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Gels and liposomes in optimized ocular drug delivery: Studies on ciprofloxacin formulations

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Abstract

Ciprofloxacin (CPFX) containing therapeutic systems were developed using gel- and liposome-based formulations to minimize tear-driven dilution in the conjunctival sac, a long-pursued objective in ophthalmology. Physicochemical properties (pH, osmolarity, viscosity, expansivity, membrane fluidity and *in vitro* CPFX release rate) of the preparations were studied by the appropriate methods. For gel preparation, the bioadhesive poly(vinyl alcohol) and polymethacrylic acid derivatives were applied in various concentrations. In our liposome-supported carrier systems, multilamellar vesicles from lecithin and α-L-dipalmithoyl-phosphatidylcholine provided the encapsulating agent. Electron paramagnetic resonance (EPR) spectroscopy was applied to study the molecular interactions in the ophthalmic formulations. The polymer hydrogels used in our preparations ensured a steady and prolonged active ingredient release. In addition, encapsulation of the CPFX into liposomes prolonged the *in vitro* release of the antibacterial agent depending on the lipid composition of the vesicles.

Keywords: Ciprofloxacin; Ocular drug delivery system; Gel; Liposome; Release

1. Introduction

Drug delivery in ocular therapy has long been a difficult task. Poor bioavailability of drugs from conventional ocular dosage forms is mainly due to the precorneal loss factors, which include tear dynamics, insufficient residence time in the conjunctival sac and non-productive absorption (Le Bourlais et al., 1998; Sultana et al., 2006).

Ciprofloxacin (CPFX) is a fluoroquinolone antibiotic against external infections of the eye, such as conjunctivitis, bacterial keratitis and keratoconjunctivitis (Appelbaum and Hunter, 2000). In addition to high potency, a desirable characteristic of topical fluoroquinolones is that concentration of the antibiotic should be maintained for sufficient long time above the minimum inhibitory concentration for relevant pathogens (Robertson et al., 2005). Efficacy of the marketed ophthalmic fluoroquinolone products, mostly aqueous solutions, is limited by poor ocular bioavailability (Lin et al., 1996; Wiechens et al., 1998–99), compelling the frequent dosing regimen and the concomitant patient compliance.

In order to improve ophthalmologic bioavailability of drugs and to lengthen the therapeutic action, introduction of novel delivery systems (Kaur et al., 2004) and enhanced vehicle viscosity have recently been proposed (Cho et al., 2003). Poly(vinyl alcohol) and polymethacrylic acid have been investigated among the possible vehicles (Lee et al., 1983; Guzek et al., 1998; Barnes and Nash, 1999; Gupta et al., 2001). The ability of disposable soft contact lenses to deliver lomefloxacin was also investigated. The contact lenses soaked in antibiotic produced higher levels of fluoroquinolone in both cornea and aqueous humor than that achieved by frequent-drop therapy for up to 8 h (Tian et al., 2001).

Other approaches include vesicular drug delivery systems, like liposomes (Bangham et al., 1965; Gregoriadis and Florence, 1993; Farkas et al., 2004). Active ingredients encapsulated in the lipid vesicles allow not only an improved solubility and transport

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through the cornea but liposomal delivery of drugs is also a tool for prolonged and controlled delivery (Sharma and Sharma, 1997; Pinto-Alphandary et al., 2000). Moreover, liposomes offer the convenience of an ophthalmic drop, and confinement of the action at the site of administration (Mainardes et al., 2005).

Our aim is to study the drug release from ciprofloxacin containing ophthalmic preparations. Here we describe the formulation and evaluation of CPFX containing hydrogels, and effects of liposomes on the drug release rate. Liposomal hydrogel system with ciprofloxacin has already been studied aiming the prevention of bacterial adhesion to catheters (DiTizio et al., 1998). Our approach is, however, completely different from that, since in case of eye drops the required consistency of a formulation should be closer to a liquid or "fluid gel" than to a "rigid" gel. To study the impact of viscosity on drug release, various concentrations of poly(vinyl alcohol) (PVA) (0.14–15% (m/m)) and polymethacrylic acid (PMA) (0.1–5%, m/m) were investigated (Hatakeyama et al., 2005) as vehicles for the formulation of CPFX eye drops. One of the aims of the present study is to evaluate the role of liposomes in the ophthalmic drug delivery, by encapsulating the CPFX into multilamellar liposomes prepared from lecithin (LEC) or α-L-dipalmithoylphosphatidylcholine (DPPC). In each case, the *in vitro* release of CPFX from the lipid vesicles was evaluated by spectrophotometry.

