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Research Article

Dexamethasone-Loaded Pseudo-Protein Nanoparticles for Ocular Drug Delivery: Evaluation of Drug Encapsulation Efficiency and Drug Release

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Ophthalmic drug delivery for treating various eye diseases still remains a challenge in ophthalmology. One perspective way of overcoming this problem is to use nanoscale biodegradable drug carriers that are able to safely deliver pharmaceuticals directly to the locus of disease and maintain a therapeutic concentration of drug for a long time. The goal of the present study was the preparation of drug- (dexamethasone-, DEX-) loaded pseudo-protein nanoparticles (NPs) and investigation of drug encapsulation efficiency and drug release kinetics. DEX-loaded pseudo-protein NPs (DEX-NPs) were successfully prepared by the nanoprecipitation method. DEX-NPs were characterized by size (average diameter, AD), size distribution (polydispersity index, PDI), and surface charge (zeta-potential, ZP) using the dynamic light scattering technique. DEX encapsulation characteristics were determined using the UV-spectrophotometric method, and kinetics of DEX release from DEX-NPs was studied according to the dialysis method in PBS at 37°C. The obtained results showed that size of DEX-NPs varies within 143.6–164.1 nm depending on DEX content during the preparation. DEX incorporation characteristics were determined—encapsulation efficiency (EE) and actual drug loading (DL) were high enough and reached 55.1 and 10.2%, respectively. The kinetics of DEX release from DEX-NPs showed a typical biphasic release pattern—an initial rapid (burst) release and further much more continuous slow release. Based on the obtained data, we can conclude that the elaborated DEX-NPs have potential for the application in ophthalmology as ocular drug delivery nanocarriers.

1. Introduction

Ocular drug delivery represents a central challenge in modern ophthalmology. Anatomical and physiological features of the eye make the special barriers that prevent the penetration of drugs from the surface (cornea) to the internal ocular tissues (choroid and retina). Due to the ocular barriers, it is a problem to deliver effective drug concentrations to the posterior tissues of the eye [1]. These ocular barriers include anatomical (tissue) barriers such as conjunctiva and cornea and precorneal factors like blinking, solution drainage, tear film, tear turnover, and induced lacrimation [2]. It is particularly difficult to deliver drugs to the eye *via* noninvasive techniques such as systemic or topical administration [3]. After topical instillation, very low amount of drug reaches the posterior segment of the eye (choroid and retina) due to the short time of presence of drops on the ocular surface as well as the presence of tissue barriers and the aqueous flow inside the eye [4, 5]. Systemic administration of drugs is also limited due to possible systemic adverse effects and complicated access to the target tissue, mostly because of the blood-retinal barriers [3].

Nowadays, the most effective method of drug administration to the internal tissues of the eye is intravitreal injection, which, however, represents an invasive mode of drug delivery and may be associated with a lot of side effects such as endophthalmitis, haemorrhages, ocular hypertension, damage of lens, or retinal detachment [6, 7]. Owing to the abovementioned problems, treatment of chronic eye diseases such as glaucoma, age-related macular degeneration, diabetic retinopathy, retinal degeneration, and cataracts still remains a challenge [8, 9].

To overcome the problems related to ocular drug delivery, the use of nanoscale drug carriers has been proposed. Among the variety of drug delivery nanocarriers, biodegradable (i.e., resorbable in the organism) polymeric nanoparticles (NPs) look more promising since they have the ability to be safely cleared from the body after the fulfilment of their function [10]. Furthermore, nanocarriers can provide sustained/prolonged drug release over a long period of time, as they gradually release molecules of drug during their degradation process [6]. The use of NPs, designed in due manner, can provide high corneal penetration and exclude the need for painful injections and, therefore, reduce the risk of complications [11].

