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Gelled oil particles: A new approach to encapsulate a hydrophobic metallophthalocyanine

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A B S T R A C T

Chloroaluminum phthalocyanine (ClAlPc) is a promising sensitizer molecule for photodynamic therapy, but its hydrophobicity makes it difficult to formulate. In this study, we have efficiently encapsulated ClAlPc into gelled soybean oil particles dispersed in water. 12-Hydroxystearic acid (HSA) and polyethyleneimine (PEI) were the gelling and stabilizing agents, respectively. The preparation process involved hot emulsification above the gelation temperature (T_{gel}), followed by cooling to room temperature, which gave a colloidal dispersion of gelled particles of oil in aqueous medium. The gelled particles containing ClAlPc had a medium diameter of 280 nm, homogeneous size distribution (polydispersity index ≈0.3) and large positive zeta potential (about +50 mV) and showed a spherical morphology. The gelled oil particle formulations exhibited good physical stability over a 6-month period. ClAlPc interfered with the HSA self-assembly only slightly, and decreased the gelation temperature to a small extent; however it did not affect gelation process of the oil droplets. The amounts of PEI and HSA employed during the preparation allowed us to control particle size and the dispersion stability, a phenomenon that results from complex electrostatic interactions between the positively charged PEI and the negatively charged HSA fibers present on the gelled particles surface. In summary, by using the right ClAlPc, HSA, and PEI proportions, we prepared very stable dispersions of gelled soybean oil particles with excellent ClAlPc encapsulation efficiency. The obtained colloidal formulation of gelled oil particles loaded with ClAlPc shall be very useful for photodynamic therapy protocols.

1. Introduction

Strategies from particulate colloidal dispersion have led to significant advances in drug delivery system research applied to the encapsulation of hydrophobic molecules [1,2]. Such approaches have become essential, because about 40% of the new molecules identified by pharmaceutical companies are practically insoluble in water. The lipophilic character of a molecule limits its bioavailability and absorption, thus preventing the pharmacological and pre-clinical evaluations of hydrophobic drugs [1–4]. In this context, several drug delivery systems based on lipid core have been developed to enhance the low solubility and bioavailability of lipophilic molecules [5,6].

Gelled particles have emerged as a successful system for drug delivery. These gelled particles comprise hydrogels and organogels, and they combine the unique properties of a gel with those of colloidal particles (e.g., very small size) [7–9]. Organogels (Ogg) are semisolid materials in which an organic solvent (e.g., vegetable oil) is entrapped in a three-dimensional fibrous network formed by self-aggregation of the organic gelator. Such gels are normally obtained by directly dissolving the gelator in oil at high temperature, followed by cooling of the solution to a temperature below the gelation transition temperature (T_{gel}). At T_{gel}, the oil flow is no longer observed over long periods [10,11]. Ogg are interesting drug delivery systems for different administration routes [12,13], and their potential as hydrophobic reservoirs has been demonstrated [14–17]. Recently, a new approach based on an original family of lipophilic drug carriers consisting of organogel particles has been successfully developed as a nanocarrier system applied to dye molecules [7,18]. The stabilizing and gelling agents are the main components in the process of Ogg particles production. These

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Fig. 1. Chemical structure of chloroaluminum phthalocyanine (ClAlPc).

agents directly influence the particle's physicochemical parameters, such as average diameter and zeta potential, as well as the colloidal dispersion physical stability. Both Ogg particle production and stability depend on the interfacial interactions between the particle and stabilizing agent, reinforced by a possible complexation between the organogelator and stabilizing agent. This complexation occurs mainly through electrostatic interactions with the organic gelator fibers present at the surface of the Ogg particles [7,18].

