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Oil-Based Nanoparticles Containing Alternative Vegetable Oils (Grape Seed Oil and Almond Kernel Oil): Preparation and Characterization

Juliana S. ALMEIDA [1], Luciane JEZUR [2], Marcia C. FONTANA [1], Karina PAESE [3], Cristiane B. SILVA [2]; Adriana R. POHLMANN [4], Silvia S.GUTERRES [3] & Ruy C.R. BECK [1]*

[1] *Programa de Pós-Graduação em Ciências Farmacêuticas, Departamento de Farmácia Industrial, Universidade Federal de Santa Maria, Santa Maria, RS Brazil;*

[2] *Curso de Farmácia, Universidade Federal de Santa Maria, Santa Maria, RS, Brazil;*

[3] *Faculdade de Farmácia, Universidade Federal do Rio Grande do Sul (UFRGS), Av. Ipiranga, 2752, 90610-000, Porto Alegre, RS, Brazil;*

[4] *Departamento de Química Orgânica, Instituto de Química, Universidade Federal do Rio Grande do Sul (UFRGS), CP 15003, 91501-970, Porto Alegre, RS, Brazil*

SUMMARY. The use of two alternative vegetable oils (grape seed oil and almond kernel oil) to prepare nanoparticulated delivery systems (nanocapsules and nanoemulsions) for active substances was evaluated. They were prepared by interfacial deposition of preformed polymer (poly-ε-caprolactone) or spontaneous emulsification, respectively. All formulations presented nanometric size, polydispersity index below 0.30, negative zeta potential and spherical-shaped particles. Benzophenone-3, as a model substance was efficiently entrapped in these systems, independent on the type of oily phase. Its association did not alter significantly the physicochemical properties of the nanoparticle dispersions, which remained adequate until 6 months of storage. Nanocapsules and nanoemulsions prepared with both vegetable oils were suitable to delay benzophenone-3 photodegradation under UV radiation.

INTRODUCTION

Nanostructured materials such as polymeric nanoparticles, nanoemulsions, liposomes and dendrimers have been widely studied in the pharmaceutical and cosmetic field over the last years [1-4]. Active substances can have some properties improved by their incorporation in these biomedical nanomaterials, such as aqueous solubility, photostability, distribution after topical or systemic application, efficacy and bioavailability [2,5-9].

Nanocapsules are submicrometric particles (mean size below 1 µm) composed of an oily core surrounded by a thin polymeric wall [10-12]. They are formulated in presence of surfactants in order to stabilize the particles [13]. Nanoemulsions are nanometric-sized emulsions having droplets up to 500 nm, being also formulated in presence of surfactants [14]. Because of their small size, which allows them to permeate through biological barriers, these systems showed potential use following topical (ocular, dermal) or systemic administration [15,16].

The development of suitable nanocapsule suspensions or nanoemulsions for pharmaceutical or cosmetic application requires the adequate selection of their adjuvants, like polymers (for NC only), surfactants, and oils [11,17-19]. Nowadays, special attention has been given to the use of biodegradable polymers in the preparation of nanocapsules due to its degradation to non-toxic and non-reactive metabolites, which can be excreted by the organism [20]. Surfactants are necessary to obtain small and stable oil droplets. Their type and concentration can affect the physicochemical properties of the nanoparticles, such as size, polydispersity index and drug loading [11,17-19]. In addition, the type of oily phase used as the core in preparation of polymeric nanocapsules or nanoemulsions can also

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* Author to whom correspondence should be addressed. *E-mail:* ruybeck@smail.ufsm.br

have an influence mainly on the mean particle size and polydispersity index due to the difference in its viscosity, hydrophobic characteristic and interfacial tension [11,18].

Grape seed oil and almond kernel oil are vegetable oils with known antioxidant activities, based on their chemical constitution. Grape seed oil is rich in phenolic components, linoleic acid and tocopherols [21,22]. Almond kernel oil also presents high concentrations of linoleic acid [23,24]. This way, both oils could be interesting alternatives to prepare nanocapsules or nanoemulsions as delivery systems for cosmetic or pharmaceutical active substances.

