soares, & Vaz-Pires, 2011).

Pure gold is safe to eat and has been used as a food decoration in food industry. Gold (E175) is approved as a food additive in the European Union. (EFSA ANS Panel, 2016; Panyala, Peña-Méndez, & Havel, 2009). In 2016, the EFSA Panel noted that no data on subchronic, chronic toxicity or genotoxicity of elemental gold are available. In the same report it has been stated that the daily mean intake of gold from the regular diet can be estimated to be in the range of 0.01 μg/kg bw/day to 0.02 μg/kg bw/day. Dietary exposure from the food additive and the regular diet would lead to a mean intake for children from 0.03 to 0.10 μg/kg bw/day. On average, dietary exposure from the food additive would represent around 30% of the total exposure (from both the food additive and the regular diet) (EFSA ANS Panel, 2016). In this regard, there is growing interest in the investigation of gold metal by nanotechnological applications due to its biocompatibility, ease of surface functionalization, and optical properties. Gold particles can be synthesized in different dimensions and shapes such as nanoflowers (Penders, Stolzoff, Hickey, Andersson, & Webster, 2017), nanotubes (Liu, Zhu, Weng, Li, & Zhao, 2020), nanostars (Lin, Sun, Kong, & Lin, 2021), nanocapsules (Singh, König, & Jaisval, 2018), nanorods, nanocages, and nanospheres (Cobley, Chen, Cho, Wang, & Xia, 2011). According to recent studies, gold in different morphologies showed antibacterial properties (Borzenkov et al., 2020; Okkeh et al., 2021; Omerović et al., 2021; Wang et al., 2020; Zheng, Setyawati, Leong, & Xie, 2017). It has been reported that an increase in the membrane tension of bacterial cells was caused by the adsorption of gold nanoparticles leading to mechanical deformation of the membrane, and eventually cell rupture and death (Linklater et al., 2020; Okkeh et al., 2021). Boatemaa, Ragunathan, and Naskar (2019) determined the detection and inhibition of *S. typhi* strains via gold nanoparticles with a size of 40–60 nm. Although nanoparticles within the size range of 80–100 nm are unable to freely translocate across the bacterial cell membrane, they still can inactivate bacteria (Okkeh et al., 2021). The ultra-small gold nanoclusters (<2 nm) can easily traverse into the cell wall pores to kill bacteria (Zheng, Setyawati, Leong, & Xie, 2021). Suganya et al. (2017) determined antibacterial and antibiofilm potential of zein with gold nanoparticles (50–80 nm along with different shapes, such as nanospheres and nanoplates), by microtiter plate technique and agar well diffusion method. They have stated that presence of zein molecules enhance the binding capacity between gold nanoparticles and outer membrane components of gram positive (*B. pumilus* and *B. subtilis*) and Gram negative (*S. sonnei* and *P. aeruginosa*) bacteria. In this way, gold nanoparticles create electronic effects due to their size, reduced surface area, then penetrate inside the bacteria to cause cell death.

These above-mentioned studies indicated that gold particles could be ideal materials to prevent microbial growth in food products. However, as stated before antibacterial mechanism action for gold depends on its size and bacterial strain to which they are applied. Therefore, the use of gold (15 nm) in zein nanofibers, could be an effective strategy to inhibit microbial spoilage in fish meat. The goal of the present study was to fabricate zein-gold nanofibers conjugate and to investigate its functionalities on sea bream fillets. Electrical conductivity, and dielectric values (i.e. ε', ε'', $\varepsilon''/\varepsilon'$) of zein-gold particle solution was investigated to evaluate the effect of incorporated gold nanosphere on its electrospinnability. Fabricated nanofibers were characterized by their zeta potential, hydrodynamic radii, polydispersity index, molecular, and morphological properties. Finally, microbiological deterioration using specific parameters such as TMAB counts and dielectric changes on sea bream fillets were evaluated to investigate the coating effect of complex nanofibers.

2. Material and method

2.1. Material

Zein from maize, purity of 98%, molecular weight (Mw) ≃22–24 kDa, was purchased from Sigma-Aldrich Inc. (Saint Louis, MO, USA). Gold nanospheres (15 nm, NanoXact, 0.05 mg/mL in water) coated with polyvinylpyrrolidone was kindly received from NanoComposix Inc. (San Diego, CA, USA). Plate count agar (VM888763 930) was purchased from Merck & Co., Inc. (Kenilworth, NJ, USA). Technical ethanol (96%) was obtained from Aven Chemical (Tusba, Van, Turkey). Distilled water was used throughout all experiments. Gilthead sea bream samples (*Sparus* *aurata*) were purchased from Metro market (Kagithane, Istanbul, Turkey), filleted and deskinned.

2.2. Method

2.2.1. Feed solution properties

Ethanol-liquid gold nanosphere solution (80:20 v/v) was used for dissolving zein at concentration of 30 g/100 mL. The mixture was blended for 30 min at 600 rpm using a stirrer to obtain feed solution.

Electrical conductivity of feed solution was measured according to methodology described by Dias Antunes et al. (2017). Measurement was done in triplicate with a unit of microsiemens/centimeter (μs/cm) using WTW LF95 electrical conductivity meter (Wissenschaftlich-Technische Werkstätten GmbH & Co., Weilheim, Germany) at 22 °C.

Dielectric constant (ε′), dielectric loss factor (ε′′) and loss tangent (tan $\delta = \varepsilon''/\varepsilon'$) values of solution was obtained as the methodology described by Krishnaswamy and Orsat (2015), using Network Analyzer instrument (Agilent Technologies E5061B ENA Series, Santa Clara, CA, USA) with Agilent 85070E Option 030 dielectric probe kit (Santa Clara, CA, USA) at room temperature (22 °C). Measurements were performed in triplicate and were investigated at 300 MHz and 3000 MHz frequencies. Software graphs for ε′, ε′′, ε′′/ε′, and cole-cole values are also provided in Supplementary Material S1–S4.

2.2.2. Electrospinning process

The electrospinning process was conducted with Inovenso NE100 (Basaksehir, Istanbul, Turkey) equipped with a syringe pump unit (New Era Pump Systems, Inc., Farmingdale, NY, USA), and an external high voltage supply (Nanofen, Yenimahalle, Ankara, Turkey). The solution was electrospun at a 0.8 mL/h feed rate with an applied voltage of 6 kV and an 18 G needle was utilized. The distance between the aluminum foil covered on the collector and needle was 15 cm.

2.2.3. Structure of nanofibers

Morphology of zein nanofibers integrated with gold nanospheres were examined using FE-SEM (FEI, Quanta Feg 250, Hillsboro, OR, USA) under low vacuum with 50.000 times magnification with a working distance of 8 mm. An accelerating voltage of 10 kV was determined for obtaining secondary electron images. The nanofiber diameter distribution was obtained by measuring 50 random nanofibers ($n = 50$) with the Image J program (National Institutes of Mental Health, Bethesda, Maryland, USA). Furthermore, TEM images were presented in Supplementary Material (S5–S6).

2.2.4. Zeta potential and dynamic light scattering measurements

The zeta potential and dynamic light scattering measurements (DLS) measurements of zein nanofibers integrated with gold nanospheres were performed using Malvern Zetasizer Nano ZS ZEN3600 equipment (Malvern Instruments Limited, Malvern, Worcestershire, UK). Ethanol or distilled water was used as dispersant as performed by Dede & Altay (2021). Nanofibers were dispersed at 0.1 g/100 mL in ethanol using Silent Crusher S (Heidolph Instruments, GmbH & Co. KG, Schwabach, Germany) and in water by magnetic stirrer for 60 s and 180 s, respectively. Then, dispersions were filtrated through a Whatman® Schleicher & Schuell filter paper (Grade 595 ½, 90 mm diameter). Methodology of zeta potential and translational diffusion coefficient was modified from Okutan, Terzi, and Altay (2014). Filtrated dispersions were analyzed using a scattering angle of 173°. Zeta potential analysis was done in disposable folded capillary cell model DTS1070 (Malvern Panalytical Inc., Malvern, Worcestershire, UK). The hydrodynamic radius and polydispersity index analyses were developed from methodologies of Chen, Qiu, Tang, Xing, and Zhao (2021) and Saunders, Noack, Dzombak, and Lowry (2015). Measurements were performed by dropping filtrated samples in the disposable cuvettes.

2.2.5. FTIR spectroscopy

Infrared spectra were obtained at a 4/cm resolution with IRAaffinity-1 FTIR spectrometer (Shimadzu Corp., Tokyo, Japan) equipped with single reflection Attenuated Total Reflectance (ATR). Measurements were done by accumulating 10 scans. The nanofiber sample was measured in transmission mode and the measurement was recorded in the range of 600–4000/cm at room temperature (22 °C).