In order to optimize ophthalmic preparations it is necessary to take care of some special requirements. The most important physical and chemical parameters, such as pH, osmolarity, expansion and viscosity of the samples were measured.

In our study, the encapsulation efficiency, as a key parameter in liposomal drug delivery (Edwards and Baeumner, 2006), was also determined. Since molecular interactions between the encapsulated drug and its liposomal carrier system can have an influence on the drug liberation, electron paramagnetic resonance (EPR) spectroscopy was applied to monitor the molecular interactions between CPFX and lipid molecules (Bedard and Bryan, 1989; Montero et al., 1994; Cevc, 2000; Hernández-Borrell and Montero, 2003; Budai et al., 2004). We studied also the lipid composition and the phase transition parameters of lipids that influence the drug release.

2. Materials and methods

2.1. Preparation of ophthalmic solutions

2.1.1. Poly(vinyl alcohol) containing dilute gel (0.14% PVA)

PVA (molecular weight 72000 Da) was obtained from Reanal Inc. 30.0 g of 0.14% (m/m) of aqueous solution was prepared from PVA and distilled water (Aqua ad injectabilia). Isotonic conditions were fulfilled using 0.9% (m/m) sodium chloride (Sigma Chemical Co.); furthermore, 1.0% (m/m) microbiological preservative (7.0 g methyl-paraoxy-benzoate and 3.0 g propyl-paraoxy-benzoate dissolved in 90.0 g ethanol) was added to ensure the microbiological stability. The concentration of CPFX (Sigma Chemical Co.) was 0.1% (m/m) in the solution.

2.1.2. Polymethacrylic acid containing dilute gel (0.1% PMA)

PMA (940 NF) was obtained from Noveon Inc. 30.0 g of 0.1% (m/m) of aqueous solution was prepared from PMA and distilled water (Aqua ad injectabilia). The solution contained 4.0% (m/m) sorbitol (Sigma Chemical Co.), 1.0% (m/m) microbiological preservative (7.0 g methyl-paraoxy-benzoate and 3.0 g propyl-paraoxy-benzoate dissolved in 90.0 g ethanol) was added to ensure the microbiological stability. The solution contained EDTA disodium salt (complexon III) (Reanal Inc.) in a concentration of 0.1% (m/m). The concentration of CPFX was 0.1% (m/m) in the solution.

2.2. CPFX-containing gels

2.2.1. Preparation of CPFX-containing gels

PVA hydrogels were prepared in concentrations of 5, 7, 10 and 15% (m/m) – denoted as 5% PVA; 7% PVA; 10% PVA and 15% PVA. In order to get a more homogeneous gel one cycle of freezing (−15 °C) and thawing (100 °C) of the samples were carried out. PMA hydrogels in concentrations of 3% and 5% (m/m) were prepared. In case of CPFX-treatment the drug was finally added to the gels in a final concentration of 0.1% (m/m); this concentration is one third of the commercially available product Ciloxan® – Alcon, Fort Worth, Texas – used recently by Solomon et al. (2005).

2.3. Examination of physical parameters

2.3.1. pH and osmolarity

The pH of the dilute gels was measured with a Metrohm 6.0234.110 combined glass electrode. The electrode system was calibrated with four NIST buffers. Osmolarity was measured by the use of a Knauer 2320 type osmometer, which allows measurement of the osmolarity in case of dilute gels. (There is no regulation how to measure the pH or osmolarity in case of eye creams in Pharmacopoea Hungarica VIII.)