Phthalocyanines are photosensitizer drugs (PS) belonging to the *second-generation* of sensitizer molecules used in photodynamic therapy (PDT). They replace the less effective *first-generation* PS such as hematoporphyrins [6]. Metallophthalocyanines result from incorporation of diamagnetic metals like zinc, aluminum, or silicon into the phthalocyanine macrocycle. Phthalocyanines containing Al or Zn are considered to be the most photoactive compounds, because their large triplet state quantum yields and long triplet lifetimes give the highest singlet oxygen ($^1O_2$) production [6,19]. Cytotoxic species burst, such as $^1O_2$, occurs when visible light of appropriate wavelength (normally the maximum absorption wavelength of the PS) interacts with a PS in the presence of oxygen, leading to cell death and tumor destruction [20,21]. Chloroaluminum phthalocyanine (ClAlPc) (Fig. 1) is a promising PS because of its strong absorption in the visible spectral region (670–680 nm), exceptional thermal and chemical stability, and excellent photophysical and photochemical properties [19,22–24]. Despite all the potential advantages of ClAlPc for PDT, this metallophthalocyanine is practically insoluble in water, which prevents its direct application in biological fluids. In addition, hydrophobic phthalocyanines dimerize or aggregate, in aqueous medium, which inactivates the PS and diminishes the production of singlet oxygen [24,25] or other reactive oxygen species.

In this study, we prepared organogel particles containing ClAlPc (ClAlPc Ogg particles) using soybean oil and a low-molecular-weight organic gelator, such as 12-hydroxystearic acid (HSA). We obtained a colloidal suspension of ClAlPc Ogg particles by dispersing the ClAlPc Ogg in an aqueous solution containing the cationic polymer polyethyleneimine (PEI) as stabilizing agent. We characterized the resulting ClAlPc Ogg particles in terms of the average diameter, size distribution, zeta potential, encapsulation efficiency, physical stability, and morphology. In addition, we assessed the thermal behavior of Ogg and ClAlPc Ogg by differential scanning calorimetry. Therefore, the purpose of this study was to develop an innovative class of colloidal particles able to encapsulate a hydrophobic photosensitizer drug for application in PDT.

2. Materials and methods

2.1. Materials

Aluminum phthalocyanine chloride (85% pure), branched polyethyleneimine (PEI, Mn = 600 g/mol), soybean oil, and methanol were purchased from Sigma (Sigma–Aldrich Co., St. Louis, MO, USA). 12-Hydroxystearic acid (HSA, 99%) was obtained from Alfa Aesar GmbH (Karlsruhe, Germany). All the other chemicals were of analytical grade. Ultrapure water was used to prepare the colloidal dispersions.

2.2. Preparation and optimization of ClAlPc Ogg particle formulations

ClAlPc Ogg particles were prepared according to the method described by Kirilov et al. [18], with some modifications. This method involved two steps: organogel preparation and the subsequent formation of the Ogg particle dispersion.

Firstly, the ClAlPc Ogg was prepared by dissolving the gelling agent (HSA) and ClAlPc in soybean oil at 75 °C for 40 min. After complete HSA and ClAlPc dissolution in the oil, the mixture was cooled naturally to room temperature leading to a compact gel formed. Then, 10 ml of an aqueous solution containing the stabilizing agent (PEI) was added to the previously obtained gel. This mixture was heated to 75 °C (above the gel melting temperature) for 40 min, which gave in an oil layer on top of the aqueous solution. Next, the hot mixture was dispersed by ultrasound sonication (Vibra Cell, Bioblock Scientific) for 10 min using a 13-mm probe operating at a power of 400 W and frequency of 20 Hz. The resulting colloidal suspension was cooled to room temperature, to give a dispersion of ClAlPc Ogg particles. All the formulations were stored at 4 ± 2 °C for a period no longer than 6 months.

Table 1
Main components of the ClAlPc Ogg particle formulations and experimental results from physicochemical characterization.

Formulations	PEI (%)[a]	HSA (%)[b]	Mean size (nm)[c]	PdI[c]	ζ Potential (mV)[c]	ClAlPc loading (%)
F1	0.075	6	349.8 (±8.5)	0.41 (±0.04)[e]	+65.5 (±1.9)[f]	10.7 (±0.4)
F2	0.15	6	310.4 (±14.1)	0.47 (±0.01)[e]	+56.7 (±1.2)[f]	22.3 (±0.2)
F3	0.30	6	282.7 (±3.0)[d]	0.34 (±0.03)	+49.3 (±1.8)	60.3 (±1.7)[g]
F4	0.625	6	283.8 (±5.9)[d]	0.37 (±0.02)	+45.7 (±1.8)	63.1 (±2.8)[g]
F5	0.30	3	Unstable formulation (creaming and phase separation)			
F6	0.30	4	323.6 (±9.8)	0.32 (±0.04)	+52.6 (±0.8)	41.6 (±1.3)
F7	0.30	8	249.4 (±7.8)[d]	0.33 (±0.02)	+53.9 (±1.4)	38.4 (±1.7)
F8	0.30	15	Unstable formulation (compact aggregate)			

