ANTI-GLAUCOMA DRUG-LOADED CONTACT LENSES PREPARED USING SUPERCRITICAL SOLVENT IMPREGNATION

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Abstract

Post-processing drug impregnation/dispersion of finished and/or commercially available polymer-based biomedical devices is a recent and attractive approach for the development of multifunctional biomedical devices and implants, drug release systems and tissue engineering applications. This strategy permits, after the polymer-based device is already prepared and processed, the impregnation and deposition of bioactive species on these items, according to the biomedical envisaged application and to the required therapeutic drug levels, without interfering with their chemical synthesis and other processing steps. Therapeutic ophthalmic articles, like drug-loaded contact lenses, are already known to improve drug absorption through the cornea which is a practical solution to improve the therapeutic efficiency in the treatment of several eye diseases, namely glaucoma, as well as to avoid the occurrence of undesired systemic side-effects. In this work, commercial silicone-based hydrogel contact lenses (Balafilcon A, Bausch & Lomb® PureVision™) were impregnated with two anti-glaucoma drugs (acetazolamide and timolol maleate) using a discontinuous Supercritical Solvent Impregnation (SSI) methodology. Pressure and temperature, as well as impregnation time and depressurization rate, were kept constant (17 MPa, 40 ºC, 90 min, 0.06 MPa/min, respectively), in order to study and elucidate the co-solvent (ethanol and water) nature and concentration effects on the impregnation efficiencies and processed contact lenses properties. Solvent mixtures of $scCO_2$+EtOH and $scCO_2$+H_2O (5, 10 and 15 % molar) were used. *In vitro* drug release kinetics studies were performed and the amounts of released drugs were determined spectrophotometrically. Other analytical techniques (DSC, oxygen permeability and SEM) were employed. Results demonstrated the feasibility of preparing acetazolamide and timolol maleate impregnated therapeutic Balafilcon A contact lenses using $scCO_2$+EtOH and $scCO_2$+H_2O solvent mixtures and are discussed in terms of how the employed solvent mixtures nature and compositions have influenced drug loading efficiency, drug release profiles and contact lenses physical and thermomechanical properties.

INTRODUCTION

Despite glaucoma drugs efficiency, their costs may impact the decisions in glaucoma medical management and, currently, its treatment constitutes an important financial burden to health care systems worldwide including the USA [1]. Because of the known limitations associated to topical eye drops formulations, several ophthalmic polymer-based drug delivery systems have been developed in the last 30 years. Furthermore, and since the drug is easily eliminated/washed by tear circulation when topical formulations are applied directly into the eye, this drug loss presents also a potential risk of systemic side-effects occurrence. On the

contrary, it is already well known that the use of drug-loaded contact lenses may overcome these issues because drugs will be released in a controlled fashion and will remain mainly trapped between the lens and the cornea, on a thin fluid layer named post-lens tear film, thus avoiding drug washing as well as increasing drug residence time and, consequently, its bioavailability.

Ophthalmic drugs were already incorporated into several polymeric controlled drug release systems (CDR's) systems with, for example, cortisone and retinol [2], ibuprofen [3], pilocarpine [4] and indomethacin [5]. Other ophthalmic drug incorporation studies were also performed by SSI and using a CO_2 and CO_2+cosolvent mixtures as solvents [6][7][8].Timolol maleate (a beta-blocker) was loaded in some polymers, like acrylates and chitosan/carbopol blends, by reverse phase evaporation [9] and by quasi-emulsion solvent diffusion [10], respectively. The solubility of this drug was measured in scCO_2+EtOH mixtures and its solubility (using 0.2 % (molar) of EtOH) was found to be approximately 0.15 g/L, at 40 ºC and 19 MPa [11]. For timolol maleate, the maximum recommended therapeutic dose (MRTD) is 1mg/kg-bw/day [12]. Acetazolamide is usually administered orally (as pills/tablets) or topically (as eye drops). However, and mainly because of its low bioavailability, the required high doses necessary for these administration routes can induce the occurrence of systemic side effects. The recommended acetazolamide defined daily dose (DDD), for both enteral and parenteral routes, is 0.75 g [13]. Acetazolamide solubility was measured in scCO_2+EtOH (5 % molar), and was 0.027 g/L, at 40 ºC and 17.2 MPa [14].