2.3.2. Viscometry

The viscosity of the prepared formulations was determined using a Haake RheoStress 150 Rheometer type rotational viscometer on 10 ml of the sample. The measurement was carried out at 37 °C at various angular velocities. A typical run involved changing the angular velocity from 1 rpm to 100 rpm at a controlled ramp speed. The device registered viscosity values in every 20 s.

2.3.3. Extensometry

The principle of the measurements is that 1 g of the gel between two horizontal parallel glass plates (15 × 15 cm) is exposed to increasing vertical pressure (1 g, 2 g, 5 g, 10 g, 20 g, 50 g, 100 g, 200 g, 500 g and 1000 g weights on the upper glass plate, during a time interval of 1 min). At the end of each time interval, the extent of the expansion (the size of the expanded spot) characterizes the expansion capacity of the given sample. Expansion measurements were carried out only on gels containing the gel forming polymer in a concentration of at least 10%

(CPFX-free and CPFX-treated 10% PVA and 15% PVA were studied). In case of lower polymer concentration, the hydrogels studied in present work possess such a low viscosity, which prevents a determination of their expansion capacity.

2.4. CPFX containing liposomes

2.4.1. Preparation of CPFX containing liposomes

Multilamellar vesicles (MLV) were prepared using the thin-film hydration method. 70 mg lipid, α-L-DPPC (Sigma Chemical Co.) or lecithin (LEC) (Sigma Chemical Co.) and the appropriate amount of CPFX were dissolved in absolute ethanol (Merck). The mixture was dried to thin-film under nitrogen stream. The rest of the solvent was removed from the lipid film in 30 min using a vacuum-line and the samples were stored in a desiccator overnight. Thin films of the lipid were hydrated above the main-transition temperature of DPPC and LEC, at ∼50 °C with 10.0 ml of the hydrating solution (0.14% PVA or 0.1% PMA as given in Section 2.1), vortexed during 45 min, resulting in a fine MLV suspension with a final lipid and CPFX concentration of 7 mg/ml and 0.1% (m/m), respectively. Liposomes neither were extruded, nor were the non-encapsulated CPFX-fractions removed. Based on liposome composition it can be calculated, that CPFX-treated DPPC and LEC liposomes contained the drug in a lipid to CPFX molar ratio of ∼3.16:1.

2.4.2. Measurement of encapsulation efficiency

400 μl of freshly prepared MLV-CPFX samples were centrifuged with an Eppendorf centrifuge (11000 × *g*, 20 min) through Microcon YM-10 Centrifugal Filter Devices (Millipore Inc.) with a cut-off value of 10 kDa. The concentration of CPFX in the filtrate, representing the amount of CPFX out of the liposomes, was determined by spectrophotometry (Unicam 2 UV/VIS spectrophotometer) at the wavelength of 273 nm. The absorbance of a 0.1% (m/m) CPFX hydrating solution served as 100% for determination of the encapsulation efficiency.

2.4.3. EPR measurements on liposomes

Liposomes were prepared as above, with the difference that the spin label 5-doxyl-stearic acid (SL-5) (Sigma Chemical Co.) was co-dissolved with the lipid in absolute ethanol. The molar ratio of the spin label to lipid molecules was 1:100. Spectra were registered with an EMX6 Bruker X-band on-line spectrometer. Temperature dependence of the spectra was measured by controlling the temperature within the sample with a precision of 0.1 °C. 100 kHz modulation frequency and maximally 2 G modulation amplitude were used. The applied microwave power was 15 mW; scan speeds of 167.77 or 335.5 s with 2048 points on 100 G field interval were taken. To characterize the fluidity of the lipid membrane in case of SL-5 the outer peak separation ($2A_{max}$) was determined at the temperatures of 20, 37 and 45 °C.