[a] % w/v.
[b] % w/w, based on oil phase weight.
[c] Mean ± SEM (n = 3).
[d] $p < 0.05$ Compared to *F1*, *F2*, and *F6* formulations.
[e,f] $p < 0.05$ Compared to *F3*, *F4*, *F6*, and *F7* formulations.
[g] $p < 0.05$ Compared to *F1*, *F2*, *F6*, and *F7* formulations.

To optimize the ClAlPc Ogg particle formulation, the concentrations of the stabilizing (PEI) and gelling (HSA) agents were varied from 0.075% to 0.625% (w/v) and 3–15% (w/w), respectively, as shown in Table 1. Soybean oil and ClAlPc concentrations were fixed at 5% (w/v) and 0.003% (w/w), respectively. For all the formulations, HSA was considered as an oil additive, so its weight percentage was based on the oily phase only. Each colloidal dispersion was prepared in triplicate.

2.3. Physicochemical characterization of ClAlPc Ogg particles

2.3.1. Particle size and polydispersity index analysis

The mean diameter and polydispersity index (PdI) of the ClAlPc Ogg particles were determined at 25 °C by photon correlation spectroscopy (PCS) at a scattering angle of 173° (Zetasizer® Nano ZS, Malvern PCS Instruments, UK). Samples were prepared by diluting 30 μl of gelled nanoparticles with 3 ml of ultrapure water. The reported values are mean ± SEM for three different batches of each colloidal dispersion.

2.3.2. Zeta potential measurement

The zeta potential (or ζ potential) of the ClAlPc Ogg particles was measured by electrophoretic mobility assayed on Zetasizer® Nano ZS (Malvern PCS Instruments, UK). The Smoluchowski model was used to estimate the ζ potential from electrophoretic mobility. The analyses were performed at 25 °C, and the samples were appropriately diluted (1/100) with ultrapurified water and the pH measured. The reported values are mean ± SEM for three different batches of each colloidal dispersion.

2.4. ClAlPc quantification in Ogg particles

2.4.1. Encapsulation efficiency (EE) and ClAlPc loading determination

The ClAlPc content in Ogg particles was determined by spectrofluorimetry (Fluorolog-3 Spectrofluorimeter, Horiba, New Jersey, USA) using a previously validated analytical method with some modifications [26]. Briefly, an aliquot of ClAlPc Ogg particles was accurately weighed (100 mg) and dissolved in 7.5 ml of methanol. The resulting mixture was sonicated for 15 min and diluted with methanol, to give a final volume of 10 ml. In addition, the free ClAlPc content in the formulations was determined by measuring the non-incorporated ClAlPc present in the clear ultrafiltrate obtained after separation of the aqueous phase by ultrafiltration/ultracentrifugation (Microcon Ultracel YM-100, Millipore, Ireland) at 12857*g* for 1 h, at 4 °C (Centrifuge 5810 R, Eppendorf, Hamburg, Germany). Both free and encapsulated ClAlPc were measured by spectrofluorimetry at fixed excitation and emission wavelengths of 615 and 674 nm, respectively. The efficiency of ClAlPc incorporation into the Ogg particles was calculated by:

$$\begin{aligned}\mathrm{EE}(\%) = &\text{total ClAlPc content}(\mu g\ g^{-1}) \\ &- \text{free ClAlPc concentration}(\mu g\ g^{-1}) \\ &/\text{total ClAlPc content}(\mu g\ g^{-1}) \times 100 \end{aligned} \quad (1)$$

$$\begin{aligned}\text{ClAlPc loading}(\%) = &\text{ photosensitizer concentration in} \\ &\text{the formulation}(\mu g\ g^{-1})/ \\ &\text{theoretical ClAlPc concentration}(\mu g\ g^{-1}) \\ &\times 100 \end{aligned} \quad (2)$$

2.5. Stability assessment

The effect of storage time on the particle size and ζ potential of the ClAlPc Ogg particles was assessed by storing colloidal dispersions in sealed glass bottles kept at 4 °C and in the dark for six months. The reported values are mean ± SEM for three different batches of each colloidal dispersion.