The most widely used biodegradable polymers for constructing drug delivery systems are polyesters such as polylactic acid (PLA), polylactic-co-glycolic acid (PLGA), and poly-ε-caprolactone (PCL). However, degradation products of these synthetic polymers are considered to be relatively toxic and induce undesired changes in cells [12]. In addition, these biomaterials revealed lower affinity to living tissues (mainly due to the absence of hydrophilic CO-NH bonds) that decreases the bioavailability of NPs prepared from these materials [13]. Furthermore, the acidic products that are released during the degradation of polyesters transiently decrease pH of vitreous humor (up to pH = 7) and, therefore, increase the risk of inflammation. For this reason, another class of biodegradable synthetic polymers—poly(ester amide)s (PEAs) combining useful properties of two important classes of polymers, polyesters and polyamides—looks more promising for biomedical applications. The naturally occurring α-amino acid-based PEAs are also mentioned as pseudo-proteins due to the composition similar to proteins. These pseudo-proteins contain both the hydrophilic CO-NH links (which increase polymer-tissue affinity) and easily hydrolysable ester bonds (which improve biodegradability of polymers) in the backbone [14–19]. During their degradation, pseudo-proteins release weakly acidic nontoxic products, thereby preventing significant changes in pH of vitreous body and reducing risk of inflammation. Moreover, it was shown that pseudo-proteins have excellent biocompatibility with ocular tissues [14, 15]. Due to the abovementioned advantages, for constructing biodegradable nanoscale drug delivery containers, we have chosen the naturally occurring α-amino acid-based PEAs—pseudo-proteins.

In the reported research [20], we systematically studied the capability of several pseudo-proteins to form stable NPs using the nanoprecipitation (polymer deposition) method. As a result, it was established that the pseudo-protein based on sebacic acid (8), L-leucine (L), and 1,6-hexanediol (6) labelled as 8L6 (Figure 1) forms stable NPs having the particle size (around 100–200 nm) that is suitable for the intraocular drug transportation. Furthermore, in this work, we showed that the pseudo-protein 8L6 was the best in terms of particle size, stability, and cell compatibility. In another work [21], we prepared fluorescently tagged 8L6 pseudo-protein NPs and performed *in vivo* study of their penetration through ocular barriers using the experimental animals (wild-type C57BL/6 mice). The *in vivo* study showed that the NPs are able to penetrate cornea and sclera and get even into the retina. These findings may open the path to clinical application of the new type of NPs by loading them with ocular drugs.

For the treatment of various neovascular diseases on the back of the eye, steroids have been used in the recent years, such as dexamethasone (DEX, Figure 1). Intravitreally injected DEX has been used to treat not only macular degeneration but also various other eye diseases [22]. Furthermore, DEX is a hydrophobic drug, and it can be easily incorporated into the polymeric NPs by the nanoprecipitation method since molecules of DEX have affinity to hydrophobic matrix of 8L6 NPs and will not be easily released from the NPs due to strong hydrophobic interactions. The goals of the present work are the preparation of drug-(DEX-) loaded pseudo-protein NPs and investigation of drug encapsulation efficiency and drug release kinetics. Furthermore, finding optimal (the highest) polymer concentration in the organic phase during the fabrication process of NPs (nanoprecipitation) for maximum drug entrapment (encapsulation) was included in the present work.

2. Materials and Methods

2.1. Materials. The drug dexamethasone (DEX) (powder, 98% pure), the surfactant Tween 20 (sorbitan monolaurate, MW 1228), phosphate-buffered saline (PBS, pH = 7.4), and dimethyl sulfoxide (99.9% pure) were purchased from Sigma-Aldrich (Germany). All the chemicals were used as received. The dialysis tubes (MWCO 25 kDa) were purchased from Sigma-Aldrich (Germany) as well.