2.5. In vitro CPFX release studies

In vitro CPFX release measurements on the formulations was carried out by filling 10.0 g of CPFX (0.1%, m/m) loaded samples into vials in triplicate. The samples were separated with a cellulose acetate membrane possessing a pore size of 4 nm from the dissolution medium. The vials were immersed in 40.0 ml phosphate buffer (pH 7.4). The temperature and stirring rate were 37 °C and 50 rpm, respectively. Aliquots of 2.0 ml were taken at 30 min intervals from the release medium and replaced by equal volume of the receptor medium at each sampling point. The aliquot was diluted with the dissolution buffer and the amount of CPFX was determined at 273 nm using a Unicam 2 UV/Vis spectrophotometer.

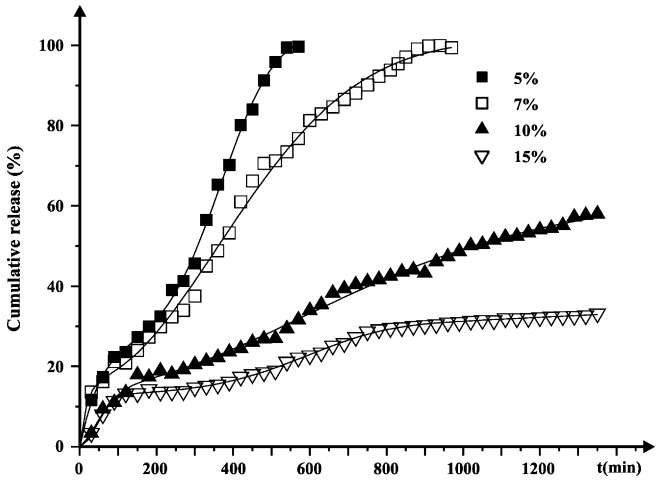

Fig. 1. Cumulative release of CPFX from PVA gels of various concentrations (5%, 7%, 10% and 15% (m/m)) *in vitro*. Concentration of CPFX was 0.1% (m/m) in all samples.

2.6. Data analysis of the release studies

Curves given in Figs. 1–3 are the results of a non-linear fit to the experimental points. The half-time of the release (Table 2) was determined using the best-fit parameters of the analysis. Weibull-function has been used during the analysis. In most cases, the best fit was obtained with a sum of two Weibull-functions of different release constants and shape parameters, thus description of the best fit is not possible with a single

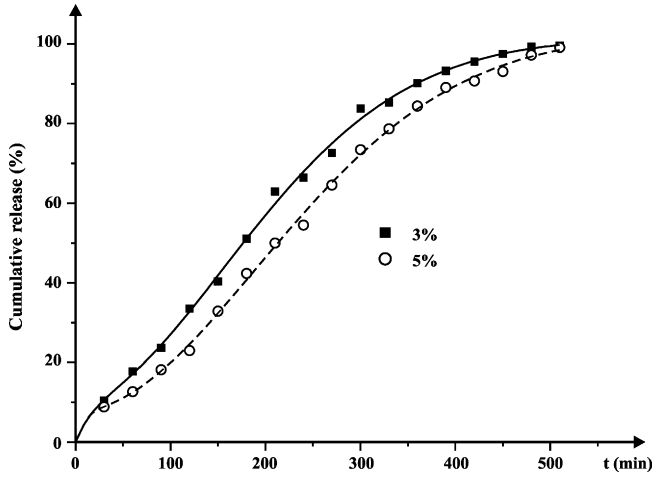

Fig. 2. Release of CPFX from PMA gels of various concentrations (3% and 5% (m/m)) with 0.1% (m/m) CPFX *in vitro*.

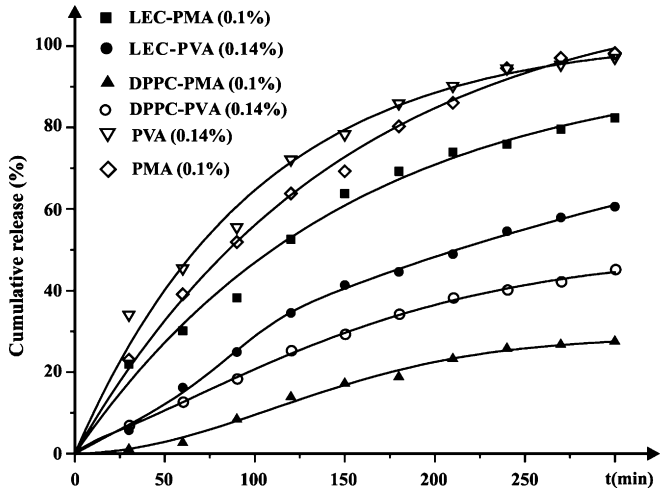

Fig. 3. Cumulative amount of CPFX released as a function of time from various formulations with 0.1% (m/m) CPFX *in vitro*.

