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Comparative study of nanoemulsions based on commercial oils (sunflower, canola, corn, olive, soybean, and hazelnut oils): Effect on microbial, sensory, and chemical qualities of refrigerated farmed sea bass

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ABSTRACT

The effects of oil-in-water nanoemulsions using different commercial oils (sunflower, canola, corn, olive, soybean, and hazelnut oils) on the sensory, chemical (total volatile basic nitrogen (TVB-N), thiobarbituric acid reactive substances (TBARs), peroxide value (PV), free fatty acids (FFA), water holding capacity (WHC), and pH) and microbiological qualities (mesophilic aerobic bacteria, total psychrophilic bacteria, and *Enterobacteriacae* bacteria) of sea bass (*Dicentrarchus labrax*) fillets stored at 2±2°C were investigated. The quality of sea bass fillets can be improved using nanoemulsions. This is a unique preservation technique given that purified oils were used without any other chemical compounds. The sensory analyses suggested that nanoemulsion treatment extended the shelf life of the sea bass from 8 days (the control) to 10 days. However, normal acceptability limits for bacterial counts were exceeded after 6 days for the control and 8 days for treated groups. The different commercial oil nanoemulsions noticeably retain the organoleptic quality parameters.

Industrial relevance: Nanoemulsions are regarded as self-preserving antimicrobials since the water present in them is effectively bound by its structure and access to the water by microorganisms is restricted. Nanoemulsions have adverse effects on the structure and function on bacteria by destabilising the organism's lipid envelope. Nanoemulsions can be used to extend the shelf life of fresh fish.

1. Introduction

The application of nanotechnology techniques to foods may lead to modification of food texture, taste, sensory attributes, colour, ability to process, and stability during storage. Nanotechnology can also improve the water solubility, thermal stability, and oral bioavailability of functional compounds (Huang, Yu, & Ru, 2010; McClements, Decker, Park, & Weiss, 2009). The term "nanoemulsion" refers to an almost thermodynamically stable isotropically clear dispersion of two immiscible liquids, such as oil and water, stabilised by an interfacial film of surfactant molecules (Thakur, Walia, & Kumar, 2013). Nanoemulsions can act as carriers or delivery systems for lipophilic compounds, such as nutraceuticals, drugs, flavours, antioxidants, and antimicrobial agents (Sanguansri & Augustin, 2006; Weiss et al., 2008; McClements et al., 2009; Ezhilarasi, Karthik, Chhanwal, & Anandharamakrishnan, 2013). The advantages of nanoemulsions include high physical stability, high bioavailability, and low turbidity, making them attractive systems for application in the food, cosmetics, and pharmaceutical industries (Ghosh, Mukherjee, & Chandrasekaran, 2013). Oil-in-water emulsions are important vehicles for the delivery of hydrophobic bioactive compounds into a range of food products (Kentish et al., 2008). The preparation of nanoemulsions with small droplet size is of particular interest, leading to a creamier mouth feel and greater emulsion stability (McClements, 2004). In addition, the formation and stability of nanoemulsions are highly dependent on the nature of the oil phase used, for example, its polarity, water solubility, interfacial tension, and rheology. Vegetable oils are mainly composed of triglycerides (98%) and small amounts of mono- and diglycerides. The fatty acids of the different oils vary in their carbon chain length and in the number of double bonds (Barnwal & Sharma, 2005).

Nanoemulsions can be produced using a variety of methods, generally classified as high energy approaches (e.g., high-pressure homogenisers, microfluidisers, and ultrasonic devices) and low energy approaches (e.g., spontaneous emulsification (SE), phase inversion temperature (PIT), phase inversion composition (PIC), and emulsion inversion point (EIP) methods) (Anton & Vandamme, 2009; Kentish et al., 2008; Leong, Wooster, Kentish, & Ashokkumar, 2009; Silva, Cerqueira, & Vicente, 2012; Tadros, Izquierdo, Esquena, & Solans, 2004; Velikov & Pelan, 2008; Yin, Chu, Kobayashi, & Nakajima, 2009). In this study nanoemulsion

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preparation used a high energy approach with an ultrasonic homogeniser. This mechanical device can generate intense disruptive forces and is capable of producing the tiny droplets needed for nanoemulsions (McClements, 2010).