To the best of our knowledge, there is no report in the literature on the use of these vegetable oils as functionally adjuvants in the preparation of nanocapsule and nanoemulsions. In this context, the aim of this work was to evaluate the feasibility of their use as alternative vegetable oils to prepare oil-based nanomaterials as delivery systems of active substances. Nanocapsules and nanoemulsions were prepared and characterized by means of mean size, polydispersity index, pH, zeta potential, and stability under storage. In order to demonstrate a practical application of these alternative oils in the development of the oil-based nanomaterials, we evaluated the incorporation of a model substance (benzophenone-3) to the systems and their properties to delay benzophenone-3 photodegradation under UV radiation.

MATERIAL AND METHODS

Materials

Grape seed oil (GSO), almond kernel oil (AKO) and benzophenone-3 (B3) were obtained from PharmaSpecial (São Paulo, Brazil). Poly-ε-caprolactone (PCL) and sorbitan monostearate (Span 60®) were acquired from Sigma (São Paulo, Brazil). Polysorbate 80 (Tween 80®) was supplied by Henrifarma (São Paulo, Brazil). All other chemicals and solvents presented pharmaceutical or HPLC grade and were used as received.

Preparation of nanocapsules and nanoemulsions

Nanocapsule suspensions (NC) were prepared (n = 3) by the interfacial deposition of preformed polymer method as described by Fessi *et al.* [25]. Briefly, an organic solution consisted of the oily phase – GSO or AKO (3.3 mL), a low HLB (hydrophilic-lipophilic balance) surfactant – Span 60® (0.776 g), the polymer (PCL) (1.0 g) and acetone (267.0 mL) was added under moderate magnetic stirring to an aqueous solution (533.0 mL) containing a high HLB surfactant - Tween 80® (0.776 g). The magnetic stirring was maintained for 10 min. Then, acetone was eliminated and the aqueous phase concentrated by evaporation under reduced pressure to a final volume of 100 mL (10 $mg.mL^{-1}$ of polymer and 3.30% v/v of oil). Nanoemulsions (NE) were prepared (n = 3) by the spontaneous emulsification method as described by Martini *et al.* [26]. To prepare the nanoemulsions, the presence of the polymer in the organic solution was omitted. All formulations were storage protected from the light.

Benzophenone-3-loaded nanoparticles were prepared at a final concentration of 1.00 $mg.ml^{-1}$, being dissolved (100 mg) in the organic solution during the nanoparticle preparation. Quali-quantitative composition of each formulation is described in Table 1.

Characterization of nanocapsules and nanoemulsions

Particle size analysis and polydispersity indices

Particle sizes and polydispersity indices (n = 3) were measured by photon correlation spectroscopy after adequate dilution of an aliquot of the suspension in purified water (Zetasizer Nanoseries, Malvern Instruments, Worcestershire, UK).

Formulation	PCL	Span 60®	GSO	AKO	Tween 80®	B3
NC-AKO	1.00 g	0.77 g	–	3.3 ml	0.77 g	–
NE-AKO	–	0.77 g	–	3.3 ml	0.77 g	–
NC-GSO	1.00 g	0.77 g	3.3 ml	–	0.77 g	–
NE-GSO	–	0.77 g	3.3 ml	–	0.77 g	–
B3-NC-AKO	1.00 g	0.77 g	–	3.3 ml	0.77 g	0.10 g
B3-NE-AKO	–	0.77 g	–	3.3 ml	0.77 g	0.10 g
B3-NC-GSO	1.00 g	0.77 g	3.3 ml	–	0.77 g	0.10 g
B3-NE-GSO	–	0.77 g	3.3 ml	–	0.77 g	0.10 g

Table 1. Quali-quantitative composition of nanoparticles – 100 ml (NC: nanocapsules; NE: nanoemulsion; B3: benzophenone-3) containing grape seed oil (GSO) or almond kernel oil (AKO) as oily phase.

Zeta potential

Zeta potentials were determined after dilution of the samples in 10 mmol L^{-1} NaCl aqueous solution using Zetasizer Nano Series (Zetasizer Nanoseries, Malvern Instruments, Worcestershire, UK).

pH

pH values of suspensions were determined by immersion of the electrode directly in the suspension using a calibrated potentiometer (MPA-210 Model, MS-Tecnopon, São Paulo, Brazil), at room temperature.