2.2.6. Coating of fish fillets with nanofibers

After the production of nanofibers on aluminum foil, deskinned sea bream fish fillets were treated and coated by rubbing the samples to nanofibers according to methodology described by Meral et al. (2019a). Thus, the zein nanofibers integrated with gold nanospheres were absorbed by the surface of the fish flesh. Control samples were not coated with nanofiber (untreated). Then, untreated and nanocoated samples were stored at 4 ± 1 °C and analyzed on initial, 1, 2, 3, 4, 7, and 8th days.

2.2.7. Microbiological analysis

TMAB count was determined according to FDA standard protocol (FDA, 2001). For each treatment, 10 g of sample was homogenized for 2 min in a stomacher (IUL Instruments, Barcelona, Spain) with 90 mL peptone water obtained from 0.1 g/100 mL peptone from meat. Serial dilutions from 10^1 to 10^7 were prepared. Diluted samples were placed on plate count agar and then incubated at 35 °C for 48 h ($n = 12$).

2.2.8. Dielectric properties of fish samples

Dielectric analyses of fish samples were performed from a methodology as described by Çetinkaya, Altay, and Zafer (2021). Distilled water (10 mL) was added to 1 g of each sample, and it was homogenized for 50 s with Silent Crusher S homogenizer (Heidolph Instruments GmbH & Co., Schwabach, Germany) at 75000 rpm. Network Analyzer (Agilent Technologies E5061B ENA Series, Santa Clara, CA, USA) with a 200 mm probe (Model/Part Number 85070-20037, Santa Clara, CA, USA) used to determine ε′ (300 MHz) and ε′′ (3000 MHz) at room temperature.

2.2.9. Statistical analysis

Collected data was conducted using IBM SPSS Statistics 28.0 (IBM Corp.,Armonk, NY, USA) program to determine solution properties and the TMAB, ε′, ε′′ differences of nanocoated and uncoated group samples during the 8-day experimental period. Mean values of the samples were further analyzed using Tukey's multiple range comparison test, whereas a significant ($p < 0.05$) main effect was found.

3. Result and discussion

3.1. Feed solution properties

The preparation, characterization, and surface coating of zein nanofiber containing gold nanosphere is shown in graphical abstract. The electrical conductivity, i.e. ε′, ε′′, and ε′′/ε′, of the feeding solution is given in Table 1. Electrical conductivity of the feed solution was found to be 808.67 ± 2.082 μS/cm. Wang, Zhao, Barker, Belton, and Craig (2019) reported a higher electrical conductivity (1.43 ± 0.01 mS/cm) of zein (30 wt%) in water/ethanol (30/70 v/v). Liquid gold nanosphere in zein solution might decrease the solution's electrical conductivity, leading to

Table 1
Electrical conductivity, ε′, ε′′, and ε′′/ε′ values of zein (30 g/100 mL) solution containing ethanol/liquid gold nanospheres (80/20 v/v).

Electrical Conductivity (μs/cm)	808.67 ± 2.082	
Dielectric Properties	300 MHz	3000 MHz
Dielectric constant (ε′)	78.64 ± 0.115	76.92 ± 0.075
Dielectric loss factor (ε′′)	1.06 ± 0.057	11.08 ± 0.057
Loss tangent (ε′′/ε′)	0.0135	0.145

a decrease in the number of soluble species with charge-carrying ability, likely because of the interaction of gold nanospheres with zein. The electrospinning process requires the transfer of electrical charge to the polymer droplet at the injection needle to the elongated fluid jet. The conductivity of the feed solution indicates the capacity of electrons to move in the solution. Therefore, electrical conductivity may influence the electrospinnability of the feed solution (le Corre-Bordes, Hofman, & Hall, 2018; Moradkhannejhad et al., 2018; Okutan et al., 2014). Electrical conductivity value can be evaluated as essential parameter for the formation of smooth fibers. A high conductivity (>10 mS/cm) of feed solution may result in necklace, beaded, nonuniform, ribbon-like and branched jet formation. For instance, soy protein isolate solution showed electrical conductivity of 13.06 ms/cm but did not form nanofibers due to instability in jet formation (Aslaner, Sumnu, & Sahin, 2021; Seethu et al., 2020). Therefore, a low electrical conductivity is preferable for nanofiber formation since the feeding solution is subjected to weaker electrical forces (Moradkhannejdah et al., 2018). A decrease in conductivity of solution could reduce beading because of less stretching of the polymer jets, which tends to produce thicker nanofibers (Aslaner et al., 2021; Facchi, Souza, Almeida, Bonafé, & Martins; Moradkhannejhad et al., 2018). However, 16% pullulan feed solution had a very low conductivity (0.06 mS/cm) that was not able to form desirable nanofibers (Seethu et al., 2020). These studies indicated that electrical conductivity depends on polymer concentration and type, affecting the nanofiber structure. Electrical conductivity of the feeding solution, i.e. 808.67 ± 2.082 μS/cm produced smooth gold-zein nanofibers.

The dielectric properties (ε', ε'', and $\varepsilon''/\varepsilon'$) of solutions may depend on factors such as water content, frequency, temperature, chemical composition, and the structure of the material (Icier & Baysal, 2004; Samuel & Trabelsi, 2012; Sosa-Morales et al., 2010). Furthermore, the size and concentration of metal nanoparticles remarkably affect the dielectric properties of polymer/metal materials (Zare & Shabani, 2016). In this regard, the dielectric properties of feed solution influenced the structure and diameter of the nanofibers. Electrospinning of feed solution with higher ε' would give a stronger jet split capability in the high electric field (Jørgensen, Qvortrup, & Chronakis, 2015; Mendes & Chronakis, 2019). Therefore, a feed solution with relatively high ε' can facilitate the formation of thin fibers (Facchi, Souza, Almeida, Bonafé, & Martins, 2021). Pure water shows a relatively high ε' of 80.4 at 20 °C due to the presence of dipole moments. Thus, H^+ and OH^- bonds in water tends to polarize, with the influence of an applied electrical field. The ε' for gold nanoparticles by the Turkevich method ranges from 78.71 to 40.41 (Krishnaswamy & Orsat, 2015). Zein solution containing ethanol/liquid gold nanospheres (80/20 v/v) showed in line ε' values, i.e. 78.64 ± 0.115 and 76.92 ± 0.075. These values were slightly lower than ε' of water. Nguyen et al. (2020) reported that the higher concentration of proteins resulted in the reduction of available moisture content, where the ε' was also decreased. Table 1 shows that the higher frequency of 3000 MHz resulted in higher dielectric properties than that obtained at lower frequency of 300 MHz. The frequency dependency of dielectric properties was in accordance with previous studies (Isik, Altay, & Capanoglu, 2018; Krishnaswamy & Orsat, 2015). Krishnaswamy and Orsat (2015) stated that the ε'' and $\varepsilon''/\varepsilon'$ increased with an increase in frequency (200 MHz–20 GHz) which could also increase charge mobility within the polymer matrix and ion diffusion in the direction of the electric field.

3.2. Morphology of nanofibers

SEM image of zein nanofibers integrated with gold nanosphere is shown in Fig. 1. The average diameter of zein nanofibers was 438 nm. Minimum and maximum fiber diameters were 85 nm and 789 nm, respectively. Fig. 1 shows that gold particles are well distributed on the surface of fibers. Yilmaz et al. (2016) successfully produced curcumin-loaded zein nanofibers (less than 350 nm in diameter) and did not find remarkable morphological difference between zein and

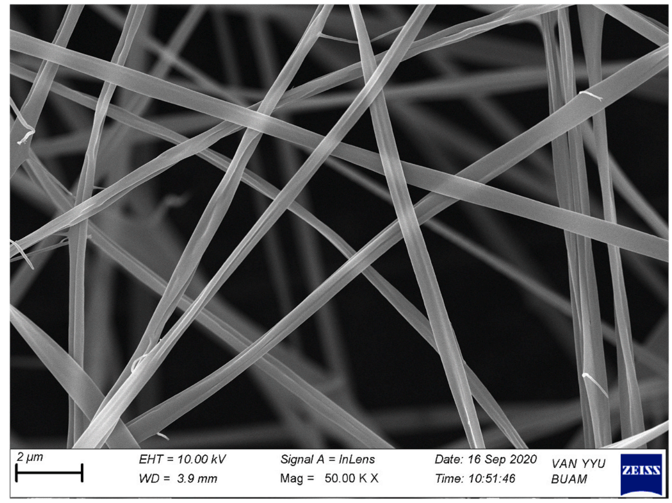

Fig. 1. SEM image of zein nanofibers integrated with gold nanospheres. Scale bar is 2 μm. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

curcumin loaded nanofibers. It has been reported that conductivity and fiber diameter showed an inverse relationship, where higher conductivity of feeding solutions produced thinner diameter fibers owing to increasing electrostatic force and jet elongation (Aslaner et al., 2021; Uygun, Yildiz, Sumnu, & Sahin, 2020). The electrospinning of solutions that had a low electrical conductivity resulting in a larger fibers formation because of less stretchability (Facchi et al., 2021). Thus, higher diameter of zein-gold nanofibers (438 nm) could be attributed to the presence of gold nanosphere which reduced the electrical conductivity properties in the feeding solution.