2.6. Morphological study

The morphology of the ClAlPc Ogg particles was evaluated by transmission electron microscopy (TEM) (JEOL JEM-1200EX, 70 kV). To this end, the samples were diluted (30 mg) in 3 ml of ultrapure water, negatively stained with 1% (w/v) sodium phosphotungstate solution, and fixed on copper grids.

2.7. Differential scanning calorimetry of organogels

Thermal properties of the organogel without ClAlPc (unloaded organogel, Ogg) and of the ClAlPc-loaded organogel (ClAlPc Ogg) were examined in a differential scanning calorimeter (Mettler Toledo, Virolay, France). Both organogels were prepared with 3%, 4%, 6%, and 8% (w/w) HSA and for ClAlPc Ogg a fixed amount of the PS (0.003%, w/v) was added. All the gel samples were melted at 75 °C, and 7–9 mg drops of the sol were placed in hermetically sealed aluminum pans. An empty aluminum pan was used as reference. DSC runs were carried out between 5 and 90 °C, and the samples were scanned at a constant rate of 2 °C min^{-1} in the heating–cooling cycle. The sol-to-gel transition temperatures (T_{gel}) were determined in triplicate for each sample.

2.8. Statistical analysis

All the experiments were performed in triplicate and data are expressed as the mean value ± SEM. Statistical analysis was performed by One-way ANOVA followed by Tukey's pairwise comparisons using the Prism 3.0® GraphPad Software. The statistical significance was set at $p < 0.05$.

3. Results

3.1. Characterization and optimization of the ClAlPc Ogg particles

3.1.1. Effects of the stabilizing and gelling agents on mean diameter and size distribution

Firstly, the influence of the stabilizing agent concentration on the average size and size distribution of the colloidal formulations was evaluated while keeping the HSA concentration at 6%. To this end, we prepared ClAlPc Ogg particles with PEI concentrations ranging from 0.075% to 0.625% (w/v). Table 1 summarizes the mean diameter and size distribution results for the ClAlPc Ogg particles prepared at four PEI concentrations, *F1*, *F2*, *F3*, and *F4*. Colloidal formulations prepared at PEI concentrations lower than 0.15% (*F1* and *F2*) had significantly different ($p < 0.05$) mean size and PdI values from those prepared at larger PEI concentrations (*F3* and *F4*). Both *F3* and *F4* presented particles with lower mean diameter and narrower size distribution compared with *F1* and *F2*; i.e., a concentration of at least 0.30% PEI furnished smaller particles and homogeneous dispersion. It can be seen from the data in Table 1 that increasing PEI percentage provided small particles and slightly reduced PdI.

Next, we varied the gelling agent (HSA) concentration from 3% to 15% (w/w in relation to the oil phase total weight) while maintaining a fixed PEI concentration of 0.30% (w/v) and an oil concentration of 5% (w/v). We obtained unstable dispersions at the lowest and highest HSA concentrations (3% and 15%, Table 1), which resulted in creaming or compact aggregation after the preparation process. As Table 1 shows, the particle size decreased for the ClAlPc Ogg particles prepared with 4%, 6%, or 8% HSA (*F6*, *F3*, and *F7*, respectively), and this was associated with increasing HSA