Benzophenone-3 content

Benzophenone-3 content ($mg.ml^{-1}$) was determined (n = 3) after dissolution of nanocapsules or nanoemulsions in acetonitrile (1 ml of suspension to 25 ml of acetonitrile) and assayed by high performance liquid chromatography – HPLC. The chromatographic system consisted of a Gemini RP-18 column (150 x 4.60 mm, 5 µm, Phenomenex, Torrance, USA) and a Shimadzu instrument (LC-10AVP Pump, UV-VIS SPD-10AVP Module, Class-VP Software, Shimadzu, Tokyo, Japan). The mobile phase at a flow rate of 1.0 ml.min-1 consisted of acetonitrile/water (85:15% v/v) containing 1% of acetic acid. The volume injected was 20 µL and benzophenone-3 was detected at 286 nm. Validation of the HPLC assay demonstrated that this method was linear ($y = 74529x - 1860.3$, $r^2 = 0.9999$, n = 5) in the range of 1 – 20 $\mu g.ml^{-1}$ and precise (RSD: 0.65% for repeatability and 0.68 % for intermediate precision). The specificity was tested in presence of the nanoparticle adjuvants and demonstrated that they did not alter the benzophenone-3 assay [27,28].

Encapsulation efficiency

Free benzophenone-3 was determined in the clear supernatant following separation of nanoparticles (NC and NE) from aqueous medium by a combined ultrafiltration-centrifugation technique (Ultrafree-MC® 10,000 MW, Millipore, Bedford, USA). Encapsulation efficiency (%) was calculated by the difference between the total and free benzophenone-3 concentrations determined in the nanoparticles (drug content) and in the ultrafiltrate, respectively, using the HPLC method described above.

Morphological analyses

Morphological analyses were conducted by transmission electron microscopy (TEM; Jeol, JEM 1200 Exll, *Centro de Microscopia* - UFRGS) operating at 80 kV. The diluted suspensions were deposited in Formvar-Carbon support films on specimen grid (Electron Microscopy Sciences, 400 mesh), negatively stained with uranyl acetate solution (2% m/v) and observed at different magnifications.

Stability studies

All NC and NE formulations were stored at room temperature and protected from light for 6 months. Particle size, polydispersity index, zeta potential and pH were evaluated after 6 months. Drug content and encapsulation efficiency were determined after 3 and 6 months of storage.

Photostability study

After preparation, the formulations (NC and NE) were placed in transparent quartz cells with 5 mm optical path and exposed to UVC radiation (Phillips TUV lamp – UVC long life, 30 W). The cells were irradiated in a box of mirror for 168 h (7 days). The formulations were placed at a distance of 10 cm from the fluorescent lamps. After 72 and 168 h (3 and 7 days, respectively), the total concentration of benzophenone-3 was quantified. Benzophenone-3 was assayed by HPLC after the dissolution of 200 µl of samples with acetonitrile according to the method previously described. A benzophenone-3 methanolic solution was used as a model of free benzophenone-3, prepared at a concentration of 1 $mg.ml^{-1}$.

Statistical analysis

Formulations were prepared and analyzed in triplicate. Results are expressed as mean ± SD (standard deviation). One-way analysis of variance (ANOVA) or two-way analysis of variance (ANOVA) was employed in the comparison of the experimental data. Post-hoc multiple comparisons were done by Tukey's test for significance at p–values less than 0.05. All analyses were run using the SigmaStat Statistical Program (Version 3.0, Jandel Scientific, USA).

RESULTS AND DISCUSSION

Nanoparticulated systems have been widely studied in the pharmaceutical and cosmetic area in the last years [2,4]. These drug delivery systems presented advantages compared to other systems and classical dosage forms for parenteral, oral and topical administration. Aiming to study the potential to use alternative vegetable oils to prepare nanostructured delivery systems, we

Formulation	Mean size (nm)	PDI	Zeta potential (mV)	pH
NC-AKO	243 ± 06 [a]	0.20 ± 0.00 [a]	-7.34 ± 0.67 [a]	6.97 ± 0.24 [a]
NE-AKO	275 ± 49 [a]	0.27 ± 0.01 [b]	- 6.94 ± 0.64 [a]	6.51 ± 1.19 [a]
NC-GSO	228 ± 07 [a]	0.19 ± 0.02 [a]	-8.22 ± 1.34 [a]	6.82 ± 0.02 [a]
NE-GSO	239 ± 03 [a]	0.30 ± 0.02 [b]	-8.80 ± 1.11 [a]	7.16 ± 0.08 [a]

Table 2. Physicochemical characteristics of nanoparticles (nanocapsules - NC and nanoemulsion - NE) containing grape seed oil (GSO) or almond kernel oil (AKO) as oily phase (n = 3, mean ± standard deviation). Means, in column, with the same letter are not significantly different (ANOVA, $p \leq 0.05$).

prepared nanocapsules and nanoemulsions containing grape seed oil (GSO) or almond kernel oil (AKO) as oily phase.