2.2. Preparation of Empty NPs at Different Polymer Concentrations. Pseudo-protein 8L6, selected for constructing NPs, was synthesized *via* interfacial polycondensation as reported previously [17, 19]. Empty NPs based on the pseudo-protein 8L6 were prepared according to the nanoprecipitation method under the optimal conditions that were previously established for pseudo-proteins [20]. In brief, a predetermined amount of 8L6 (10, 15, 20, 25, 30, and 35 mg) was dissolved in 1.0 mL of dimethyl sulfoxide (DMSO), and the obtained solution (organic phase) was added dropwise to 10 mL of water (water phase) containing 50 mg of the surfactant Tween 20 at an average stirring rate (700 rpm). Suspensions of the obtained blank NPs were stirred additionally for 15 min and then were washed twice

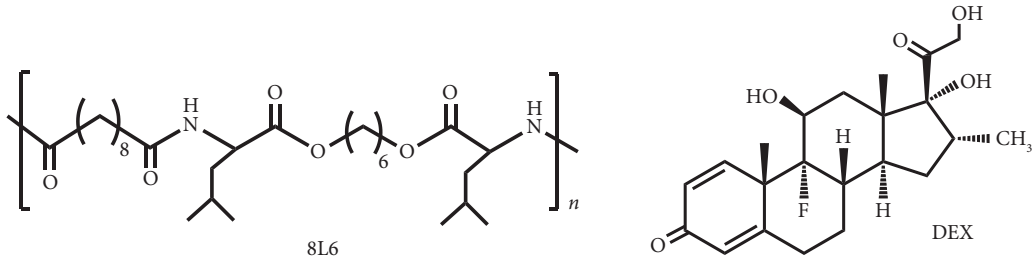

Figure 1: Chemical structures of pseudo-protein 8L6 and the drug DEX.

with distilled water by centrifugation at 30,000 rpm for 40 min using the Optima™ MAX-XP Ultracentrifuge (Beckman Coulter, Inc., USA). The obtained NPs were kept at low temperature (4°C) and used for stability study.

2.3. Preparation of DEX-Loaded NPs at the Optimal Polymer Concentration. DEX-loaded NPs based on the pseudo-protein 8L6 (labelled as DEX-NPs) were prepared according to the nanoprecipitation method (see Figure 2). In brief, the optimal amount of the pseudo-protein 8L6 (30 mg) was dissolved in 1.0 mL of dimethyl sulfoxide (DMSO) along with a predetermined amount of DEX (10, 20, 30, 40, and 50 wt% from the polymer mass), and the obtained solution (organic phase) was added dropwise to 10 mL of water (water phase) containing 50 mg of Tween 20 at an average stirring rate (700 rpm). Suspensions of DEX-NPs were stirred additionally for 15 min and then were washed twice with distilled water by centrifugation at 30,000 rpm for 40 min using the Optima™ MAX-XP Ultracentrifuge (Beckman Coulter, Inc., USA). The obtained purified DEX-NPs were immediately used for drug incorporation studies. Part of the purified DEX-NPs was also kept at low temperature (4°C) for stability study. The aggregation of DEX-NPs (if any) was evaluated visually by the appearance of small polymeric aggregates in the suspension.

2.4. Determination of NPs' Characteristics and Evaluation of Their Stability. The obtained NPs were characterized by size (average diameter (AD)), particle size distribution (polydispersity index (PDI)), and zeta-potential (ZP), which were determined by the dynamic light scattering (DLS) method using the analyser machine Zetasizer Nano ZS (Malvern Instruments, Malvern, UK). PDI values smaller than 0.05 are mainly seen with highly monodisperse standards, PDI ≤0.5 shows a narrow particle distribution (NPs are considered to be monodisperse), and PDI >0.5 displays a wide particle distribution [23–25]. AD, PDI, and ZP are presented as averages of three independent parallel experiments ± -standard deviation (SD). Stability of the prepared NPs versus time (upon storage at low temperature) was studied by measuring AD and PDI at predetermined time points using the DLS method.

The morphological examination of DEX-NPs was performed on a transmission electron microscope (Philips Tecnai 10, Philips, Eindhoven, the Netherlands) using negative staining of the samples with uranyl acetate solution (1%). In brief, a drop of concentrated suspension of NPs (30 mg/mL) was placed on a carbon grid surface with a filter paper. A drop of uranyl acetate solution was added to the surface of the carbon-coated grid. After 1 min incubation, excess fluid was removed and the grid surface was air-dried for 24 h at room temperature before being loaded into the microscope.