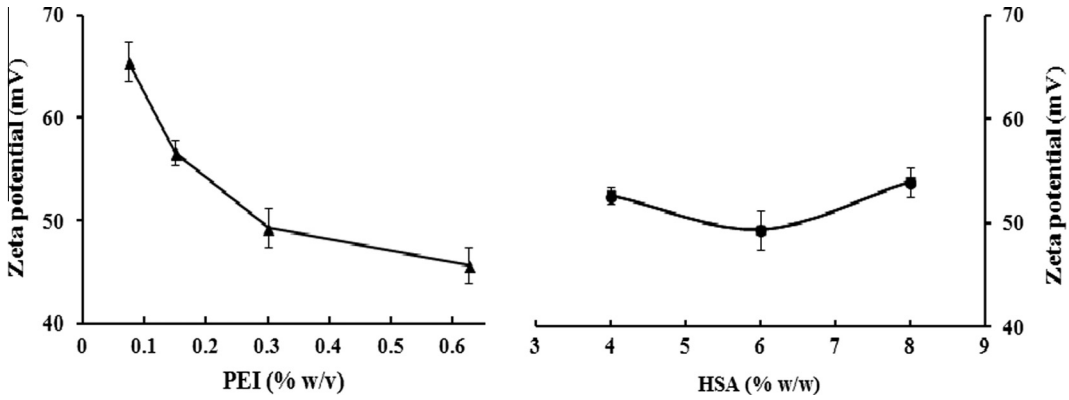

Fig. 2. Zeta potential variation of ClAlPc Ogg particle dispersions under the influence of PEI (left, fixed HSA 6%, w/w) and HSA (right, fixed PEI 0.3%, w/v) concentrations.

concentration ($p < 0.05$). HSA concentration did not affect the size distribution of the ClAlPc Ogg particle formulations, so the PdI of *F3*, *F6*, and *F7* did not differ ($p > 0.05$).

3.1.2. Effects of the stabilizing and gelling agents on zeta potential

All the formulations of ClAlPc Ogg particles had significantly positive surface charge (Table 1). The colloidal dispersions prepared with 0.075% and 0.15% PEI (*F1* and *F2*) presented higher ζ potential values than those formulations with 0.30% and 0.625% PEI (*F3* and *F4*). Increasing PEI percentage reduced the ζ potential of ClAlPc Ogg particles. On the other hand, for the formulations containing 4% and 8% HSA (*F6* and *F7*) the surface charge of gelled particles did not vary ($p > 0.05$) (Fig. 2). The pH values were 6.1, 6.0, 6.8, 6.5, 6.7, 6.8 for *F1*, *F2*, *F3*, *F4*, *F6*, and *F7* samples, respectively.

3.2. Encapsulation efficiency (EE) and ClAlPc loading determination

The EE for ClAlPc into Ogg particles was approximately 100% for all the formulations, as expected for a lipophilic photosensitizer drug. On the other hand, The ClAlPc loading varied considerably, from 10.7% (±0.4) to 63.1% (±2.8). We verified variation among formulations with fixed HSA concentration and different PEI concentrations (*F1*, *F2*, *F3*, and *F4*), and also between these formulations and those prepared with different HSA percentages and fixed PEI concentration (*F3*, *F6*, and *F7*) ($p < 0.05$) (Table 1).

F1 and *F2* formulations prepared with the lowest PEI concentrations (0.075% and 0.15%) had 6-fold lower ClAlPc loading than *F3* and *F4* formulations. The loading for ClAlPc into *F3* and *F4* preparations did not differ ($p > 0.05$), and these formulations had the highest loading rate of all the samples (Table 1). To assess the influence of the stabilizing agent (PEI) concentration on ClAlPc loading, we prepared ClAlPc Ogg particle dispersions with 0.20% and 0.25% PEI (w/v) and fixed HSA concentration (6%), which gave loading values of 32.9% and 48.4%, respectively. These findings confirmed that the PEI concentration influences ClAlPc loading into the gelled colloidal particles, and that the loading rate increased with rising PEI percentage. The highly positive correlation ($r \approx 0.99$) among PEI percentages from 0.075% to 0.30% corroborated the strong correlation between total ClAlPc content and PEI concentration (Fig. 3). For all the samples, partial loss of ClAlPc Ogg particles accounted for decreased ClAlPc loading, and a coating appeared on the glass surface of the vessel.

3.3. Assessment of the stability of ClAlPc Ogg particle dispersions

Physical stability of ClAlPc Ogg particles was assessed as a function of time over six months. To this end, we stored all the colloidal dispersions obtained from the optimization study at 4 °C, in the

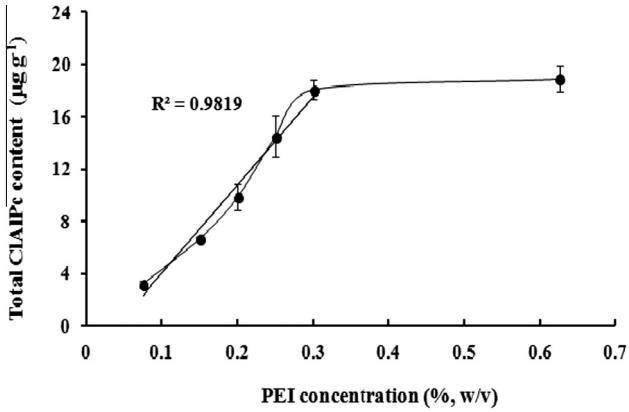

Fig. 3. Influence of PEI concentration on the total content of ClAlPc in the colloidal dispersions.