Preparation and characterization

Nanocapsules and nanoemulsions were prepared by interfacial polymer deposition and spontaneous emulsification, respectively. All formulations appeared macroscopically homogeneous and their aspects were similar to a milky bluish opalescent fluid (Tyndall effect), regardless of the type of oily phase (GSO or AKO) or the vesicle structure (nanocapsule or nanoemulsion). Physicochemical characteristics of the formulations are presented in Table 2. All formulations presented mean particle size in the nanometric range (220 - 280 nm), acidic pH and negative zeta potential (between -6.0 and -9.0 mV). By a one-way ANOVA analysis, these parameters were similar for all formulations ($p > 0.05$). On the other hand, NC presented a narrowed particle size distribution, as can be observed by their lower polydispersity indices (0.19 - 0.20) compared to NE (0.27 – 0.30, $p \leq 0.05$). Polydispersity indices below 0.25 indicate an adequate homogeneity of these systems [7].

In order to obtain a better evaluation of the influence of the oily phase and the vesicle structure on the physicochemical characteristics of the formulations, we also carried out a two-way ANOVA analysis. This analysis showed the influence of the oily phase on the mean particle size as well as on the zeta potential. Formulations prepared with AKO showed a higher mean size and lower zeta potential ($p \leq 0.05$). According to Schaffazick *et al.* [11], the kind of oily phase used as the core in the preparation of polymeric nanocapsules could have a great influence on the particle mean size and polydispersity index due to the difference in its viscosity, hydrophobic characteristic and interfacial tension. Polydispersity indices were influenced by the vesicle structure ($p \leq 0.05$). NC presented lower polydispersity indices than NE, regardless of the kind of oily phase, as showed previously by the one-way ANOVA analysis.

Considering the overall results just after the preparation, the formulations prepared with GSO or AKO as oily phase presented physicochemical characteristics similar to those prepared with capric/caprilic triglyceride mixture [9,19], an oily phase widely used to prepare NC and NE [1,3,29,30].

Figure 1 shows the images obtained by transmission electron microscopy, revealing that nanocapsules and droplets of the nanoemulsions were spherical in shape. The results corroborate the particle size analysis, showing that the particles are present in the nanometric range (close to 200 nm). The morphological analysis allows observing the influence of the type of oily phase on the mean particle size, as demonstrated by the two-way analysis of variance previously commented.

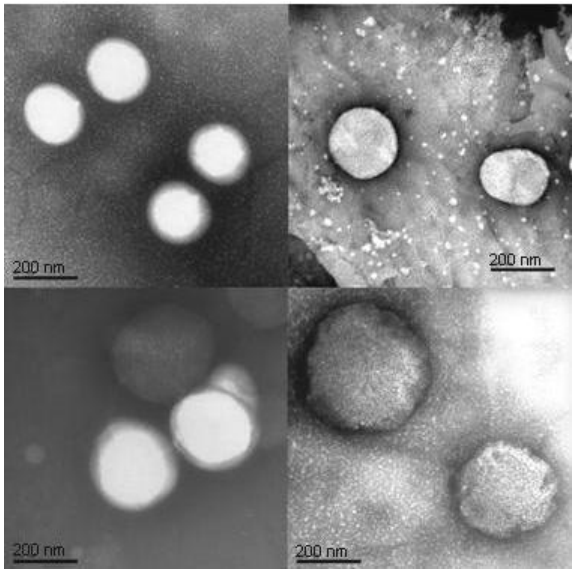

Figure 1. Transmission electron microscopy images of nanocapsules containing grape seed oil (**A**), nanocapsules containing almond kernel oil (**B**), nanoemulsion containing grape seed oil (**C**) and nanoemulsion containing almond kernel oil (**D**). Bar = 200 nm (150,000 x).