2.5. Determination of Drug Incorporation. The effectiveness of DEX incorporation into the pseudo-protein NPs was studied using a spectrophotometric method as reported elsewhere [26, 27]. Two main characteristics of the drug incorporation process—encapsulation efficiency in percentage (EE%) and actual drug loading in percentage (DL %)—were determined using the following procedure: freshly prepared and purified (by centrifugation) suspensions of DEX-NPs were lyophilized, the freeze-dried DEX-NPs were dissolved in DMSO, and DEX concentrations were determined by measuring absorbance of the solution at $\lambda = 254$ nm and comparing it with a calibration curve. The exact concentrations of DEX were calculated using equations acquired from the calibration curve. EE% and DL% were calculated using the following equations:

$$\text{EE\%} = \frac{\text{weight of DEX encapsulated into the NPs}}{\text{weight of DEX initially added in solvent}} \times 100\%,$$
$$\text{DL\%} = \frac{\text{weight of DEX encapsulated into the NPs}}{\text{weight of NPs containing DEX}} \times 100\%. \quad (1)$$

For finding optimal polymer/drug ratio in order to reach maximum DEX loading, different concentrations (10, 20, 30, 40, and 50 wt% from the polymer mass) of DEX were tested.

2.6. In Vitro Drug Release Study. The *in vitro* release behaviour of DEX from DEX-NPs was analysed under sink conditions using the dialysis method as described by Krishnan et al. [27]. In brief, 6 mL of freshly prepared suspension of DEX-NPs (containing 2.45 mg DEX) was loaded into dialysis tube with MWCO 25 kDa. Afterwards, the tube was immersed in 100 mL of phosphate-buffered saline (PBS, pH = 7.4, release media) under moderate stirring (300 rpm) at 37°C. At predefined time points, 10 mL of the release media was taken, and the same volume of fresh PBS was added to the media in order to maintain a constant volume. PBS containing the released DEX collected from each time point was freeze-dried, and then, the resulting

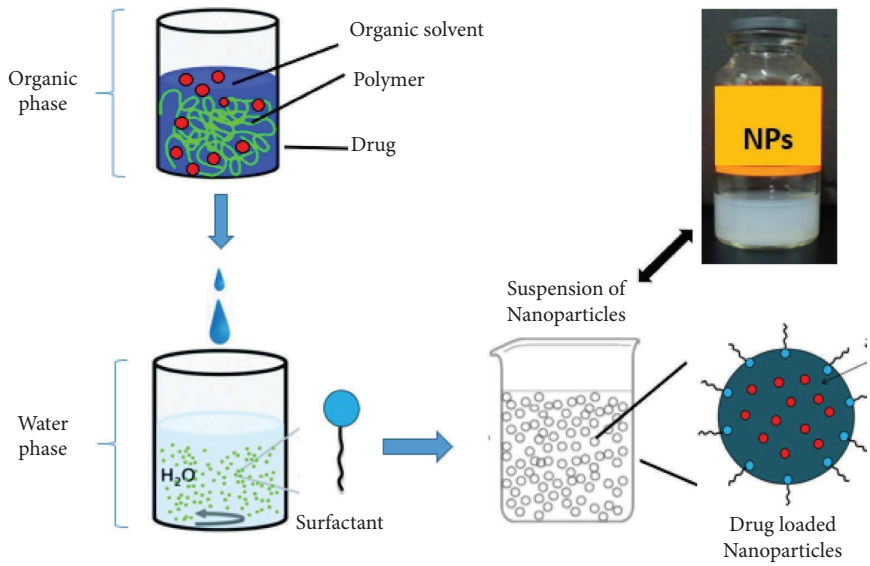

Figure 2: Scheme of the nanoprecipitation method.

solid was dissolved in DMSO again. Afterwards, DEX concentration was determined by measuring the absorbance of the solution at $\lambda = 254$ nm using a UV-spectrophotometer. The cumulative release profile was computed by dividing the amount of drug released in one specific time point by the total amount initially loaded (i.e., 2.45 mg). For comparison, 2.45 mg of DEX powder was examined under the same experimental conditions. Three parallel experiments were conducted, and results were presented as averages.