release constant. This has the consequence that in characterization of the release process of the systems studied the release half-time was calculated using the best-fit parameters. Best fit to the experimental points were obtained by the Origin 6.0 software from Microcal Software Inc., USA. Standard errors for the fitting parameters were taken from the out-put of the software. Standard error of mean (SEM) for the half-time values given in Table 2 was calculated by the rule of error propagation.

3. Results and discussion

3.1. Physical and rheological characteristics

pH and osmolarity of the selected PVA and PMA dilute gels were measured with and without CPFX; the corresponding data are collected in Table 1. In the solution of 0.14% PVA, the 0.1% (m/m) CPFX caused a decrease of 1.6 pH. On the contrary, in case of 0.1% PMA a much smaller decrease, about 0.2 pH value, was measured in the presence of CPFX. pH of hydrogels containing liposomes was measured in the supernatant having centrifuged the samples. It was found that the presence of LEC or DPPC (which are mostly in zwitterionic form under the conditions studied) does not significantly alter the pH values of solutions compared to liposome-free gels (data not shown here). As expected, the CPFX-loaded samples are of slightly higher osmolarity than the corresponding CPFX-free ones. Comparing the values of Table 1 with those of the special requirements for the ophthalmic formulations – pH from ~6 to 9 and osmolarity between 220 and 450 mosmol/kg (Völker-Dieben et al., 1987) – our formulations satisfy the given requirements.

Plasticity is a desirable characteristic of the consistency of topical formulations (Bousmina, 1999). The rheological behavior of the sample is of great importance since it is applied in thin layers. For that reason it is necessary to prepare plastic formulations because of their low resistance to flow under high shear conditions, whereas the flow at rest is zero (Ruiz Martinez et al., 2007). The gels examined showed plastic properties and the steady state viscosities are collected in Table 1. The results reflect that increasing concentrations of hydrogels increase the viscosity of the samples. Evaluating the viscosity data of PVA and PMA gels with different concentrations of the gel forming material (between 0.14% and 10%, m/m for PVA and 0.1% and 5%, m/m for PMA), an exponential dependence was found. Semi-logarithmic plots of the viscosities against concentration can be well fitted with straight lines having regression coefficient very close to one (0.994 and 0.998 for CPFX-free and CPFX-treated PVA hydrogels, 0.971 and 0.975 for CPFX-free and CPFX-treated PMA gels, respectively).

It was found that extent of the expansion depends mainly on the concentration of the viscosity enhancer; moreover, the presence of CPFX does not have a significant effect on the size of the extended spot. Varying the loading between 0 and 1000 g, the size of the expanded spot increases; it is in the range of 6.23–7.63 cm^2 and 6.15–7.54 cm^2 for CPFX-free and CPFX-treated 10% PVA, respectively. Expanded spot sizes of 5.79–6.30 cm^2 and 5.65–6.08 cm^2 were determined for CPFX-free and CPFX-treated 15% PVA.