Nanoemulsions are considered as self-preserving antimicrobials due to the fact that the water present in them is effectively bound by its structure and access to the water by microorganisms is limited (Al-Adham, Khalil, Al-Hmoud, Kierans, & Collier, 2000). It has been reported that nanoemulsions have adverse effects on the structure and function on bacteria (Friberg, 1984; Jones, Song, Kaszuba, & Reboiras, 1997) by destabilising the organism's lipid envelope (Baker, Hamouda, Shih, & Andrzej, 2003). Little work has been done using nanoemulsions in seafood. Joe et al. (2012) investigated the influence of nanoemulsions on the microbiological, chemical, and sensory qualities of Indo-Pacific king mackerel (*Scomberomorus guttatus*) steaks stored at 20 °C for 72 h. Yazgan (2013) studied the effect of oil-in-water sunflower nanoemulsions on the sensory, chemical, and microbiological qualities of sea bream and sea bass fillets stored at 2 ± 2 °C.

The demand for sea bass (*Dicentrarchus labrax*) has increased over the past 15 years in many countries, especially in Europe, because of its high nutritional value, and characteristic good flavour. Therefore, sea bass producers have expanded production on the Mediterranean coast (Kyrana & Lougovois, 2002).

The present study was conducted to investigate the possible use of a number of different commercial oil-based nanoemulsions to determine which would be best for extending the shelf life of fish.

2. Materials and methods

2.1. Preparation of nanoemulsions

Nanoemulsions were prepared according to Hamouda et al. (1999). Oils used were purchased from the local market (Adana, Turkey) and were at least 5 months before their expiry date. The oil phase of the oil-in-water (O/W) nanoemulsions consists of commercial oil (14% of the total emulsion), ethanol (3%), and a surfactant (Tween 80, 3%, generally regarded as safe (GRAS)) (Sigma-Aldrich, Lyon, France) and represents 20% (*v*/*v*) of the emulsion. The components of this oil phase were mixed and kept for 1 h at 86 °C and after which the compounds were mixed with water (80%). The mixture was homogenised using an ultrasonic homogeniser (Optic Ivymen System CY-500, Barcelona, Spain) for 15 min.

2.2. Physical properties of emulsions

Viscosity and the average particle size of droplets in the emulsions were measured using a dynamic light scattering instrument (Malvern Mastersizer 2000, Malvern, UK). Thermodynamic stability was determined according to Shafiq et al. (2007). Briefly, nanoemulsions were centrifuged (Universal 32R, DJB Labcare Ltd., Buckinghamshire, England) at 13,000 × *g* for 30 min at 4 °C and maintained at 25 °C for 48 h. Six cycles of centrifugation were done, and the appearance of these formulations was studied using turbidity, phase separation, precipitation, demulsification, and creaming. Refractive index was measured using an Abbe-type refractometer (Schimidt + Haensch ATR W2, Berlin, Germany). Density was determined (kg/m^3) at 26 °C using a picnometer (ISO LAB Boro 3.3: 50 ml at 20 °C, İstanbul, Turkey). Surface tension was measured using a gonyometer (Attension Theta, Biolin Scientific, Espoo, Finland). Table 1 shows some of the properties of the oil-in-water nanoemulsions based on the commercial oils.