The SSI technique is based on the use of a supercritical solvent (for example, carbon dioxide) as a carrier for the drug into a preformed polymeric matrix (films, pellets, particles, etc.) [15]. The supercritical solvent will also swell/plasticize the polymeric matrix, thus increasing its volume and facilitating a fast diffusion of the mobile phase containing the drug. The depressurization step will lead to drug deposition inside the polymer by two main mechanisms: impregnation and dispersion/deposition [16]. Small amounts of cosolvents can improve the overall process, by increasing solvent polarity (thus promoting drug solubility) or by increasing polymer swelling and plasticization. Therefore, the amount of impregnated/dispersed drug can be controlled by changing the operational pressure and temperature process conditions, as well as by choosing the appropriate cosolvents and varying their compositions.

This work is part of global research project involving the development and the process optimization of polymer-based ophthalmic drug delivery systems using supercritical CO_2 impregnation/dispersion methods, namely therapeutic contact and intraocular lenses, hydrogels and particles for topical applications and biodegradable copolymer blends for ophthalmic implants [6][7][8]. In this work, we studied cosolvent nature and concentration effects on the impregnated drug amounts in contact lenses (Balafilcon A, Bausch & Lomb® PureVision™). Ethanol and water were the chosen cosolvents and were employed at 5, 10 and 15% (%molar), in order to increase acetazolamide and timolol maleate solubility in the scCO_2 phase and to verify other possible effects on employed contact lenses. Pressure and temperature, as well as impregnation time and depressurization rate, were kept constant (17 MPa, 40 ºC, 90 min, 0.06 MPa/min, respectively). Drug release assays were carried out for the impregnated systems. Some processed contact lenses thermomechanical properties, as well as their oxygen permeability, were analyzed and compared to non-processed articles. Surface morphology was observed using scanning electronic microscopy.

MATERIALS AND METHODS

The employed supercritical impregnation apparatus was described in Patent EP 1 611 877 A1 (Unit I) [6][8][17]. Employed contact lenses were water-containing (wet) Balafilcon A

(Bausch & Lomb® PureVision™). Ethanol (EtOH) and water (H_2O) were the chosen cosolvents and were employed at 5, 10 and 15% (%molar). Acetazolamide (ACZ, hydrophobic and having a relatively low solubility in $scCO_2$) and timolol maleate (TM, hydrophilic and having a relatively low solubility in $scCO_2$) were the chosen ophthalmic drugs. Pressure and temperature, as well as impregnation time and depressurization rate, were kept constant (17 MPa, 40 ºC, 90 min, 0.06 MPa/min, respectively). Magnetic stirring was always employed in order to dissolve and homogenize the drug in compressed fluid mixtures ($scCO_2$+EtOH or $scCO_2$+H_2O).

Kinetics of drug release studies were performed for all prepared systems using a spectrophotometric method. Release experiments were carried out during 8 hours for timolol maleate and 2.5 h for acetazolamide, in physiological serum, under agitation (100 rpm) and at 37 ºC. Released drug concentration was calculated using previously determined calibration curves. Total (accumulated) drug released amounts were determined for all experiments. Kinetics experimental data were fitted using a linear regression analysis by a curve composed of three straight lines. Fitting was done by minimizing the least regression error (in the least square sense) using the fminsearch function of Matlab (R2007a).

Contact lenses oxygen permeability was measured (in 0.9% NaCl), using a Createch permeometer, model 210T (Rehder Development Company, Castro Valley, CA USA), fitted with a radius cell and keeping the polarographic cell in a sealed box. Obtained results are presented as $Dk\times10^{12}$, $(cm^2/s)(cm^3\ O_2/cm^3.mmHg)$ and compared to the literature.

Thermal analysis (on processed and non-processed freeze-dried lenses) was performed by differential scanning calorimeter (DSC - Q100 model, TA Instruments). Nitrogen was used and the samples were equilibrated at - 40 ºC until 400 ºC. Hermetic aluminum pan was used as reference and freeze-dried samples were analyzed. All assays were duplicated.

Scanning Electron Microscopy (SEM) was employed to observe lenses surfaces and cross sections (after cut), at 25 kV (Jeol, JSM-5310 model, Japan). Samples were coated with gold, approximately 300 Å, in an argon atmosphere, and observed for processed and non-processed samples.