3. Results and Discussion

The empty and DEX-loaded pseudo-protein NPs were successfully prepared using the nanoprecipitation (polymer deposition) method. Nanoprecipitation is a simple and cost-effective technique of preparing polymeric NPs. This method is based on the precipitation mechanism. Precipitation of polymer macromolecules occurs after the addition of an organic phase (polymer solution) to a water phase (solution of a surfactant in water) in a four-step mechanism: (1) supersaturation, (2) nucleation, (3) growth by condensation, and (4) growth by coagulation [28]. According to this technique, drug loading is achieved by dissolving drugs in an organic phase along with a polymer (i.e., matrix of NPs). Encapsulation of drugs to NPs occurs during their formation process.

3.1. Preparation and Characterization of Empty NPs. In order to increase the degree of drug encapsulation to the NPs, we determined the maximal polymer concentration in the organic phase during the nanoprecipitation process, since it is obvious that the higher the polymer (i.e., matrix of NPs) content during the nanoprecipitation, the higher the degree of drug encapsulation. For this study, we prepared empty (unloaded) pseudo-protein NPs at different polymer concentrations in the organic phase (see Section 2.2 in "Materials and Methods") and studied the influence of polymer concentration on the formation of NPs. Results of the study are given in Table 1.

The obtained results show that the higher the polymer concentration in the organic phase, the bigger the average diameter (AD) of the formed NPs. Thus, increasing the polymer concentration from 10 to 35 mg/mL increases AD from 91.7 to 158.3 nm. At the tested highest concentration of the polymer (35 mg/mL), partial aggregation of NPs was observed. For this reason, the polymer concentration of 30 mg/mL was selected as optimal and used for further experiments to prepare drug-loaded NPs. As to PDI, the obtained empty NPs demonstrated a narrow particle distribution (PDI <0.5).

3.2. Preparation and Characterization of DEX-NPs. DEX-NPs were prepared by the nanoprecipitation method at the selected optimal concentration of polymer in the organic phase (30 mg/mL). Particle size of DEX-NPs with different contents of DEX measured by DLS revealed that AD of DEX-NPs is slightly increasing depending on the rise of DEX concentration in organic phase (Table 2). Thus, AD of DEX-NPs with 10 wt% of DEX was 143.6 nm, whereas AD of DEX-NPs with 50 wt% of drug was increased up to 164.1 nm. It should be noted that in the case of the highest concentration of DEX (50 wt%), partial aggregation of the formed NPs occurred during the preparation process.

As regards polydispersity, obtained DEX-NPs are characterized by a narrow particle size distribution. As shown in Table 3, PDI of DEX-NPs ≤0.1 that shows practically monodispersity of the obtained systems. It should be underlined that PDI values of 0.2 and below are desired and acceptable for polymer-based NPs [24]. Hence, in respect of a size distribution, the elaborated DEX-NPs are suitable for practical application. With regard to the surface charge, the generated DEX-NPs had quite high negative charge (zeta-potential, ZP) that reached up to −31.5 mV in the case of 20 wt% of DEX. Again, in the case of the partially

Table 1: Influence of the polymer concentration in the organic phase on the formation of NPs.

Concentration of polymer in the organic phase (mg/mL)	AD (nm) ± SD	PDI ± SD
10	91.7 ± 0.9	0.086 ± 0.013
15	102.1 ± 0.7	0.095 ± 0.011
20	113.4 ± 1.2	0.101 ± 0.016
25	123.5 ± 0.6	0.097 ± 0.013
30	138.4 ± 0.7	0.093 ± 0.012
35	158.3 ± 1.5[§]	0.269 ± 0.026[§]

AD, average diameter; PDI, polydispersity index; SD, standard deviation. [§]Partially aggregated sample. Data presented are averages of three independent experiments.

Table 2: Main characteristics of the freshly prepared DEX-NPs with different concentrations of DEX.