3.3. Zeta potential and dynamic light scattering measurements

Stability of electrospun nanofibers was evaluated by zeta potential measurements. Dispersions with zeta potential values at above +25 mV or below −25 mV can be considered as a stable colloidal dispersion (Okutan et al., 2014). Nanofiber dispersed in ethanol showed a relatively high zeta potential, i.e. +41.73 ± 3.1 mV, indicating the nanofibers can be well dispersed for a relatively long time. However, changing the solvent to water reduced the value to +5.1 ± 2.49 mV, indicating the occurrence of particle-particle attractive interactions leading to aggregation since zein is a hydrophobic protein (Table 2). According to Yilmaz et al. (2016) zeta potential values of 2.5% and 5% curcumin loaded zein (based on the weight 30% zein powder) nanofibers were calculated as −24.1 ± 2.44 mV and −29.8 ± 2.16 mV, respectively. It has been reported that more negative charges were carried out by increasing curcumin concentration in zein nanofibers

Table 2
Zeta potential, translational diffusion coefficient, hydrodynamic radius, polydispersity index, and major axis values of dispersed (0.1 g/100 mL) electrospun zein nanofibers.

	Dispersed in ethanol	Dispersed in water
Zeta potential (mV)	+41.7 ± 3.1	+5.1 ± 2.5
Translational diffusion coefficient (D) ($\mu m^2/s$)	1.85 ± 0.09	2.03 ± 0.03
Hydrodynamic radius of aggregated nanofiber (nm)	221 ± 9.94	242 ± 3.03
Polydispersity index of aggregated nanofiber	0.36 ± 0.02	0.22 ± 0.01
Calculated major axis for nanofiber in a solvent (nm)	693.07	629.78

Data represent average amounts ± standard deviations of three replications.

(Yilmaz et al., 2016). We observed similar phenomenon that oppositely charge of zein, and gold particles resulted in attractive interactions between zein and gold nanospheres, suggesting an increase of the thickness of charged layer on the nanofiber surface due to increasing positive charge.

The translational diffusion coefficient of complex nanofibers in ethanol and water dispersions was 1.85 $\mu m^2/s$ and 2.03 $\mu m^2/s$ respectively. The results showed that the sample dispersed in the water had a 9.73% higher translational diffusion coefficient than the one dispersed in ethanol. Monaghan and White (1936) showed that slightly decreased mobility of red cells by adding proteins indicated absorbed water molecules (or hydrated) on the surface of red cells. In addition, Isik et al. (2018) reported that a higher diffusion coefficient indicated a higher mobility of the polymer in the suspension. Therefore, high mobility might indicate low absorption of water (or ethanol) on the surface of a polymer in the suspension. Similarly, higher translational diffusion coefficient of zein nanofibers in water may indicate lower water absorption on the surface of nanofibers compared to zein nanofibers in ethanol.

Hydrodynamic radii of nanofibers dispersed in ethanol and water were 220.5 ± 9.94 nm and 242 ± 3.03 nm, respectively. The higher hydrodynamic radii of nanofibers in water could be attributed to hydrophobic character of zein. Zhong and Jin (2009) reported that the size of zein-based nanoparticles are typically found between 100 and 200 nm. It was observed that the increase of zein nanoparticle diameter was followed by decreasing ethanol concentration. Therefore, larger particles would be formed with a relatively high-water concentration. Furthermore, shear rate and polymer concentration may also affect the size (Zhong & Jin, 2009). It must be emphasized that the values of hydrodynamic radii obtained from the DLS are not the nanofiber diameter. This is due to the fact that the hydrodynamic radius measurement from DLS follows the theory of Brownian motion and calculates the hydrodynamic radius assuming a spherical particle (Pecora, 1985; Saunders et al., 2015). Despite the inaccuracy of the hydrodynamic radius measurement for fiber suspensions in water or ethanol, the values can provide an insight about relative extent of aggregation since the solution ionic strength and polymer type may affect aggregation state (Saunders et al., 2015).

Polydispersity index of zein/gold complexes in ethanol, i.e. 0.36, and in water, i.e. 0.22, showed a multimodal size distribution. Similarly, the polydispersity index of zein dispersions was earlier found to be in the range of 0.25–0.64 (Giteru, Azam Ali, & Oey, 2021). The multiple modal peaks could be attributed to the size difference of gold nanospheres (15 nm) and zein protein. This size distribution could confirm the existence of supramolecular nanostructures in dispersion (Chen et al., 2021).

To calculate major axis for nanofiber in a water and ethanol solvents, first F_D value was measured. equation (1) was given as below:

$$F_D = log\rho + 0.312 + \frac{0.565}{\rho} - 0.1 \Big/ \rho^2 \tag{1}$$

The equation was provided for long rods from Supporting information of Arenas-Guerrero et al. (2018). (ρ) is the length to dimeter ratio of the cylindrical shaped particle. According to Yabuki, Motonobu, and Fathona (2017), length of short nanofibers from cellulose acetate with silica particles were 52.4-15.3 μm. If the length would be taken as 52.4 μm, the aspect ratio (length/diameter) would be ~120 when considering the average diameter of zein nanofibers was 438 nm. On the other hand, Iwamoto, Lee, and Endo (2014) prepared cellulose nanofibers with aspect ratios varying from 30 to 300. These aspect ratios were reported for nanofibers as it is, not in a solution. Therefore, the aspect ratio of zein nanofibers with gold was taken as 200 to calculate F_D which was found as 2.62. After calculating F_D, then the major axis of nanofibers (L) in a solvent was calculated by using following equation (2):

$$D = \frac{k_B T}{3\pi\eta L} F_D \tag{2}$$

D is the translational diffusion coefficient obtained from DLS measurement, k_B is the Boltzman constant (1.38×10^{-23} kg m^2/s^2.K), T is temperature (25 °C + 273.15 = 298.15 K), η is the viscosity of ethanol (0.8911×10^{-3} Pa s) (Tanaka, Yamamoto, Satomi, Kubota, & Makita, 1977) or water ($0,8937 \times 10^{-3}$ Pa s) (Perry, 1950) at 25 °C. Using equation (2), major axis values were calculated as 693.07 and 629.78 nm in ethanol and water solvents, respectively (Table 2).

3.4. Molecular characterization by FTIR spectroscopy

The infrared spectrum of gold nanosphere, pure zein nanofiber, and zein nanofiber containing gold nanospheres is shown in Fig. 2. The -OH stretching vibration observed around at 3291 cm^{-1} and C-H stretching from CH_2 and CH_3 functional groups revealed at 2873, 2956 and 2928 cm^{-1}. The characteristic peak observed at 1643 cm^{-1} (amide I) is related to the C=O stretching of peptide chain, while 1537 cm^{-1} (amide II) was related to the N-H in plane vibrations. 1236 cm^{-1} (amide III) and 1444 cm^{-1} were related to C-N stretching vibrations. These results were in accordance with previous studies (Dashdorj et al., 2015; Pérez-Guzmán & Castro-Muñoz, 2020; Yilmaz et al., 2016). Amplitude of peak with wavenumber between 3400 and 3200 cm^{-1}, representing mainly OH groups observed in spectrum. For zein-based materials, peaks can be observed at a range of 3294–3350 cm^{-1} which corresponds to the -OH functional groups, while the peaks identified between 2968 and 2922 cm^{-1} correspond to C–H groups. 1538–1546 cm^{-1} for N–H, 1649–1653 cm^{-1} for C=O, and 1428–1451 cm^{-1} for C–N has also been previously observed (Dashdorj et al., 2015; Pérez-Guzmán & Castro-Muñoz, 2020; Yilmaz et al., 2016).