Formulation	Mean size (nm)	PDI	Zeta potential (mV)	pH
B3-NC-AKO	242 ± 11 [a]	0.21 ± 0.01 [a]	-8.76 ± 0.96 [a]	7.03 ± 0.04 [a]
B3-NE-AKO	233 ± 06 [a]	0.28 ± 0.06 [b]	-9.37 ± 1.35 [a]	7.08 ± 0.02 [a]
B3-NC-GSO	242 ± 11 [a]	0.16 ± 0.02 [a]	- 7.28 ± 1.00 [a]	6.99 ± 0.11 [a]
B3-NE-GSO	237 ± 12 [a]	0.23 ± 0.04 [a,b]	- 7.03 ± 0.34 [a]	7.20 ± 0.15 [a]

Table 3. Physicochemical characteristics of benzophenone-3-loaded nanoparticles (nanocapsules - NC and nanoemulsion - NE) containing grape seed oil (GSO) or almond kernel oil (AKO) as oily phase (n = 3, mean ± standard deviation). Means, in column, with the same letter are not significantly different (ANOVA, $p \leq 0.05$).

Formulation	Benzophenone-3 content ($mg.ml^{-1}$)	Encapsulation efficiency (%)
B3-NC-AKO	0.95 ± 0.04	99.98 ± 0.01
B3-NE-AKO	0.96 ± 0.04	99.98 ± 0.01
B3-NC-GSO	1.00 ± 0.06	99.98 ± 0.01
B3-NE-GSO	0.97 ± 0.04	99.99 ± 0.00

Table 4. Total content and encapsulation efficiency of benzophenone-3-loaded nanoparticles (nanocapsules - NC and nanoemulsion - NE) containing grape seed oil (GSO) or almond kernel oil (AKO) as oily phase (n = 3, mean ± standard deviation).

After observing the feasibility to obtain colloidal systems (NC and NE) with two alternative vegetable oils, we chose a substance model to be entrapped in these systems. Benzophenone-3 was chosen because of its sun protection properties, which could be efficiently associated to the antioxidant capacities of the oily phases. Physicochemical characteristics of the benzophenone-3-loaded systems are presented in Table 3.

All results are similar to those presented by the unloaded-systems, presenting nanometric mean particle size (230 – 252 nm), polydispersity index below 0.30, acidic pH range and negative zeta potential (-7.0 to – 9.5 mV). The presence of the sunscreen did not modify the colloidal characteristics of both systems. Regarding benzophenone-3 content and its encapsulation efficiency, all formulations showed content according to the theoretical concentration and encapsulation efficiency close to 100 % (Table 4), regardless of the oily phase or the vesicle structure. These results showed that the use of these vegetable oils could be an interesting alternative to be used in the development of dermatological and cosmetic formulations.

Stability studies

Taking into account the possibility of using these systems in the development of new products, we stored the developed formulations (containing benzophenone-3 or not) at room temperature (25 °C) and protected from light. After 6 months, the physicochemical characteristics of the systems were reanalyzed (Tables 5 and 6 for unloaded- and benzophenone-3-loaded nanoemulsions and nanocapsules, respectively).

As can be observed in Table 5, after 6 months of storage unloaded-NC and NE showed similar mean particle size and polydispersity indices compared to the initial characteristics (Table 1), confirmed by a two-way ANOVA. However, all formulations presented a significant decrease in pH values, regardless of the formulation. The decline of pH values was more pronounced for those formulations containing GSO as oily core. Regarding NC, this decrease is usually explained due to the polymer hydrolysis or the relaxation of the polymer chains [11]. However, in our study this decrease was present in formulations with or without polymer (NC and NE, respectively). Thus, the decrease in pH values observed in our study could be explained by the hydrolysis of the triglyceride chains and the respective increase of the free fatty acids

Formulation	Mean size (nm)	PDI	Zeta potential (mV)	pH
NC-AKO	222 ± 09 [a]	0.18 ± 0.00 [a,b]	-08.59 ± 0.62 [a]	4.88 ± 1.14 [a]
NE-AKO	238 ± 42 [a]	0.26 ± 0.06 [a,b]	-10.23 ± 1.31 [a]	4.78 ± 0.80 [a]
NC-GSO	220 ± 04 [a]	0.15 ± 0.05 [a]	-10.30 ± 1.41 [a,b]	3.25 ± 0.17 [a]
NE-GSO	239 ± 23 [a]	0.34 ± 0.11 [b]	-13.00 ± 0.47 [b]	3.58 ± 0.35 [a]

Table 5. Stability evaluation - Physicochemical characteristics of nanoparticles (nanocapsules - NC and nanoemulsion - NE) containing grape seed oil (GSO) or almond kernel oil (AKO) as oily phase after 6 months of storage at room temperature and protected from light (n = 3, mean ± standard deviation). Means, in column, with the same letter are not significantly different (ANOVA, $p \leq 0.05$).