DEX content (wt% of the polymer)	AD (nm) ± SD	PDI ± SD	ZP (mV) ± SD
10	143.6 ± 0.8	0.069 ± 0.010	−26.6 ± 1.4
20	151.8 ± 0.3	0.098 ± 0.025	−31.5 ± 3.2
30	155.2 ± 4.6	0.065 ± 0.034	−26.3 ± 1.7
40	156.9 ± 2.0	0.102 ± 0.017	−28.4 ± 2.7
50	164.1 ± 9.5[§]	0.272 ± 0.074[§]	−16.3 ± 0.8[§]

AD, average diameter; PDI, polydispersity index; ZP, zeta-potential; SD, standard deviation. [§]Partially aggregated sample. Data presented are averages of three independent experiments.

aggregated sample with 50 wt% of DEX, ZP was low: −16.3 mV. We assume that negative ZP of DEX-NPs is caused by a partial hydrolysis of ester links of the polymer 8L6 that generates free carboxyl groups—carboxylate anions.

The morphological examination of DEX-NPs was performed by transmission electron microscopy (TEM). For this study, DEX-NPs prepared at optimal drug concentration—20 wt% of DEX (see Section 3.3)—were selected. TEM images of the DEX-NPs are given in Figure 3. According to the DLS analysis, the DEX-NPs prepared at 20 wt% of DEX had the average diameter of 151.8 nm. TEM analysis of these NPs also confirmed DLS measurement results—the size of DEX-NPs was in good accordance with the DLS data and no aggregates were observed. As shown in Figure 3, DEX-NPs had spherical shape.

3.3. Drug Incorporation and Stability Studies. In order to reach maximum drug loading, various concentrations of DEX were used during the preparation of NPs. We tested EE % and DL% values at five different concentrations of DEX: 10, 20, 30, 40, and 50 wt% from the polymer mass. The obtained results are shown in Table 3. The calibration curve for DEX in DMSO is depicted in Figure 4. As we can see from the results, the lowest values of EE% and DL% (25.9% and 2.5%, respectively) were obtained at the lowest concentration of DEX (10 wt%). The maximum value of EE% up to 62.8% was reached at 30 wt% of DEX, whereas maximum DL% (19.0%) was revealed for DEX-NPs with 40 wt% of DEX content. Nonetheless, concentrations of DEX at 30 and 40 wt% could not be selected as optimal due to poor stability of DEX-NPs at these concentrations of drug. At maximum value of DEX content (50 wt%), partial aggregation of forming NPs occurred leading to significant decrease of EE% and DL% values to 18.0% and 8.2%, respectively.

As shown in Table 2, increasing the content of DEX in NPs led to negligible changes in main characteristics of NPs. Consequently, poor stability of the NPs with high DEX content (30–50 wt%) might not be stipulated by slight shifts of size, PDI, and zeta-potential of the NPs. The loss of stability could be presumably attributed to the high content of hydrophobic drug in polymeric matrix: excessive number of DEX molecules incorporated into the matrix of NPs may weaken chain-chain interactions among polymeric macromolecules and loosen the solid matrix of the NPs, thereby decreasing their stability.

As shown in Table 3, stable DEX-NPs were obtained only in the case of 10 and 20 wt% of DEX content. Based on the obtained results, the concentration of DEX at 20 wt% was selected as optimal owing to the relatively high values of EE % and DL%—55.1% and 10.2%, respectively. Relatively high encapsulation efficiency of DEX could be explained by its high hydrophobicity which consequently reduces the possibility of drug loss by diffusion during the formation of NPs. Comparable values of drug incorporation were obtained by other studies using amphiphilic polymers as carriers for the drug DEX [27, 29]. It was shown that EE% of DEX to poly-ε-caprolactone NPs reached 52.6% [27], whereas for DEX-loaded PLA NPs, EE% was only 48.5% [29].