It can be seen from Fig. 2 that the spectrums of pure zein nanofiber and zein nanofiber with gold showed similar trend basically. However, with a closer look, important differences were clearly detected (Fig. 3 and Fig. 4). For instance, after the encapsulation of gold nanospheres, similar characteristic high C=O stretching peaks (a) observed at 1643 cm^{-1} and 1632 cm^{-1} with a decreased transmittance. This could be attributed to the interaction of amide functional groups with gold nanospheres. Similarly, in a study by Puthiyaveetil Yoosaf et al., 2019), these peaks were observed at 1638 cm^{-1} for zein film with gold nanoparticles and at 1644 cm^{-1} for gold nanoparticle caped zein ligand. Authors stated that decreasing of C=O peaks attributed to the distribution of charge on –N–C=O moiety (Puthiyaveetil Yoosaf et al., 2019). In amide I and amide II region, new shoulders (b) observed with C=O carbonly stretch vibrations. These shoulders were detected in gold nanosphere spectrum but not in pure zein.

N-H functional groups play an important role in the stabilization of zein nanofibers. Dias Antunes et al. (2017) have stated that lower wave number in amide regions show better structural stability, with an increased hydrogen bonding interaction occurred at the N-H group. As can be seen from Fig. 3, a decrease of transmittance and shifting (from 1649 cm^{-1}–1643 cm^{-1}) in N-H vibration was observed for the spectrum of zein nanofiber with gold, compared to pure zein nanofiber. These differences could be attributed to increasing molecular weight after the attachment of gold nanospheres to nitrogen atoms which also affected the vibration intensity of the N-H bond (Wei, Sun, Qian, Ye, & Ma, 2009).

Fig. 4 shows that there were disappeared bands for pure zein (c, d, e), which changed to the stretching peaks with cleaving and elongation for zein nanofiber with gold. For instance, bands at 1452 cm^{-1} and 1447 cm^{-1} appeared as stretching peaks at 1454 cm^{-1} and 1445 cm^{-1}, respectively. In previous studies, a band around 1297 and 1303 cm^{-1} was observed for silver loaded zein materials which thought to be generated by the newly formed Ag–N coordination bonds (Dashdorj et al., 2015; Zhang, Luo, & Wang, 2010). Therefore, a band at 1308 cm^{-1} (f) and a stretching peak at 1300 cm^{-1} (g) may indicate possible Au-N interaction.

An enhancement in the signal at 1171 cm^{-1} and 1124 cm^{-1} (h) detected corresponding to carbonyl group. These changes indicated the

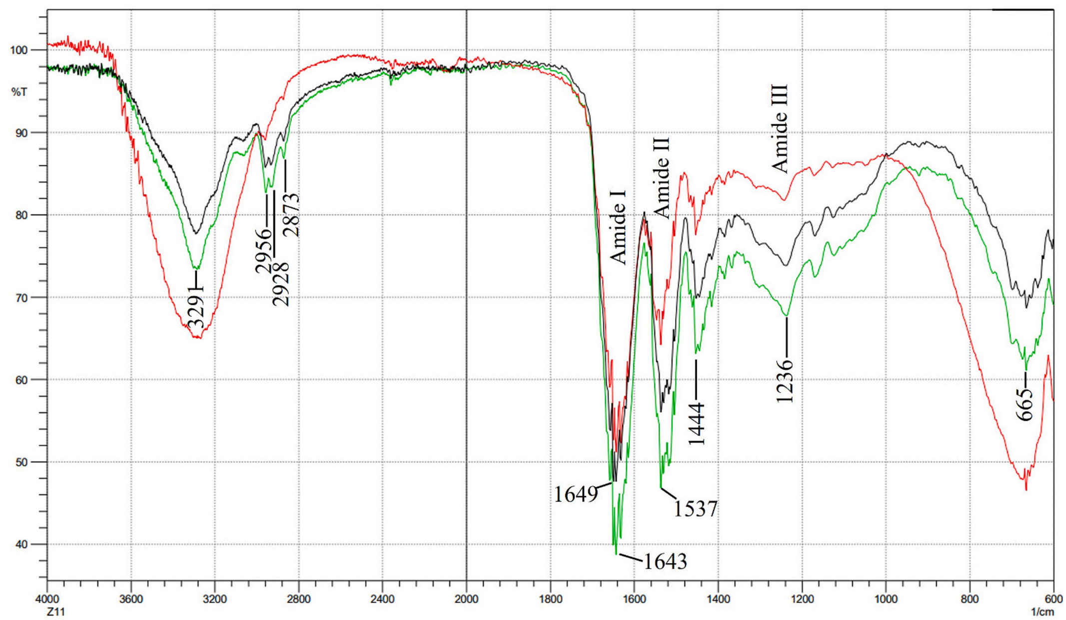

Fig. 2. FTIR spectrum of gold nanosphere (▬), pure zein nanofiber (▬), and zein nanofiber integrated with gold nanospheres (▬). (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

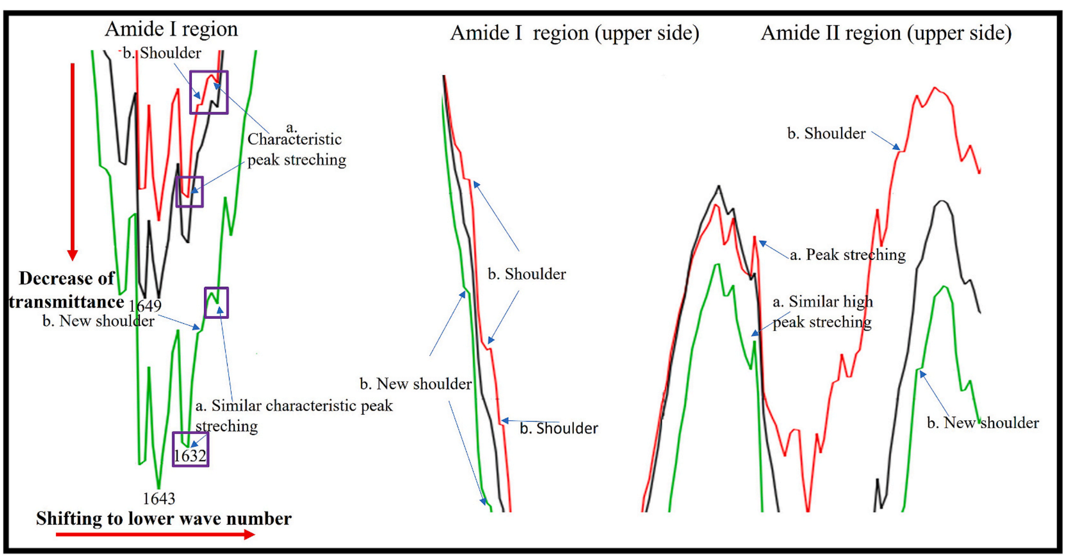

Fig. 3. Enlarged peak assignments of gold nanosphere (▬), pure zein nanofiber (▬), and zein nanofiber integrated with gold nanospheres (▬) in amide I and amide II regions. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

reduction of the gold ions which was coupled to the oxidation. Low oxidation process proved that gold nanospheres were successfully attached to the zein nanofibers. The results were in accordance with previous study (Chandran, Chaudhary, Pasricha, Ahmad, & Sastry, 2006). C-N stretching amine was observed at 1049 cm^{-1} for pure zein and shifted to 1053 cm^{-1} for zein with gold nanospheres (i). Suganya et al. (2017) produced zein-coated gold nanoparticles and have stated that 1096 cm^{-1} indicated C-N stretching of amine. For gold spectrum peaks observed at 665 cm^{-1} and 658 cm^{-1} (j) might indicate the vibration frequency of Au–O ionic bond groups. This peaks were also observed in zein nanofiber with gold spectrum. Furthermore, narrowing band was detected at 670 cm^{-1} after integration. These trends were previously observed by Gharibshahi, Saion, Gharibshahi, Shaari, and Matori (2017) who detected Ag-O ionic bond at 513 cm^{-1}.

In summary, all above mentioned findings confirmed the binding between gold ion and zein molecules, that promotes integration of the gold nanospheres to the zein nanofibers.