3.2. Studies on CPFX containing liposomes

Since the amount of the entrapped CPFX was calculated by measuring the non-entrapped portion of the drug in the fil-

Table 1
Physical parameters (pH, osmolarity and viscosity) determined for the formulations studied with and without CPFX-treatment

Type of gel	$cc_{Polymer}$ (%, m/m)	pH		Osmolarity (mosmol/kg)		Viscosity (mPa s)	
		CPFX- free	CPFX treated	CPFX-free	CPFX treated	CPFX-free	CPFX treated
PVA	0.14	6.8 ± 0.1	5.2 ± 0.1	304 ± 2	313 ± 1	1.7 ± 0.1	1.8 ± 0.1
	5.0	6.1 ± 0.2	5.2 ± 0.1	n.m.	n.m.	69 ± 3	54 ± 4
	7.0	5.9 ± 0.2	5.3 ± 0.2	n.m.	n.m.	238 ± 8	238 ± 7
	10.0	5.9 ± 0.2	5.3 ± 0.2	n.m.	n.m.	1285 ± 11	1313 ± 9
	15.0	5.9 ± 0.2	5.4 ± 0.2	n.m.	n.m.	1500 ± 15	1570 ± 22
PMA	0.1	6.5 ± 0.2	6.3 ± 0.1	362 ± 1	381 ± 1	2.5 ± 0.2	2.5 ± 0.1
	3.0	6.9 ± 0.1	6.7 ± 0.1	n.m.	n.m.	95 ± 2	92 ± 5
	5.0	6.9 ± 0.2	6.7 ± 0.2	n.m.	n.m.	329 ± 8	340 ± 7

In case of CPFX-treatment the CPFX concentration was 0.1% (m/m). Each value given in the table was calculated from $n = 3$ parallels, and are given by the mean ± S.D. n.m.: parameters cannot be measured due to the consistency of the samples with the device and method given in experimental part.

trate, the term encapsulation efficiency refers to CPFX localized both inside the entrapped aqueous volume of the liposomes and incorporated/bound into the bilayer. Encapsulation efficiency was determined for liposomes prepared from LEC or DPPC in the presence of a 0.1% (m/m) final CPFX concentration. PMA containing LEC and DPPC liposomes showed $11.2 \pm 3.5\%$ (average $\pm$ S.D.) and $13.7 \pm 3.9\%$ encapsulation, respectively. For PVA containing LEC and DPPC liposomes $14.1 \pm 2.9\%$ and $13.0 \pm 3.4\%$ encapsulation efficiencies were determined. Statistical analysis of the entrapment efficiencies does not show significant difference between the liposomes with different lipid composition. In our earlier experiments on similar MLV systems, the EPR signals outside of the MLVs were quenched by paramagnetic relaxation and we observed for the incorporated volume about 7% (Budai et al., 2004). Taking into account the encapsulation efficiency and the volume of the liposomes, the concentration of the CPFX in the liposomes is about 2 times higher than outside of the liposomes. This observation may well assume specific molecular interactions between the lipid molecules and CPFX.

According to the results of Bedard and Bryan (1989), the molecular interaction between CPFX and phospholipid containing bilayers is based on ionic and hydrophobic forces. It was also described (Merino et al., 2002) that CPFX interacts electrostatically at the lipid headgroups of liposomes bearing negative charge. Ofloxacin and lomefloxacin fluoroquinolones in our earlier works have been shown to interact mainly with the lipid head groups of liposomal membranes resulting in a slight increase of the membrane rigidity below the phase transition temperature of the bilayer (Budai et al., 2005).

Doxyl-group of the SL-5, used in the present study, are adjacent the lipid head group, apparently the most sensitive region in the fluoroquinolone-lipid interaction. In case of pure and CPFX-treated LEC- and DPPC-liposomes, we determined the outer peak separation values ($2A_{max}$) at selected temperatures. Greater values of this parameter correspond to less fluid environment. The presence of CPFX does not alter significantly the $2A_{max}$ values measured on LEC liposomes. The presence of CPFX in DPPC-liposomes causes an increase ($\sim$1 G) in the $2A_{max}$ values at temperatures below the phase transition temperature of the DPPC ($\sim$42 °C). Such a change indicates the formation of a more rigid bilayer structure in the presence of the antibacterial agent (the smallest acceptable change of $2A_{max}$ is about 0.3 G under the experimental conditions of the present study). Evaluating the role of gel forming PMA in liposomes it can be concluded that PMA does not cause a significant change in the membrane rigidity. At 20 °C we registered $2A_{max}$ values of 51.8 ± 0.4 G for PMA-free LEC liposomes, and 52.44 ± 0.3 G for PMA containing (0.1%, m/m) LEC liposomes. EPR spectroscopy allows investigation of liposomal fusions using the phenomenon of paramagnetic line-broadening (Béni et al., 2006). Using PVA and PMA at the respective 0.14% and 0.1% (m/m) concentrations no linewidth broadening has been observed, indicating that PVA or PMA at these concentrations does not cause fusion of the liposomes.