dark. The mean size and zeta potential were used as stability indices throughout this evaluation. *F1* formulation, with low amount of PEI and larger size, showed instability signs such as slightly creaming followed by separating the phases after first month (Fig. 4). The size and zeta potential of *F2* and *F6* formulations changed slightly, while *F7*, with the higher proportion of HSA increasing the fibers interconnection between particles [27], presented significantly altered size associated with creaming process after six months. *F3* and *F4* were the most physically stable formulations, since they presented minor variation on particle size and zeta potential (Fig. 4).

3.4. Morphological study

Morphology of ClAlPc Ogg particles was examined by TEM. Fig. 5 corresponds to the image of *F3* formulation. ClAlPc Ogg particles were spherical and had average diameters of less than 300 nm, corroborating the photon correlation spectroscopy analysis results. Moreover, the bright areas observed on the surface of the ClAlPc Ogg particles possibly result from zones of high HSA-fiber density. It is worth noting that the dried samples used in the TEM analyzes could be deformed due to particles and grid interactions (aggregate), and/or oil evaporation from the particle core.

3.5. Differential scanning calorimetry of Ogg and ClAlPc Ogg

The thermal properties of Ogg and ClAlPc Ogg were investigated in the temperature range of 5–90 °C by differential scanning calorimetry (DSC). Fig. 6 shows the DSC heating and cooling

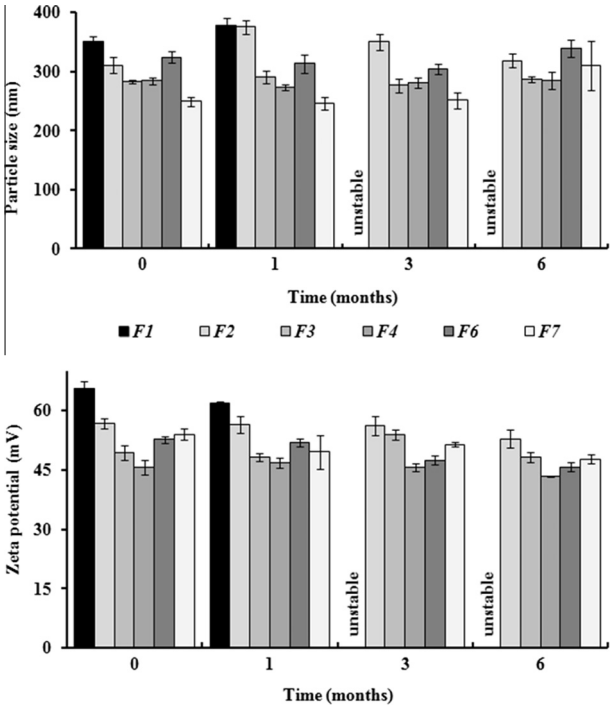

Fig. 4. Aging influence on the average diameter and zeta potential of ClAlPc Ogg particles.

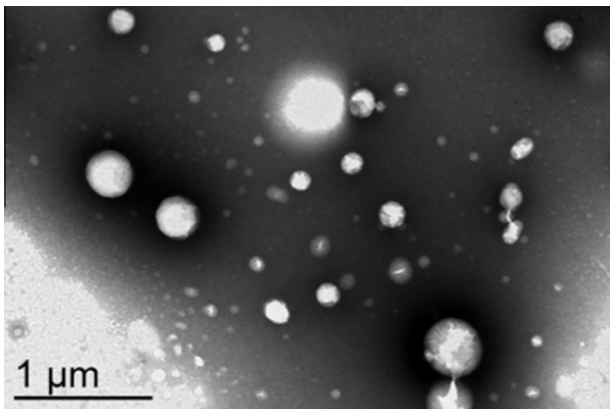

Fig. 5. TEM micrograph of ClAlPc Ogg particles (*F3* formulation).

thermographs for ClAlPc Ogg and the unloaded organogel (Ogg). During heating, endothermic peaks were observed between 56.0 and 64.1 °C for Ogg and 56.2 and 63.3 °C for ClAlPc Ogg, for different gelling agent (HSA) concentrations. These peaks correspond to first order endothermic transitions typical of the melting process. The melting characteristics of all the samples (i.e., beginning and end of melting, peak melting temperature, and enthalpy) increased with rising HSA concentration. Although the transitions occurred over a temperature range, we took the peak temperatures as the melting temperatures (T_m). The melting properties of the materials prepared with a similar HSA percentage did not differ significantly ($p > 0.05$).