RESULTS

Figure 1 presents the results of drug released masses (ACZ and TM) for the contact lenses processed at 17 MPa and 40 ºC, and using 5% (molar) of cosolvents (EtOH and H_2O). It is considered that the total impregnated drug mass corresponds to the maximum value of total (accumulated) drug released for all systems. As can be seen, ACZ was impregnated in lower extents for both employed cosolvents. Moreover, when H_2O is the employed cosolvent and when compared to EtOH results, ACZ was just impregnated with very low amounts of drug. This is probably due to the high hydrophobic character of this drug: when small amounts of H_2O dissolve in CO_2, it will not just increase the polarity of the supercritical phase but also will introduce some hydrophilic character to that phase, thus not increasing too much the solubility of ACZ in it. Furthermore, H_2O is also present in the contact lenses (wet lenses, 36% (w/w) of water) and this will not favor the impregnation/deposition of a hydrophobic drug.

On the contrary and for the above explained reasons, TM is a highly hydrophilic and water-soluble drug and thus will be impregnated in higher extent than ACZ when H_2O is employed as the cosolvent (more than 10× of impregnated drug). However, an important issue should be noted: the H_2O solubility in $scCO_2$ is very low at these experimental conditions, approximately 5430×10^{-6} (molar) [18]. Therefore, the indicated 5% (molar) H_2O composition was not really achieved in the supercritical mobile phase.

On the contrary, the indicated 5% (molar) composition of EtOH was achieved since EtOH is highly soluble in $scCO_2$ at the experimental conditions. Therefore, the polarity of the mobile phase was increased in a higher extent (than the obtained with the lower amount of H_2O) and TM solubility on the mobile was strongly increased. In addition, TM is a hydrophilic drug thus having a higher compatibility for the wet contact lenses, promoting its impregnation. This explains the obtained highest impregnation results for TM+5% (molar) EtOH.

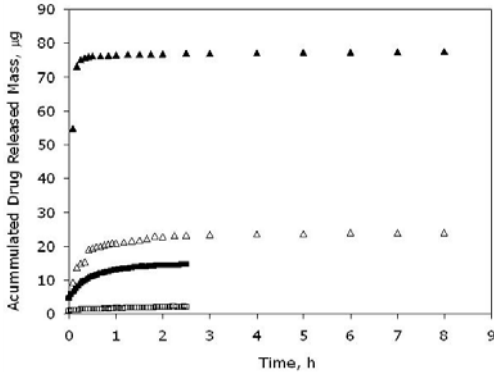
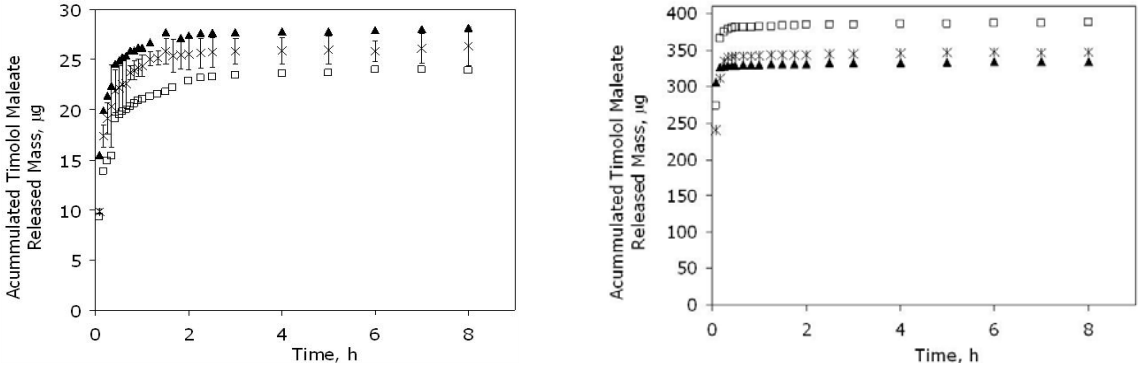


Figure 1. Accumulated drug released mass from contact lenses, using 5% (molar) of cosolvent: ■ ACZ-EtOH □ ACZ-H_2O ▲ TM-EtOH Δ TM-H_2O

Figure 2 shows the results of TM released masses for the contact lenses processed at 17 MPa and 40 ºC, and using 5%, 10% and 15% (molar) of cosolvents (H_2O and EtOH). For H_2O, **Figure 2(A)**, and as referred above, the indicated compositions do not correspond to the true compositions at the mobile supercritical phase, which must be all equal and approximately 5430×10^{-6} (molar) [18]. Therefore, adding more H_2O into the cell will not increase the amount of water in that phase. Despite it seems that there is some apparent tendency (higher amounts of H_2O will correspond to higher amounts of impregnated drug), the replicated experiments (performed with 10% (molar) of H_2O) showed that the obtained standard deviation will enclose the results obtained with 5% and 10% of H_2O. Therefore, we can conclude that there is no real difference between all these experiments and the obtained drug released values are quite similar (~25µg) for all indicated compositions.