3.3. In vitro release studies

Fig. 1 shows the cumulative amount of the released CPFX for various concentrations of gel forming PVA as a function of time. Half-time values for the CPFX release are collected in Table 2. These values were determined from the plotted data as well as those that were determined based on the best-fit parameters (Antal et al., 1997). All the gels contained 0.1% (m/m) CPFX. 5% PVA and 7% PVA released half of the CPFX after 300 and 360 min, respectively. For these samples, the complete CPFX release can be observed after 550 and 900 min. On the contrary, only about 17% and 15% of the CPFX was released after 300 min from formulations containing 10% PVA and 15% PVA, respectively. Half-time values for the release in these latter cases are about 1000 and 5500 min for 10% PVA and 15% PVA, respectively. These observations suggest that for a formulation of prolonged release, higher initial drug concentration is necessary to ensure the therapeutic antibiotics level, compared to gels of lower PVA concentrations (Solomon et al., 2005).

In vitro drug release from PMA containing systems was also studied to evaluate the role of the gel forming material in ophthalmic systems. As Fig. 2 shows, the CPFX - cumulative release values are only slightly different between PMA gels with 3% and 5% concentration. Release profiles of 5% PVA and 5% PMA are remarkably close. This suggests that the release rate depends primarily on the concentration of the hydrogel, and secondarily on the type of the gel forming material (at least for these hydrogels). This has been further corroborated by our observations (Fig. 3) on 0.14% PVA and 0.1% PMA gels. The corresponding release half-times are 85 and 72 min for 0.1% PMA and 0.14% PVA hydrogels, respectively (Table 2). After 300 min, the whole amount of CPFX is released from both formulations. Use of dilute preparations – 0.1% PMA and 0.14% PVA – in combinations with lipid vesicles resulted in prolonged release. Up to a three-fold increase of the release half-time was detected incor-

Table 2
Half-times of CPFX-release from the various formulations used

Type of gel	Half-time of release[a] (min)	SEM for calculated best-fit[b] half-times (min)
0.14% PVA	72 (60–90)[c]	2.8
0.1% PMA	85 (60–90)	1.3
LEC-PMA 0.1%	110 (90–120)	5.8
3% PMA	176 (180)	4.6
LEC-PVA 0.14%	212 (210–240)	5.4
5% PMA	213 (210)	2.5
5% PVA	307 (300–330)	2.2
7% PVA	361 (360–390)	1.0
DPPC-PVA 0.14%	644 (n.d.)	5.4
10% PVA	1026 (1020–1050)	1.2
DPPC-PMA 0.1%	4614 (n.d.)	5.3
15% PVA	5507 (n.d.)	0.5

[a] Half-time values were determined using the best-fit parameters as given in the methods.

[b] SEM values were calculated according to the rule of error propagation using the standard errors of the best-fit parameters.

[c] Values in parentheses were determined from the figures of the experimental results. Intervals are given when no sampling time was in the close proximity of the 50% release. n.d.: values cannot be determined from the experimental data.