2.3. Preparation of sample

Sea bass (*D. labrax*) were obtained from a local fish farm in İzmir, Turkey. Fish were killed by dipping in ice-cold water (hypothermia). After death, the fish were transported to the laboratory in ice within 24 to 25 h from harvesting. The average length and weight of the sea bass were 29.7 ± 1.0 cm and 270 ± 23 g. They were immediately gutted, filleted with skin on, and divided into seven lots. One lot (126 fillets; the reference samples) was stored on plates (6 fillets per plate) wrapped with permeable stretch film (Prima Gıda Iht. Mad. Ambalaj. San, Mersin, Turkey). The other samples were treated with nanoemulsions. Fish fillets were immersed for 4 min in the nanoemulsions prepared with different oils and then stored 6 to a plate wrapped with stretch film. The average sea bass fillet weight was 55 ± 7 g. For each analysis day, 3 plates (total 18 fillets) were randomly selected for each group. Analyses were carried out seven times (d 0, 2, 4, 6, 8, 10 and 12) meaning 126 fillets for each group. Therefore, a total of 450 fish were used for this work. All samples were stored in a chill room (2 ± 2 °C). Data were obtained from fillets from three plates (triplicate) treated separately with duplicate measurements of the appropriate sample from each plate (n = 6).

Table 1
Properties of oil-in-water nanoemulsions based on commercial oils (n = 6).

Oils	Density (g/ml)	Viscosity (N s/m^2)	Thermo-dynamic stability	Mean droplet size (nm)	Surface tension (N/m)
Sunflower	0.9819 ± 0.01	1.94 ± 0.14	+++	212	33.2 ± 0.6
Hazelnut	0.9826 ± 0.02	1.99 ± 0.09	+++	185	33.4 ± 0.2
Canola	0.9846 ± 0.01	1.83 ± 0.13	+++	299	35.7 ± 1.1
Soybean	0.9837 ± 0.02	1.93 ± 0.14	+++	250	34.5 ± 0.8
Corn	0.9840 ± 0.02	1.86 ± 0.11	+++	250	33.2 ± 0.8
Olive	0.9840 ± 0.01	1.99 ± 0.11	+++	275	30.6 ± 0.7

2.4. Proximate analysis

Lipid content was measured by the method of Bligh and Dyer (1959). Ash and moisture contents were determined as described by AOAC. Official Method 920.153 (2002) and AOAC. Official Method 950.46 (2002), respectively. Protein was determined by the Kjeldahl procedure using a Buchi Digestion System, Model K-424 (BÜCHI Labortechnic, Flawil, Switzerland) and a Kjeltec Distillation Unit B-324 (BÜCHI Labortechnic). Percent protein was calculated using a Kjeldahl conversion factor of N × 6.25.

2.5. Sensory analysis

For sensory analysis, the quality index method (QIM) scheme developed by Bonilla, Sveinsdottir, and Martinsdottir (2007) was used with the raw fish with modification. The scheme consisted of 8 quality parameters (e.g., skin brightness, skin mucus, flesh texture, flesh-blood, odour, colour, brightness, and gaping). For each of these parameters, the scheme had 4 simple descriptors, scoring demerit points from 0 to a maximum of 3, where 0 represented best quality and higher scores (e.g., 3) indicated poorer quality. Twelve experienced panellists (who were familiar with sensory assessment) participated in the sensory evaluation. Training session involved the objective of the tests and discussion of the procedures. During training, fresh and very spoiled fish samples were used as references. A completely randomised sampling scheme was adopted using coded samples. Sensory assessment was done on each test day. Samples were assessed under natural day light conditions at 24 °C. At each sampling time, three fish for each group were randomly chosen to evaluate their sensorial attributes for both raw and cooked. Water has been served between tests to clean palates.

The measurement of the freshness of cooked fish (odour, taste, and texture) was assessed according to Torry Scheme (Howgate, 1982) with the modification by Alasalvar et al. (2001) for farmed sea bream. A hedonic scale from 10 to ≤3 was used to evaluate the fish. A score of 10 represents very fresh fish while ≤3 represents putrid or spoiled fish. Three random fillets from each group were cooked on top of

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