3.5. *Effect of nanocoating on fish fillets*

3.5.1. *Microbiological analysis of fish fillets*

The results of TMAB growth in uncoated and nanofiber coated fish fillets are shown in Table 3. The initial TMAB count of sea bream fillets was 3.80 ± 0.18 log CFU/g. Nanocoated samples showed a lower TMAB count than that of uncoated samples after the storage. Overall, the coated fish fillets showed a remarkable reduction of TMAB counts, i.e. 5.60 CFU/g, compared to that of the uncoated samples. The microbiological data were in line with recent studies, where the inhibition of rapid TMAB growth in sea bass fillets could also be obtained by coating the fish fillets with electrospun mats containing gold nanospheres

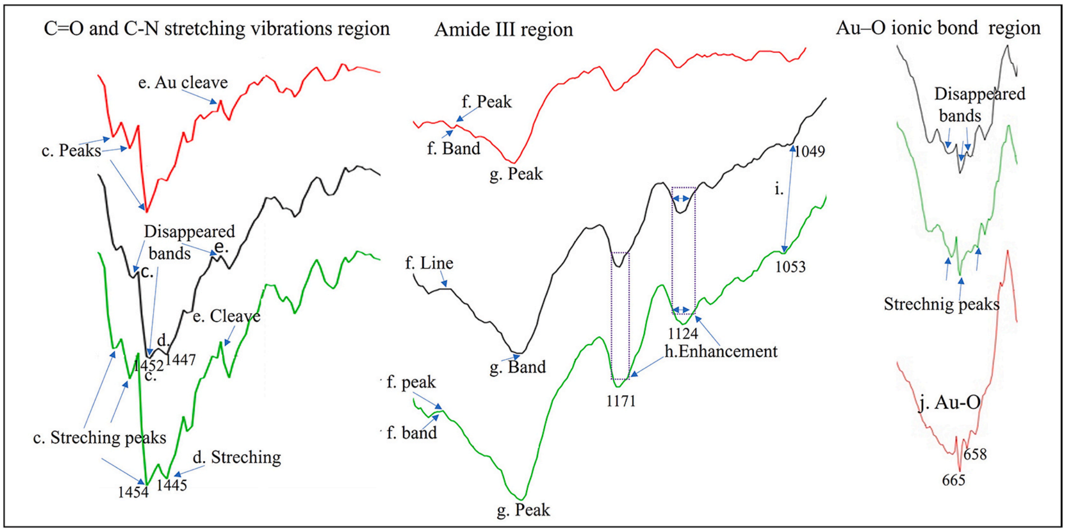

Fig. 4. Enlarged peak assignments of gold nanosphere (red), pure zein nanofiber (black), and zein nanofiber integrated with gold nanospheres (green) between 1500 cm^{-1}-600 cm^{-1}. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

Table 3
TMAB, ε′-dielectric constant, and ε″-dielectric loss factor results of nanofiber coated and uncoated fillets.

	Group	Initial	1st	2nd	3rd	4th	7th	8th
TMAB	U	3.80 ± 0.18	3.92 ± 0.09^{aA}	4.38 ± 0.23^{aAB}	4.68 ± 0.14^{aB}	4.85 ± 0.22^{aB}	5.72 ± 0.14^{aC}	6.81 ± 0.16^{aD}
	AZN		3.85 ± 0.18^{aA}	4.02 ± 0.05^{bA}	4.26 ± 0.21^{aAB}	4.43 ± 0.15^{bB}	4.67 ± 0.05^{bB}	5.60 ± 0.06^{bC}
ε′	U	75.60 ± 4.64	77.87 ± 0.72^{aA}	75.94 ± 4.63^{aA}	71.73 ± 3.70^{aA}	119.18 ± 21.98^{aB}	64.46 ± 4.53^{aA}	76.08 ± 3.92^{aA}
	AZN		78.80 ± 0.62^{aA}	77.75 ± 0.47^{aA}	77.38 ± 0.64^{aA}	75.58 ± 3.42^{aA}	64.38 ± 1.25^{aB}	74.77 ± 0.81^{aA}
ε″	U	10.55 ± 0.71	11.22 ± 0.30^{aA}	9.90 ± 1.20^{aAB}	10.46 ± 3.70^{aAB}	8.47 ± 0.52^{aB}	9.79 ± 0.40^{aAB}	10.68 ± 0.47^{aAB}
	AZN		11.62 ± 0.42^{aA}	11.14 ± 6.06^{aA}	11.20 ± 0.29^{aA}	9.89 ± 1.1^{aA}	10.24 ± 0.2^{aA}	10.41 ± 0.50^{aA}

$^{a\text{-}b}$ Within each column, different superscript lowercase letters show differences between treatment groups for same storage day ($p < 0.05$).$^{A-D}$ Within each row, different superscript uppercase letters show differences between the storage days within same analysis group ($p < 0.05$).
U: Uncoated samples; AZN: Samples treated with zein nanofibers containing gold nanospheres.

(Çetinkaya, Ceylan, Meral, Kılıçer, & Altay, 2021b). TMAB counts showed the impact of zein-gold nanofibers coating to inhibit deterioration of fish fillets. There are various mechanisms which gold nanoparticles show an antimicrobial property, i.e. interaction with the bacterial cell wall, prevention of biofilm formation, production of reactive oxygen species, and inducing leakage of intracellular components (e.g., interactions with DNA and/or proteins) (Okkeh et al., 2021). The antimicrobial mechanism depends on the shape and size of the nanomaterial. Penders et al. (2017) showed that the antibacterial action of gold-nanoflowers is associated with high-aspect ratio spikes and pillars, triggering local stress on the bacterial cell wall, resulting in membrane rupture. Nanofibers formed by other biopolymer such as chitosan interacts with the bacterial cell wall, this interaction occurs through cationic amine groups with anionic binding sites of bacterial cell walls, which damages the bacteria membrane (Arkoun, Daigle, Heuzey, & Ajji, 2017; Mohamady Hussein et al., 2021).

Moreover, a strong adhesion of gold-zein nanofibers to the membrane due to the coupling of gold/amine groups might lead to a synergistic effect in the deformation of bacteria cell. Fish fillet's surface efficiently absorbed the nanofibers due to its relatively high moisture content. The gold nanospheres slowly interacted with the outer membrane of bacteria over time, causing an increase in membrane permeability and leakage of intracellular constituents, which could damage the cells.

3.5.2. *Dielectric properties of fillets*

Table 3 shows ε′ and ε″ values of nanofiber coated, and uncoated fish fillets. On the initial day, ε′ value was 75.60, where the initial ε′ value of seabass fillet was 76.47 at 3000 MHz (Çetinkaya et al., 2021b)). On the 1st day, there was a slight increase in ε′ values, i.e. 77.87 and 78.80 for the uncoated and nanocoated groups, respectively. The nanocoated samples showed a higher ε′ on 1, 2, and 3rd day than that of the uncoated samples. Wang, Tang, Rasco, Kong, and Wang (2008) observed an increase on ε′ at 27 MHz (from 97.8 at 20 °C to 149.6 at 120 °C) and at 40 MHz but decreasing at 433 MHz (from 60.4 at 20 °C to 56.7 at 120 °C). Flocculation was observed in terms of ε′ values, since ε′ values were decreased on 2nd and 3rd day of storage for both groups. However, a significant reduction of ε′ values was observed for the uncoated group. Çetinkaya, Ceylan, et al. (2021) reported that fluctuations might occur in ε′ and ε″ during storage of fish meat. The dielectric properties of foods could be affected by the degree of water binding with constituents of the food (Samuel & Trabelsi, 2012). Li, Li, Tang, Koral, and Jiao (2019) indicated that moisture content is the most influential factor on the dielectric properties of fish meat. In this respect, the decreased ε′ was attributed to the loss of water, as it might affect the dielectric behavior. The lowest ε′ values were observed on the 7th day. Significant differences ($p < 0.05$) between storage days were observed on 4th day of storage for uncoated group (119.18) and on the 7th day for the nanocoated group (64.38). The initial ε″ value was 10.55 for the fresh fillet. The ε″ for uncoated samples and nanocoated samples ranged from 11.22 to 8.47 and 11.62 to 9.89, respectively. Results showed that fluctuations of ε′ and ε″ were higher for uncoated group, suggesting a stable shelf-life for the nanocoated group. Similarly, Çetinkaya, Ceylan, et al. (2021) proposed that the stability of dielectric properties can be provided by functional nanofibers.

4. Conclusion

The preparation, characterization, and application of zein-gold nanofibers were successfully reported. The dielectric and electrical conductivity of feeding solutions composed of gold nanospheres and zein strongly influenced the formation and properties of zein-gold electrospun nanofibers. Zeta potential and DLS measurements were provided as an useful approach to characterize nanofibers. Aggregation behavior of produced nanofibers in ethanol or water was successfully determined by DLS. FTIR data showed the interaction of amide functional groups between zein and gold ions suggesting the stabilization of the gold nanospheres by zein. The zein nanofibers integrated with gold nanospheres improved surface coating properties to reduce TMAB (17.8%) after 8 days of storage. The functionalities of zein-gold nanofibers in the reduction of TMAB were also affected by dielectric properties of fish fillet coated by nanofibers, where change in ε' and ε'', i.e. 14.84% and 10.14%, respectively, in the coated group were lower than that of uncoated group, i.e. 57.65% and 19.72%, respectively. This study may provide further development of zein-based nanofibers as edible preservatives in the food industry.