On cooling, exothermic peaks were observed from 49.8 to 58.6 °C for Ogg and from 35.5 to 51.6 °C for ClAlPc Ogg prepared with different HSA concentrations. As can be seen from Fig. 6, the gelation characteristics of organogels varied according to the HSA concentration and ClAlPc addition. Thus, the gelation process of the Ogg and ClAlPc Ogg samples differed significantly ($p < 0.05$).

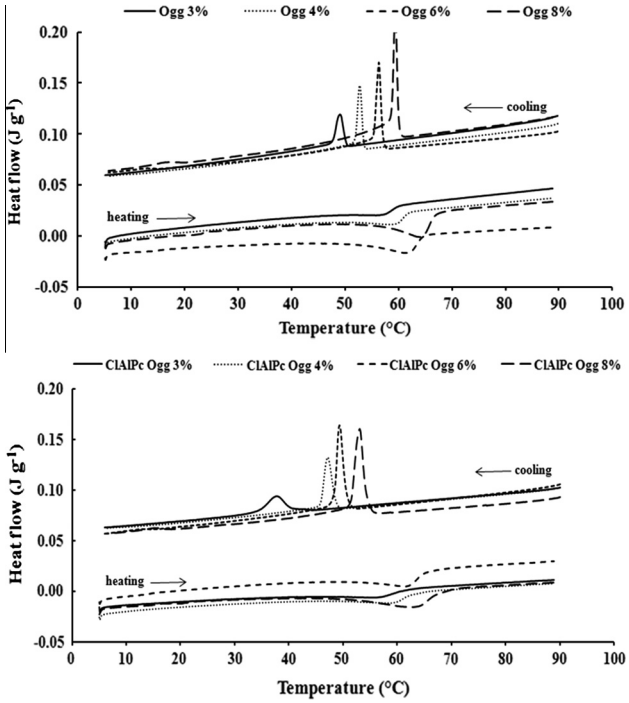

Fig. 6. DSC thermograms of unloaded organogels (Ogg) and organogels containing ClAlPc (ClAlPc Ogg) prepared with 3%, 4%, 6%, and 8% HSA (w/w).

Furthermore, ClAlPc Ogg having low HSA concentrations were more affected by ClAlPc during formation of the gelled matrix.

4. Discussion

Gelled particles have received considerable attention as drug delivery systems due to mainly their very interesting advantages such as ease of preparation and administration [8,9]. In this context, we used a new approach to encapsulate the hydrophobic photosensitizer ClAlPc; i.e. gelled oil particles.

During ClAlPc Ogg particle preparation and optimization at fixed HSA concentration (6%), increased PEI concentration (0.075–0.625%) resulted in markedly reduced mean particle size. This could be explained by increased complexation between the PEI positive charge and the negative charge of HSA fibers at the surface of the particle, leading to diminished hydrodynamic diameter of ClAlPc Ogg particles. PEI is a synthetic polymer that is very soluble in water and has high capacity for acquiring positive charge. Branched PEI contains primary, secondary, and tertiary amines, which can all be potentially protonated [28,29]. In addition, formation of this complex is influenced by the level of PEI protonation on the one hand and the flexibility of the polymer chains on the other hand [28,29]. Accordingly, the particle size analysis results could also be attributed to the low molecular weight of the branched PEI used in this work, which contains a high level of primary amines (42%) [28]. These conditions should result in maximum protonation of the amino functions with consequent high positive charge, promoting a strong electrostatic interaction between PEI and the HSA fibers.