Formulation	Mean size (nm)	PDI	Zeta potential (mV)	pH
B3-NC-AKO	237 ± 10 [a]	0.19 ± 0.02 [a]	-09.79 ± 0.88 [a]	4.77 ± 0.85 [b,c]
B3-NE-AKO	216 ± 16 [a]	0.29 ± 0.06 [a]	-12.19 ± 0.58 [a]	5.91 ± 1.95 [c]
B3-NC-GSO	221 ± 12 [a]	0.20 ± 0.04 [a]	-10.35 ± 0.63 [a]	3.17 ± 0.21 [b,c]
B3-NE-GSO	214 ± 10 [a]	0.25 ± 0.03 [a]	-12.82 ± 2.02 [a]	2.86 ± 0.15 [a,b]

Table 6. Stability evaluation – Physicochemical characteristics of benzophenone-3-loaded nanoparticles (nanocapsules - NC and nanoemulsion - NE) containing grape seed oil (GSO) or almond kernel oil (AKO) as oily phase after 6 months of storage at room temperature and protected from light (n = 3, mean ± standard deviation). Means, in column, with the same letter are not significantly different (ANOVA, $p \leq 0.05$).

Formulation	Benzophenone-3 content ($mg.ml^{-1}$)		Encapsulation efficiency (%)	
	3 months	6 months	3 months	6 months
B3-NC-AKO	0.99 ± 0.04	0.98 ± 0.02	99.37 ± 0.12	94.94 ± 3.56
B3-NE-AKO	1.02 ± 0.07	0.99 ± 0.10	97.89 ± 0.67	98.51 ± 0.49
B3-NC-GSO	0.99 ± 0.07	0.96 ± 0.07	99.07 ± 0.41	99.18 ± 0.18
B3-NE-GSO	1.04 ± 0.06	0.96 ± 0.04	99.39 ± 0.13	98.44 ± 0.15

Table 7. Stability evaluation - Total content and encapsulation efficiency of benzophenone-3-loaded nanoparticles (nanocapsules - NC and nanoemulsion - NE) containing grape seed oil (GSO) or almond kernel oil (AKO) as oily phase after 3 and 6 months of storage at room temperature and protected from light (n = 3, mean ± standard deviation).

content [31-34]. AKO and GSO are oils with high amounts of linoleic acid and other unsaturated fatty acids [21-23], which could undergo oxidative and hydrolytic reactions in contact with aqueous medium and temperature below 100 °C, forming free fatty acids [32,33].

The occurrence of oxidative/hydrolytic reactions during the storage time was confirmed by the development of a characteristic odor in all formulations after 6 months of storage. These results show that it is necessary to add antioxidant substances like butylated hydroxy anisole (BHA) or butylated hydroxy toluene (BHT) in future formulations in which these systems could be incorporated. In addition, as the decrease in pH values was higher for GSO formulations, these formulations presented a significant increase (in module) in their potential zeta values, explained also by the higher concentration of free fatty acids on the particle surface. Similar results were observed for benzophenone-3-loaded nanocapsules and nanoemulsions comparing the results presented in Table 3 and 5. In addition, the results showed no influence of this model substance in the stability of these systems (Table 5 and 6). Benzophenone-3 content and its encapsulation efficiency was not altered during the storage time, presenting values between 90–110 % and close to 100%, respectively, after 3 as well as 6 months of storage (Table 7).

Based on the results obtained by instrumental analysis, no differences on the physicochemical stability of the different vesicles (NC or NE) could be observed. However, macroscopic analysis allowed us to observe the occurrence of reversible aggregation (creaming) in the NE, regardless of the type of oily phase. This finding shows the importance of the polymer layer around the oil droplet to improve the physical stability of the systems. Creaming could probably not be detected by the particle size and polydispersity index measurements due to its reversible characteristic by little agitation.