CRediT authorship contribution statement

Turgay Cetinkaya: Writing – original draft, Data curation, Formal analysis, Investigation, Validation, Visualization, Writing – review & editing. **Wahyu Wijaya:** Writing – review & editing, Conceptualization. **Filiz Altay:** Supervision, Writing – review & editing, Resources, Funding acquisition, Formal analysis, Investigation. **Zafer Ceylan:** Supervision, Writing – review & editing.

Declaration of competing interest

The authors have no conflict of interest in this research.

Acknowledgment

This work was supported by the Research Fund of the Istanbul Technical University. Project number: 42144. The study was designed as a part of Turgay Cetinkaya's Ph.D. thesis (First advisor: Prof. Dr. Filiz Altay, and co-advisor: Assoc. Prof. Dr. Zafer Ceylan).

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.lwt.2021.112976.

References

Afonso, B. S., Azevedo, A. G., Gonçalves, C., Amado, I. R., Ferreira, E. C., Pastrana, L. M., et al. (2020). Bio-based nanoparticles as a carrier of β-carotene: Production, characterisation and in vitro gastrointestinal digestion. *Molecules, 25*(19), 4497. https://doi.org/10.3390/molecules25194497

Aman Mohammadi, M., Ramezani, S., Hosseini, H., Mortazavian, A. M., Hosseini, S. M., & Ghorbani, M. (2021). Electrospun antibacterial and antioxidant zein/polylactic acid/hydroxypropyl methylcellulose nanofibers as an active food packaging system. *Food and Bioprocess Technology, 14*(8), 1529–1541. https://doi.org/10.1007/S11947-021-02654-7

Ansarifar, E., & Moradinezhad, F. (2021). Preservation of strawberry fruit quality via the use of active packaging with encapsulated thyme essential oil in zein nanofiber film. *International Journal of Food Science and Technology*. https://doi.org/10.1111/IJFS.15130

Arenas-Guerrero, P., Delgado, Á. V., Donovan, K. J., Scott, K., Bellini, T., Mantegazza, F., et al. (2018). Determination of the size distribution of non-spherical 513 nanoparticles by electric birefringence-based methods. *Scientific Reports, 8*(1), 1–10. https://doi.org/10.1038/s41598-018-27840-0, 514.

Arkoun, M., Daigle, F., Heuzey, M.-C., & Ajji, A. (2017). Antibacterial electrospun chitosan-based nanofibers: A bacterial membrane perforator. *Food Sciences and Nutrition, 5*(4), 865–874. https://doi.org/10.1002/fsn3.468

Aslaner, G., Sumnu, G., & Sahin, S. (2021). Encapsulation of grape seed extract in Rye flour and whey protein–based electrospun nanofibers. *Food and Bioprocess Technology, 14*(6), 1118–1131. https://doi.org/10.1007/S11947-021-02627-W, 2021 14:6.

Aytac, Z., Huang, R., Vaze, N., Xu, T., Eitzer, B. D., Krol, W., et al. (2020). Development of biodegradable and antimicrobial electrospun zein fibers for food packaging. *ACS Sustainable Chemistry & Engineering, 8*(40), 15354–15365. https://doi.org/10.1021/ACSSUSCHEMENG.0C05917

Bhushani, J. A., Kurrey, N. K., & Anandharamakrishnan, C. (2017). Nanoencapsulation of green tea catechins by electrospraying technique and its effect on controlled release and in-vitro permeability. *Journal of Food Engineering, 199*, 82–92. https://doi.org/10.1016/j.jfoodeng.2016.12.010

Boatemaa, M. A., Ragunathan, R., & Naskar, J. (2019). Nanogold for in vitro inhibition of Salmonella strains. *Journal of Nanomaterials*, 1–11. https://doi.org/10.1155/2019/9268128, 2019.

Borzenkov, M., Pallavicini, P., Taglietti, A., D'Alfonso, L., Collini, M., & Chirico, G. (2020). Photothermally active nanoparticles as a promising tool for eliminating bacteria and biofilms. *Beilstein Journal of Nanotechnology, 11*(1), 1134–1146. https://doi.org/10.3762/BJNANO.11.98

Çetinkaya, T., Altay, F., & Ceylan, Z. (2021a). Determination of fish meat quality changes by fast and novel methods during storage period. *Journal of the Institute of Science and Technology, 11*(3), 2030–2040. https://doi.org/10.21597/jist.795957

Çetinkaya, T., Ceylan, Z., Meral, R., Kılıçer, A., & Altay, F. (2021b). A novel strategy for Au in food science: Nanoformulation in dielectric, sensory properties, and microbiological quality of fish meat. *Food Bioscience, 41*, Article 101024. https://doi.org/10.1016/j.fbio.2021.101024

Ceylan, Z., Meral, R., Karakaş, C. Y., Dertli, E., & Yilmaz, M. T. (2018). A novel strategy for probiotic bacteria: Ensuring microbial stability of fish fillets using characterized probiotic bacteria-loaded nanofibers. *Innovative Food Science & Emerging Technologies, 48*, 212–218. https://doi.org/10.1016/j.ifset.2018.07.002

Chandran, S. P., Chaudhary, M., Pasricha, R., Ahmad, A., & Sastry, M. (2006). Synthesis of gold nanotriangles and silver nanoparticles using aloe vera plant extract. *Biotechnology Progress, 22*(2), 577–583. https://doi.org/10.1021/bp0501423

Chen, Y., Qiu, F., Tang, C., Xing, Z., & Zhao, X. (2021). Controllable self-patterning behaviours of flexible self-assembling peptide nanofibers. *Nanoscale Advances, 3*(6), 1603–1611. https://doi.org/10.1039/D0NA00892C

Cobley, C. M., Chen, J., Cho, E. C., Wang, L.v., & Xia, Y. (2011). Gold nanostructures: A class of multifunctional materials for biomedical applications. *Chemical Society Reviews, 40*(1), 44–56. https://doi.org/10.1039/B821763G

Dashdorj, U., Reyes, M. K., Unnithan, A. R., Tiwari, A. P., Tumurbaatar, B., Park, C. H., et al. (2015). Fabrication and characterization of electrospun zein/Ag nanocomposite mats for wound dressing applications. *International Journal of Biological Macromolecules, 80*, 1–7. https://doi.org/10.1016/j.ijbiomac.2015.06.026

Dede, S., & Altay, F. (2021). Investigation of electrophoretic mobility of various nanofibers in ethanol or water. *Turkish Journal of Science & Technology, 16*(2), 269–274.

Dias Antunes, M., da Silva Dannenberg, G., Fiorentini, Â. M., Pinto, V. Z., Lim, L.-T., da Rosa Zavareze, E., et al. (2017). Antimicrobial electrospun ultrafine fibers from zein containing eucalyptus essential oil/cyclodextrin inclusion complex. *International Journal of Biological Macromolecules, 104*, 874–882. https://doi.org/10.1016/j.ijbiomac.2017.06.095

EFSA ANS Panel. (2016). EFSA Panel on Food Additives and Nutrients Sources added to Food. Scientific Opinion on the Re-Evaluation of Gold (E 175) As a Food Additive. *EFSA Journal, 14*(1), Article 4362. https://doi.org/10.2903/j.efsa.2016.4362

Facchi, D. P., Souza, P. R., Almeida, V. C., Bonafé, E. G., & Martins, A. F. (2021). Optimizing the Ecovio® and Ecovio®/zein solution parameters to achieve electrospinnability and provide thin fibers. *Journal of Molecular Liquids, 321*, Article 114476. https://doi.org/10.1016/j.molliq.2020.114476

FDA. (2001). *Bacteriological Analytical Manual: Chapter 3 Aerobic plate count*. United States Food and Drug Administration.

Franceschelli, L., Berardinelli, A., Dabbou, S., Ragni, L., & Tartagni, M. (2021). Sensing technology for fish freshness and safety: A review. *Sensors, 21*(4), 1373. https://doi.org/10.3390/s21041373

García-Moreno, P. J., Mendes, A. C., Jacobsen, C., & Chronakis, I. S. (2018). Biopolymers for the nano-microencapsulation of bioactive ingredients by electrohydrodynamic processing. In T. Gutiérrez (Ed.), *Polymers for food applications* (pp. 447–479). Cham: Springer International Publishing. https://doi.org/10.1007/978-3-319-94625-2_17.