Increased HSA concentration (4–8%) also led to in decreased ClAlPc Ogg particle diameter. However, at the lowest and highest HSA percentages, 3% and 15%, respectively, ClAlPc Ogg particles were unstable. According to Boudier et al. [7], an inadequate amount of negative charge at low HSA concentration prevents formation of a stable complex with PEI. On the other hand, at high concentration of HSA, the resulting organogel matrix is denser,

hampering close interaction between the dense HSA fibers and the PEI branched polymer. Also, increased HSA concentration in the formulations could increase the density of negative charges on the ClAlPc Ogg particle surface, resulting in better PEI adsorption. This process would decrease the polymeric corona around the ClAlPc Ogg particles, thus reducing the hydrodynamic diameter [7].

The ζ potential is an important physicochemical parameter of a colloidal dispersion. The electric charge on the surface of gelled particles influences the interaction of the dispersed particles with a biological target and physical stability of the formulations [30]. All the ClAlPc Ogg particle formulations prepared had positive ζ potential, and this parameter was mainly influenced by the PEI concentration. However, it is difficult to explain simply its variation because of the multiple interactions at the interface of the particle, especially considering the complexation between the HSA fibers and the PEI.

The encapsulation of a hydrophobic molecule in Ogg particles is related to the solubility of this molecule in the oily core. In this study, the encapsulation efficiency of ClAlPc in Ogg particles was associated with the good solubility of this photosensitizer in the soybean oil. ClAlPc loading was affected by stabilizing and gelling agent concentrations. There was a progressive increase in ClAlPc loading with increasing PEI concentration, and a maximum was reached for *F3* and *F4* formulations. On the other hand, for HSA concentration the *F3* formulation prepared with 6% (w/w) showed higher loading rate (60.3%) than *F6* and *F7* formulations. These findings could be attributed to better electrostatic interaction between PEI and HSA at concentrations 0.30% and 6%, respectively, which culminated in excellent stabilization of the ClAlPc Ogg particles and limited coating of the glass surface with these particles [7,18].

Colloidal particles dispersion stability can be controlled by the electric layer present on the dispersed phase, which generates electrostatic repulsion among the particles. *F3* and *F4* formulations showed excellent stability over six months of storage. This stability resulted from a protective corona of PEI on the particle surface leading to electrostatic and steric stabilization. On the other hand, *F1* formulation was unstable after first month. These results agree with those obtained by Kirilov et al. [18] which demonstrated that gelled particles prepared with low stabilizing agent concentration were unstable after 45 days. Therefore, the colloidal ClAlPc Ogg particle dispersion prepared with 0.30% PEI (w/v), 6% HSA (w/w), and 5% soybean oil (w/v) (*F3* formulation) showed the most appropriate characteristics for a drug delivery system: small particle size, homogeneous size distribution, and a large zeta potential.

The thermal behavior of Ogg and ClAlPc Ogg was studied by DSC. Heating prompted endothermic transitions corresponding to melting of the gel. Both the melting temperature and enthalpy increased with rising HSA concentration. The results from thermal analyses also showed that ClAlPc did not affect the melting process of Ogg or ClAlPc Ogg. However, the gelation properties changed in the case of ClAlPc Ogg. The sol–gel transition is complex and, for the low-molecular-weight gelling agent HSA, this process is based on molecular self-assembly via non-covalent interactions (hydrophobic interactions, van der Waals, and hydrogen bonding). In general, the HSA fibrous network is formed in three stages: primary nucleation of the gelator molecules, growth of the fibers, and subsequent branching formed by the three-dimensional network. HSA fiber formation starts when the solution is cooled below the gelation temperature and the solution becomes supersaturated [31,32]. Accordingly, the ClAlPc influence on the gelation process may be due to strong hydrophobic interactions between the photosensitizer and lipophilic carbon chain of HSA slightly altering the formation of the fibrous network.

5. Conclusion

We have successfully encapsulated a hydrophobic chloroaluminum phthalocyanine (ClAlPc) sensitizer into gelled particles of soybean oil. The ClAlPc Ogg particles were prepared by hot emulsification in water and cooling, and they were efficiently stabilized as a colloidal dispersion by polyethyleneimine (PEI). The amount of PEI and 12-hydroxystearic acid (HSA) used in the formulations affected particle size and colloidal dispersion stability. The phthalocyanine interacted weakly with the gelation process, leading to good encapsulation and potential use of the resulting ClAlPc Ogg particles in photodynamic therapy protocols.

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