DEX-NPs with the selected optimal DEX content (i.e., 20 wt%) were studied for stability versus time at 4°C (Table 4). AD and PDI of DEX-NPs were measured right after their fabrication, and then suspensions of the NPs were stored at low temperature. After predetermined time points, suspensions of DEX-NPs were thoroughly shaken and analysed for AD and PDI. The stability study of the obtained drug-loaded NPs upon storage is essentially important since for the practical application of drug nanocarriers in medicine, they should be highly stable and their parameters should remain unchanged over time. The obtained results showed that the generated NPs were highly stable—no substantial change of AD and PDI or aggregation was observed after 6 months of storage at low temperature (4°C).

3.4. In Vitro Drug Release Study. In vitro release of DEX from DEX-NPs and DEX powder (control group) was evaluated by incubating the samples in PBS (pH = 7.4) under sink conditions at 37°C for 30 days. Results of drug release study are shown in Figure 5. The *in vitro* release profiles for DEX from DEX-NPs revealed a biphasic release pattern, namely, an initial burst release and further much more continuous slow release. The initial burst release showed that 31.65% of initially accumulated DEX was released from DEX-NPs at the first 24 hours. From day 2 to day 30, DEX release profile had sustained and slow character. Thus, during these 29 days (i.e., from day 2 to day 30) only 30.53% of accumulated DEX was released with an average rate around 1.28% per day. During the whole experimental period (30 days), only

Table 3: Drug incorporation characteristics of the DEX-NPs.

DEX content (wt% of the polymer)	EE (%) ± SD	DL (%) ± SD	Stability of DEX-NPs upon storage at 4°C
10	25.9 ± 0.4	2.5 ± 0.3	Stable
20	55.1 ± 1.1	10.2 ± 0.3	Stable
30	62.8 ± 5.0	15.8 ± 1.1	Partial aggregation within a week
40	58.9 ± 2.8	19.0 ± 0.7	Partial aggregation within a week
50	18.0 ± 1.2	8.2 ± 0.4	Partial aggregation during the preparation

EE, encapsulation efficiency; DL, drug loading; SD, standard deviation. Data presented are averages of three independent experiments.

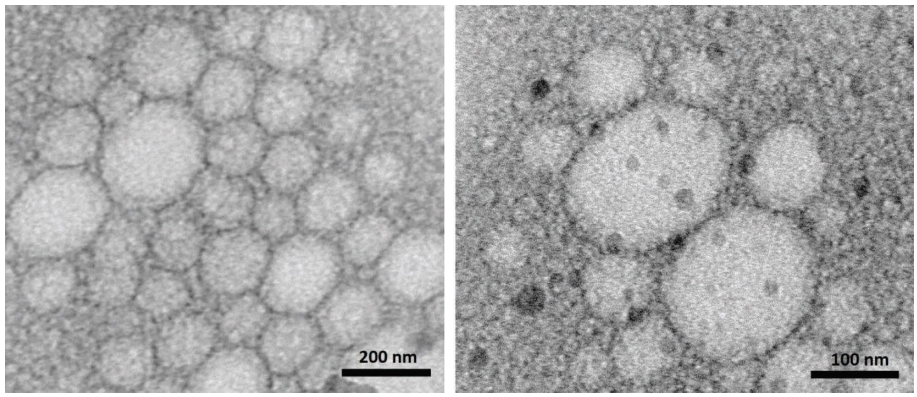

Figure 3: TEM images of the DEX-NPs prepared at 20 wt% of DEX content.

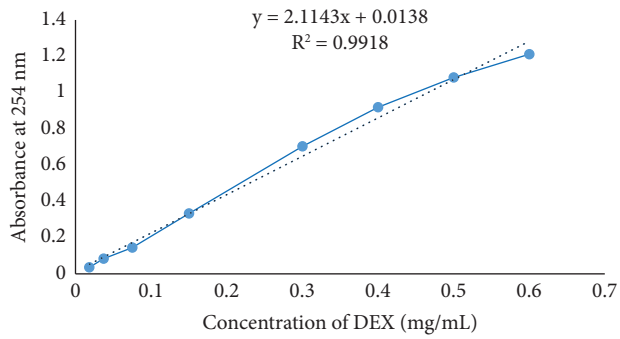

Figure 4: Calibration curve of DEX in DMSO.