Gharibshahi, L., Saion, E., Gharibshahi, E., Shaari, A., & Matori, K. (2017). Structural and optical properties of Ag nanoparticles synthesized by thermal treatment method. *Materials, 10*(4), 402. https://doi.org/10.3390/ma10040402

Giteru, S. G., Azam Ali, M., & Oey, I. (2021). Elucidating the pH influence on pulsed electric fields-induced self-assembly of chitosan-zein-poly(vinyl alcohol)-polyethylene glycol nanostructured composites. *Journal of Colloid and Interface Science, 588*, 531–546. https://doi.org/10.1016/j.jcis.2020.12.075

Hofman, D. K., & Hall, B. (2018). Guide to electrospinning denatured whole chain collagen from hoki fish using benign solvents. *International Journal of Biological Macromolecules, 112*, 1289–1299. https://doi.org/10.1016/j.ijbiomac.2018.02.088

Icier, F., & Baysal, T. (2004). Dielectrical properties of food materials - 1: Factors affecting and industrial uses. *Critical Reviews in Food Science and Nutrition, 44*(6), 465–471. https://doi.org/10.1080/10408690490886692

Isik, B. S., Altay, F., & Capanoglu, E. (2018). The uniaxial and coaxial encapsulations of sour cherry (Prunus cerasus L.) concentrate by electrospinning and their in vitro bioaccessibility. In *Food Chemistry* (Vol. 265, pp. 260–273). Elsevier Ltd. https://doi.org/10.1016/j.foodchem.2018.05.064

Iwamoto, S., Lee, S.-H., & Endo, T. (2014). Relationship between aspect ratio and suspension viscosity of wood cellulose nanofibers. *Polymer Journal, 46*(1), 73–76. https://doi.org/10.1038/pj.2013.64