Photodegradation studies

In order to evaluate the potential of these nanoparticulated systems prepared with alternative vegetable oils to prevent the photodegradation of some substances under UV radiation and to highlight their potential application in pharmaceutical and cosmetic field, we carried out a photodegradation study on benzophenone-3-loaded nanoparticles by their exposure to UVC radiation during 7 days. UVC was chosen due to its more energetic characteristic and the relative photostability of benzophenone-3 [35], allowing to reduce the experimental time. Although NE showed a lower physicochemical stability compared to NC, photodegradation studies were carried out with all formulations in order to compare the influence of the kind of oily phase

Formulation	Benzophenone-3 content (mg.ml^{-1})		
	0 h	72 h	168 h
B3-NC-AKO	0.95 ± 0.00 [a]	0.84 ± 0.04 [a,b]	0.79 ± 0.10 [b]
B3-NE-AKO	0.94 ± 0.00 [a]	0.86 ± 0.05 [b]	0.81 ± 0.10 [b]
B3-NC-GSO	0.99 ± 0.04 [a]	0.86 ± 0.03 [b]	0.78 ± 0.06 [b]
B3-NE-GSO	0.99 ± 0.02 [a]	0.85 ± 0.01 [b]	0.71 ± 0.02 [b]
MS	0.92 ± 0.03 [a]	0.74 ± 0.06 [a]	0.21 ± 0.12 [a]

Table 8. Benzophenone-3 content of free benzophenone-3 solution (methanolic solution – MS) and benzophenone-3-loaded nanocapsules or nanoemulsions after 0, 72 and 168 h of UV irradiation (n = 3, mean ± standard deviation). Means, in column, with the same letter are not significantly different (ANOVA, $p \leq 0.05$).

and the vesicle structure. A benzophenone-3 methanolic solution was used as a model of free benzophenone-3. The results of the photodegradation study are shown in Table 8. At the beginning of the experiment, all formulations as well as the methanolic solution presented similar benzophenone-3 content close to the theoretical concentration (1.00 mg.ml^{-1}). After 72 h, benzophenone-3 content in the methanolic solution decreased to 0.74 mg.ml-1, which was significantly lower ($p \leq 0.05$) than almost all nanoparticulated formulations (0.84 to 0.86 mg.ml^{-1}). However, after 168 h of UV radiation, the protective effect of the entrapment of benzophenone-3 in NC and NE prepared with GSO and AKO against photodegradation was more pronounced. Benzophenone-3 content in methanolic solution decayed from 0.92 to 0.21 mg.ml^{-1} (close to 80% of photodegradation), whereas for formulations it decayed only around 10-30% reaching mean concentrations between 0.71 and 0.81mg.ml^{-1}.

By a two-way analysis of variance, it could be observed that the kind of oily phase and the vesicle structure did not influence the protection of benzophenone-3 against the photodegradation. However, the photostability of benzophenone-3 was improved by its entrapment in the nanoparticle systems. This result suggests that benzophenone-3 is dissolved in the oily phase of the particles in a higher proportion [36] compared to its possible adsorption on the nanoparticle surfaces [11]. Solubilization of a sunscreen in the internal phase of coarse emulsions could lead to a higher photostability of formulations [37]. The results from photodegradation obtained in the present study corroborate to previous studies reported in the literature to other substances. Weiss-Angeli *et al.* [38] showed the increase in the photostability of octyl methoxycinnamate against UVA radiation after its incorporation in polymeric nanocapsules. A previous study of our research group demonstrated a lower photodegradation of tretinoin when loaded in nanocapsules or nanoemulsion compared to a free tretinoin solution [9]. Formulations developed in these two cited studies were prepared with a capric/caprylic triglyceride mixture as oily phase, pointing out the potential use of the vegetable oils suggested in our study as an alternative to compose the oily phase of such nanoparticles structures (nanocapsules and nanoemulsions).

CONCLUSIONS

This work showed for the first time the feasibility to prepare nanocapsules and nanoemulsions using grape seed oil and almond kernel oil, as alternative oily phases. Nanocapsules and nanoemulsions prepared with these oils presented nanometric size range and negative zeta potential. These parameters remained adequate after 6 months of storage at room temperature, although a decline of pH was observed for all formulations. However, nanoemulsions presented higher polydispersity indices and occurrence of creaming after 6 months of storage, regardless of the type of oily phase. The application of these alternative vegetable oils in the preparation of oil-based nanomaterials like nanocapsules and nanoemulsions with pharmaceutical or cosmetic purposes was demonstrated by an efficient entrapment of a model substance (benzophenone-3) to the nanoparticles as well as by showing their use to delay its photodegradation under UV radiation.

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