Table 4: Stability of the prepared DEX-NPs upon storage at 4°C.

Sample	Measurement time			
	Freshly prepared	After 1 month	After 3 months	After 6 months
	AD (nm) ± SD [PDI ± SD]			
DEX-NPs	**151.8 ± 0.3** [0.098 ± 0.025]	**151.9 ± 1.9** [0.089 ± 0.022]	**153.2 ± 0.2** [0.103 ± 0.022]	**157.0 ± 0.8** [0.107 ± 0.049]

AD, average diameter; PDI, polydispersity index; SD, standard deviation. Data presented are averages of three independent experiments. The average diameters of the NPs are given in bold.

62.18% of initially accumulated DEX was released. In the control group, 61.78% of DEX was released from DEX powder in the first 24 hours, while by day 2, 88.38% of DEX was released from DEX powder.

The initial rapid (burst) release is normally attributed to release of drug molecules that are adsorbed or close to the surface of NPs which are known to be permeable to water [30]. In the second stage, release of DEX from DEX-NPs was sustained and slow due to the low rate of degradation of the pseudo-protein 8L6 by hydrolysis because of the presence of CO-NH amide groups in the backbone along with the easily hydrolysable ester bonds. For this reason, the rate of hydrolytic degradation of polyesters is generally higher than the rate of hydrolytic degradation of PEAs like pseudo-protein 8L6. It is interesting to compare the obtained results with the ones for DEX release from DEX-loaded poly-

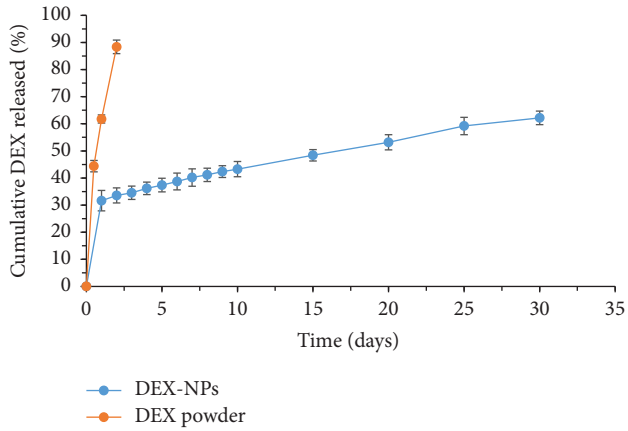

Figure 5: In vitro release profiles for DEX from DEX-NPs and DEX powder in PBS at 37°C. Note: data presented are averages of three independent experiments.

ε-caprolactone NPs (DEX-ECT2 NPs) reported by Krishnan et al. [27]. The authors showed that around 60% of DEX was released from DEX-ECT2 NPs during the first 2 days, while in our experiments, only 33.5% of DEX was released from DEX-NPs within 48 hours (Figure 5). Moreover, by day 7, in the case of DEX-ECT2 NPs, 91.4% of DEX was released, whereas in our experiments, only 40.1% of DEX was released from DEX-NPs during 7 days. It is obvious from the obtained results that the pseudo-protein 8L6-based NPs release molecules of drug much slower than poly-ε-caprolactone NPs. Therefore, the pseudo-protein 8L6-based NPs look more promising for prolonged and continuous drug delivery.

4. Conclusions

DEX-NPs were successfully obtained using the cost-effective nanoprecipitation method. The optimal/high polymer concentration in the organic phase during the preparation process of NPs for maximum drug entrapment was found, and the main characteristics of the prepared DEX-NPs were determined. DEX was successfully encapsulated into the pseudo-protein NPs with EE and DL of 55.1 and 10.2%, respectively. The kinetics of DEX release from DEX-NPs was studied as well and showed a biphasic drug release pattern with an initial burst release and further much more slow and continuous release. The elaborated DEX-NPs are suitable for medical applications in ophthalmology as drug delivery carriers.

Data Availability

The data used to support the findings of this study are included within the article.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Acknowledgments

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