Jørgensen, L., Qvortrup, K., & Chronakis, I. S. (2015). Phospholipid electrospun nanofibers: Effect of solvents and co-axial processing on morphology and fiber diameter. *RSC Advances, 5*(66), 53644–53652. https://doi.org/10.1039/c5ra10498j
Kasaai, M. R. (2018). Zein and zein -based nano-materials for food and nutrition applications: A review. *Trends in Food Science & Technology, 79*, 184–197. https://doi.org/10.1016/j.tifs.2018.07.015
Krishnaswamy, K., & Orsat, V. (2015). Insight into the nanodielectric properties of gold nanoparticles synthesized from maple leaf and pine needle extracts. *Industrial Crops and Products, 66*, 131–136. https://doi.org/10.1016/j.indcrop.2014.12.048. le Corre-Bordes,.
Li, S., Li, F., Tang, J., Koral, T., & Jiao, Y. (2019). Influence of composition, temperature, and frequency on dielectric properties of selected saltwater and freshwater fish. *International Journal of Food Properties, 22*(1), 1920–1934. https://doi.org/10.1080/10942912.2019.1693593
Linklater, D. P., Baulin, V. A., Guével, X. le, Fleury, J.-B., Hanssen, E., Nguyen, T. H. P., et al. (2020). Antibacterial action of nanoparticles by lethal stretching of bacterial cell membranes. *Advanced Materials, 32*(52), Article 2005679. https://doi.org/10.1002/ADMA.202005679
Lin, M.-H., Sun, L., Kong, F., & Lin, M. (2021). Rapid detection of paraquat residues in green tea using surface-enhanced Raman spectroscopy (SERS) coupled with gold nanostars. *Food Control, 130*, Article 108280. https://doi.org/10.1016/j.foodcont.2021.108280
Liu, Y., Zhu, J., Weng, G., Li, J., & Zhao, J. (2020). Gold nanotubes: Synthesis, properties and biomedical applications. *Microchimica Acta, 187*(11), 612. https://doi.org/10.1007/s00604-020-04460-y
Mahendia, S., Tomar, A. K., & Kumar, S. (2010). Electrical conductivity and dielectric spectroscopic studies of PVA-Ag nanocomposite films. *Journal of Alloys and Compounds, 508*(2), 406–411. https://doi.org/10.1016/j.jallcom.2010.08.075
Mendes, A. C., & Chronakis, I. S. (2019). Functional phospholipid nano-microfibers and nano-microparticles by electrohydrodynamic processing: A review. *Journal of Self-Assembly and Molecular Electronics (SAME), 7*(1), 23–44. https://doi.org/10.13052/JSAME2245-4551.7.002
Meral, R., Alav, A., Karakas, C. Y., Dertli, E., Yilmaz, M. T., & Ceylan, Z. (2019). Effect of electrospun nisin and curcumin loaded nanomats on the microbial quality, hardness and sensory characteristics of rainbow trout fillet. *Lebensmittel-Wissenschaft & Technologie, 113*, Article 108292. https://doi.org/10.1016/j.lwt.2019.108292
Meral, R., Ceylan, Z., & Kose, S. (2019). Limitation of microbial spoilage of rainbow trout fillets using characterized thyme oil antibacterial nanoemulsions. *Journal of Food Safety, 39*(4), Article e12644. https://doi.org/10.1111/jfs.12644
Miguel, G. A., Jacobsen, C., Prieto, C., Kempen, P. J., Lagaron, J. M., Chronakis, I. S., et al. (2019). Oxidative stability and physical properties of mayonnaise fortified with zein electrosprayed capsules loaded with fish oil. *Journal of Food Engineering, 263*, 348–358. https://doi.org/10.1016/j.jfoodeng.2019.07.019
Mohamady Hussein, M. A., Guler, E., Rayaman, E., Cam, M. E., Sahin, A., Grinholc, M., et al. (2021). Dual-drug delivery of Ag-chitosan nanoparticles and phenytoin via core-shell PVA/PCL electrospun nanofibers. *Carbohydrate Polymers, 270*, 118373. https://doi.org/10.1016/j.carbpol.2021.118373
Monaghan, B. R., & White, H. L. (1936). Effect of proteins on electrophoretic mobility and sedimentation velocity of red cells. *The Journal of General Physiology, 19*(5), 715–726. https://doi.org/10.1085/jgp.19.5.715
Moradkhannejhad, L., Abdouss, M., Nikfarjam, N., Mazinani, S., & Heydari, V. (2018). Electrospinning of zein/propolis nanofibers; antimicrobial properties and morphology investigation. *Journal of Materials Science: Materials in Medicine, 29*(11), 165. https://doi.org/10.1007/s10856-018-6174-x
Nguyen, L. T., Ahmad, I., & Jayanath, N. Y. (2020). Dielectric properties of selected seafood and their products. In *Encyclopedia of marine Biotechnology* (pp. 2867–2880). Wiley. https://doi.org/10.1002/9781119143802.ch127.
Okkeh, M., Bloise, N., Restivo, E., Vita, L. de, Pallavicini, P., & Visai, L. (2021). Gold nanoparticles: Can they Be the next magic bullet for multidrug-resistant bacteria? *Nanomaterials 2021, 11*(2), 312. https://doi.org/10.3390/NANO11020312
Okutan, N., Terzi, P., & Altay, F. (2014). Affecting parameters on electrospinning process and characterization of electrospun gelatin nanofibers. *Food Hydrocolloids, 39*, 19–26. https://doi.org/10.1016/j.foodhyd.2013.12.022
Omerović, N., Djisalov, M., Živojević, K., Mladenović, M., Vunduk, J., Milenković, I., et al. (2021). Antimicrobial nanoparticles and biodegradable polymer composites for active food packaging applications. *Comprehensive Reviews in Food Science and Food Safety, 20*(3), 2428–2454. https://doi.org/10.1111/1541-4337.12727
Panyala, N. R., Peña-Méndez, E. M., & Havel, J. (2009). Gold and nano-gold in medicine: Overview, toxicology and perspectives. *Journal of Applied Biomedicine, 7*(2), 75–91.
Pecora, R. (1985). Dynamic light scattering. In R. Pecora (Ed.), *Applications of photon correlation spectroscopy*. New York: Springer Science & Business Media. https://doi.org/10.1007/978-1-4613-2389-1.
Penders, J., Stolzoff, M., Hickey, D. J., Andersson, M., & Webster, T. J. (2017). Shape-dependent antibacterial effects of non-cytotoxic gold nanoparticles. *International Journal of Nanomedicine, 12*, 2457–2468. https://doi.org/10.2147/IJN.S124442
Pérez-Guzmán, C. J., & Castro-Muñoz, R. (2020). A review of zein as a potential biopolymer for tissue engineering and nanotechnological applications. *Processes, 8*(11), 1–21. https://doi.org/10.3390/pr8111376
Perry, J. H. (1950). Chemical engineers' handbook. *Journal of Chemical Education, 27*(9), 533. https://doi.org/10.1021/ed027p533.1
Puthiyaveetil Yoosaf, M. A., Jayaprakash, A., Ghosh, S., Jaswal, V. S., Singh, K., Mandal, S., et al. (2019). Zein film functionalized with gold nanoparticles and the factors affecting its mechanical properties. *RSC Advances, 9*(43), 25184–25188. https://doi.org/10.1039/c9ra04527a
Samuel, D., & Trabelsi, S. (2012). Influence of color on dielectric properties of marinated poultry breast meat. *Poultry Science, 91*(8), 2011–2016. https://doi.org/10.3382/ps.2011-01837
Sant'Ana, L. S., Soares, S., & Vaz-Pires, P. (2011). Development of a quality index method (QIM) sensory scheme and study of shelf-life of ice-stored blackspot seabream (*Pagellus bogaraveo*). *Lebensmittel-Wissenschaft und -Technologie- Food Science and Technology, 44*(10), 2253–2259. https://doi.org/10.1016/j.lwt.2011.07.004
Saunders, Z., Noack, C. W., Dzombak, D. A., & Lowry, G.v. (2015). Characterization of engineered alumina nanofibers and their colloidal properties in water. *Journal of Nanoparticle Research, 17*, 140. https://doi.org/10.1007/s11051-015-2942-4
Seethu, B. G., Pushpadass, H. A., Emerald, F. M. E., Nath, B. S., Naik, N. L., & Subramanian, K. S. (2020). Electrohydrodynamic encapsulation of resveratrol using food-grade nanofibres: Process optimization, characterization and fortification. *Food and Bioprocess Technology, 13*(2), 341–354. https://doi.org/10.1007/s11947-019-02399-4
Silva, P. M., Torres-Giner, S., Vicente, A. A., & Cerqueira, M. A. (2021). Electrohydrodynamic processing for the production of zein-based microstructures and nanostructures. *Current Opinion in Colloid & Interface Science, 56*, Article 101504. https://doi.org/10.1016/j.cocis.2021.101504
Singh, P., König, T. A. F., & Jaiswal, A. (2018). NIR-active plasmonic gold nanocapsules synthesized using thermally induced seed twinning for surface-enhanced Raman scattering applications. *ACS Applied Materials & Interfaces, 10*(45), 39380–39390. https://doi.org/10.1021/acsami.8b14445
Sosa-Morales, M. E., Valerio-Junco, L., López-Malo, A., & García, H. S. (2010). Dielectric properties of foods: Reported data in the 21st century and their potential applications. *Lebensmittel-Wissenschaft und -Technologie- Food Science and Technology, 43*(8), 1169–1179. https://doi.org/10.1016/j.lwt.2010.03.017
Suganya, P., Vaseeharan, B., Vijayakumar, S., Balan, B., Govindarajan, M., Alharbi, N. S., et al. (2017). Biopolymer zein-coated gold nanoparticles: Synthesis, antibacterial potential, toxicity and histopathological effects against the Zika virus vector *Aedes aegypti*. *Journal of Photochemistry and Photobiology B: Biology, 173*, 404–411. https://doi.org/10.1016/j.jphotobiol.2017.06.004
Tanaka, Y., Yamamoto, T., Satomi, Y., Kubota, H., & Makita, T. (1977). Specific volume and viscosity of ethanol-water mixtures under high pressure. *Review of Physical Chemistry of Japan, 47*(1), 12–24.
Turasan, H., Cakmak, M., & Kokini, J. (2019). Fabrication of zein-based electrospun nanofiber decorated with gold nanoparticles as a SERS platform. *Journal of Materials Science, 54*(12), 8872–8891. https://doi.org/10.1007/S10853-019-03504-W
Turasan, H., & Kokini, J. L. (2017). Advances in understanding the molecular structures and functionalities of biodegradable zein-based materials using spectroscopic techniques: A review. *Biomacromolecules, 18*(2), 331–354. https://doi.org/10.1021/ACS.BIOMAC.6B01455
Uygun, E., Yildiz, E., Sumnu, G., & Sahin, S. (2020). Microwave pretreatment for the improvement of physicochemical properties of carob flour and rice starch–based electrospun nanofilms. *Food and Bioprocess Technology, 13*(5), 838–850. https://doi.org/10.1007/s11947-020-02440-x
Wang, K., Qi, Z., Pan, S., Zheng, S., Wang, H., Chang, Y. X., et al. (2020). Preparation, characterization and evaluation of a new film based on chitosan, arginine and gold nanoparticle derivatives for wound-healing efficacy. *RSC Advances, 10*(35), 20886–20899. https://doi.org/10.1039/d0ra03704d
Wang, Y., Tang, J., Rasco, B., Kong, F., & Wang, S. (2008). Dielectric properties of salmon fillets as a function of temperature and composition. *Journal of Food Engineering, 87*(2), 236–246. https://doi.org/10.1016/j.jfoodeng.2007.11.034
Wang, Y. H., Zhao, M., Barker, S. A., Belton, P. S., & Craig, D. Q. M. (2019). A spectroscopic and thermal investigation into the relationship between composition, secondary structure and physical characteristics of electrospun zein nanofibers. *Materials Science and Engineering: C, 98*, 409–418. https://doi.org/10.1016/j.msec.2018.12.134
Wei, Y., Li, C., Zhang, L., Dai, L., Yang, S., Liu, J., et al. (2020). Influence of calcium ions on the stability, microstructure and in vitro digestion fate of zein-propylene glycol alginate-tea saponin ternary complex particles for the delivery of resveratrol. *Food Hydrocolloids, 106*, Article 105886. https://doi.org/10.1016/j.foodhyd.2020.105886
Wei, Y., Sun, C., Dai, L., Zhan, X., & Gao, Y. (2018). Structure, physicochemical stability and in vitro simulated gastrointestinal digestion properties of β-carotene loaded zein-propylene glycol alginate composite nanoparticles fabricated by emulsification-evaporation method. *Food Hydrocolloids, 81*, 149–158. https://doi.org/10.1016/j.foodhyd.2018.02.042
Wei, D., Sun, W., Qian, W., Ye, Y., & Ma, X. (2009). The synthesis of chitosan-based silver nanoparticles and their antibacterial activity. *Carbohydrate Research, 344*(17), 2375–2382. https://doi.org/10.1016/j.carres.2009.09.001
Yabuki, A., Motonobu, E., & Fathona, I. W. (2017). Controlling the length of short electrospun polymer nanofibers via the addition of micro-spherical silica particles. *Journal of Materials Science, 52*(7), 4016–4024. https://doi.org/10.1007/s10853-016-0663-4
Yilmaz, A., Bozkurt, F., Cicek, P. K., Dertli, E., Durak, M. Z., & Yilmaz, M. T. (2016). A novel antifungal surface-coating application to limit postharvest decay on coated apples: Molecular, thermal and morphological properties of electrospun zein–nanofiber mats loaded with curcumin. *Innovative Food Science & Emerging Technologies, 37*, 74–83. https://doi.org/10.1016/j.ifset.2016.08.008
Zare, Y., & Shabani, I. (2016). Polymer/metal nanocomposites for biomedical applications. *Materials Science and Engineering: C, 60*, 195–203. https://doi.org/10.1016/j.msec.2015.11.023
Zhang, B., Luo, Y., & Wang, Q. (2010). Development of silver-zein composites as a promising antimicrobial agent. *Biomacromolecules, 11*(9), 2366–2375. https://doi.org/10.1021/bm100488x

Zheng, K., Setyawati, M. I., Leong, D. T., & Xie, J. (2017). Antimicrobial gold nanoclusters. *ACS Nano, 11*(7), 6904–6910. https://doi.org/10.1021/ACSNANO.7B02035

Zheng, K., Setyawati, M. I., Leong, D. T., & Xie, J. (2021). Overcoming bacterial physical defenses with molecule-like ultrasmall antimicrobial gold nanoclusters. *Bioactive Materials, 6*(4), 941–950. https://doi.org/10.1016/J.BIOACTMAT.2020.09.026

Zhong, Q., & Jin, M. (2009). Zein nanoparticles produced by liquid-liquid dispersion. *Food Hydrocolloids, 23*(8), 2380–2387. https://doi.org/10.1016/j.foodhyd.2009.06.015