porating the CPFX into liposomes prepared from lecithin in case of low-concentration PVA-hydrogel (LEC-PVA 0.14%). About 80% and 60% of total CPFX can be measured in the dissolution buffers after 300 min in case of LEC-PMA 0.1% and LEC-PVA 0.14%, respectively. Among the liposomal formulations studied, the highest CPFX release half-time was obtained for DPPC-PMA 0.1% (4600 min), which is comparable to the release rate of a 15% PVA hydrogel without liposomes (5500 min; Table 2). In case of dilute PVA hydrogels (PVA 0.14%), values of the release half-time of CPFX changed from 72 min to 212 and 644 min for samples without liposome, with LEC and DPPC liposomes respectively. We found differences in the increase of release half-times between LEC and DPPC containing liposomal formulations. This can partially be attributed to the lipid composition, since LEC contains also unsaturated lipids that results in a higher fluidity at a given temperature than DPPC. In our work, liposomal formulations were prepared by hydrating the lipid thin film with appropriate concentration of PVA and PMA hydrogels. Thus, lipid vesicles produced by this method are covered by adsorbed layer of PVA or PMA (Mu and Zhong, 2006; Xie and Granick, 2002). Interaction between the liposomes and the polymer-coating depends on the types of polymer molecules and also on the fluidity of the liposomal membrane. In case of a dimyristoyl-phosphatidylcholine-PMA formulation it has been shown (Xie and Granick, 2002) that the molecular interaction between PMA as a week polyelectrolyte and the lipids depends mainly on the phase of the lipid bilayer, gel or liquid crystal. We suppose that differences in the lipid composition and the type of the polymer together might explain the differences found in the increase of release half-times. Concerning drug release kinetics, liposomal formulations of CPFX in dilute hydrogels behave like concentrated (3–10%, m/m) hydrogels without liposomes. The observed advantage of prolonged CPFX-release in liposomal formulations is due to the presence of multilamellar lipid vesicles and the adsorbed layer of polymers retarding the release. The examined gel forming auxiliary materials at the concentrations studied have no impact on the fusion of liposomes. Thus, they cannot influence the rate of CPFX-release by altering the encapsulated volume or size distribution of vesicles either.

A comparison of the release half-time values for LEC- and DPPC-liposomes, shows that DPPC prolongs the CPFX-release to a greater extent than LEC. Evaluating the molecular background, this phenomenon can be explained in terms of different composition and phase transition parameters of DPPC and LEC. α-L-DPPC used in our experiment is a synthetic product having only saturated palmithoyl chains. On the contrary, egg yolk lecithin contains also unsaturated chains; the ratio of saturated to unsaturated components is about 0.8. This difference in compositions leads to different main phase transition temperature of LEC and DPPC found to be between −15 to −7 °C and 42.5 ± 0.25 °C, respectively (Chapman et al., 1967; Budai et al., 2004). Since membrane permeability and membrane fluidity are co-dependent terms, drug formulations with LEC should be of higher permeability than with DPPC. Nevertheless, this effect can be modulated by the solubility/hydrophobicity of the drug encapsulated.

One can address the question of incorporating liposomes into more concentrated hydrogels for further prolongation of the CPFX-release. According to our observations (not shown here), there was only a slight difference between the drug releases from LEC-PVA 5% and liposome-free 5% PVA formulations. This observation suggests that for PVA hydrogels above the concentrations of 5% (m/m) the release rate is determined mainly by the viscosity of the system and contribution of the additional liposomal formulation is negligible. However, for PVA gels at concentrations below 5% (m/m), the LEC-liposomes can ensure a prolonged release profile compared to the corresponding liposome-free PVA gel. A further aspect on the use of higher hydrogel concentration or DPPC-formulation comes from the following observation. Use of 15% PVA or DPPC-PMA 0.1% formulation can result in a very low release rate. Thus, the drug concentration could remain below the therapeutic level. Thus, an optimal equilibrium between the release rate and the prolonged release time can be achieved by varying the hydrogel concentration and the initial drug concentration within a liposomal formulation of appropriate lipid composition.

4. Conclusion

Correlations can be demonstrated between the rheological parameters and the rate of CPFX release. Optimized combination of viscosity enhancers and vesicular delivery systems can result in the desired rheological characteristics and prolonged release. By increasing the concentration of the viscosity-increasing adjuvant and by use of liposomal formulation with appropriate lipid composition, higher drug concentration can be achieved at the site of action. Concomitantly the time of contact can be prolonged thus ocular bioavailability can be improved.

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