However, when EtOH is the cosolvent, **Figure 2(B)**, it seems that there is a clear tendency: as EtOH composition increases from 5 up to 15%, the amounts of impregnated drug will decrease. This tendency can be explained by the increase of solvent mixture polarity (for higher EtOH concentrations) and to the resulting much higher TM solubilities in that phase. Therefore, the partition coefficient of TM with the fluid phase will be favored over its partition coefficient with the contact lenses [6][8].



(A) H_2O **(B) EtOH**

Figure 2. Accumulated drug released mass from contact lens processed at 17 MPa, 40 ºC, using H_2O (**A**) and EtOH (**B**): □ 5 % ✕10 % ▲ 15 % (molar)

A cross-section of a TM impregnated contact lens is presented in **Figure 3**. TM particles can be observed in the impregnated lens cross-section thus showing that the employed method can impregnate drugs relatively deep into the contact lens structure. Same conclusions were taken from the observation of SEM micrographs of ACZ impregnated contact lenses.

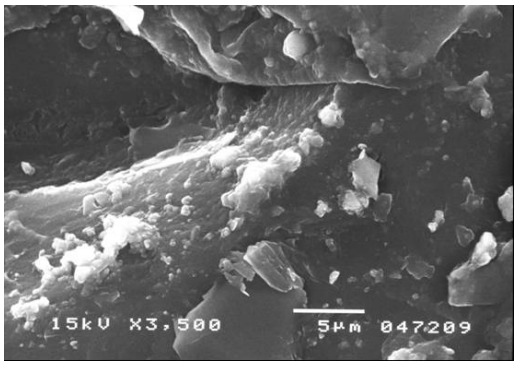


Figure 3. Cross-section SEM micrograph of a TM-impregnated contact lens, using 5% EtOH (molar)

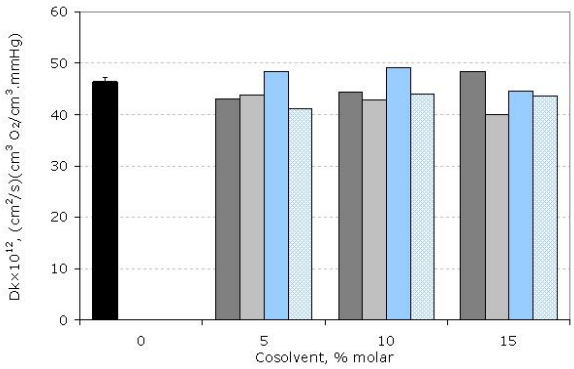


Figure 4. O_2 permeability of TM-impregnated contact lenses using cosolvents. Not released: ■ EtOH and ■ H_2O; After release: ■ EtOH, ■ H_2O and ■ Control

Table 1 presents the correlated drug release kinetics parameters. Obtained correlation errors (SSE) were quite small. The most important periods for drug release are the constant release rate period (C_{RR}), mainly due to drug at/near the surface, and the final diffusional release rate (D_{RR}). The corresponding kinetic parameters are mass transfer rate, M_{CRR} and M_{DRR} (µg/h) (mass ratio of drug in the polymer for solvent phase) and the duration of it, t_{CRR} and t_{DRR} (h). The falling rate period (M_{FRR} and t_{FRR}), is located between the two above described periods and their correlated values are not presented. For TM, and in the C_{RR} period, it is clear that mass transfer rates are much slower for the case of H_2O as the cosolvent. This is due to the corresponding lower amounts of impregnated drug, which will lead to lower drug gradients and to slower release velocities. For ACZ, the same behavior is observed and can be explained by the same reasons. ACZ presents also a much lower solubility in the release medium than TM which, in addition to the lower drug gradients, will lead to much lower release velocities than in the case of TM. For the D_{RR} period, a quite similar release behavior can be observed for both drugs. It is also clear that, for all systems, the period of constant release (t_{CRR}) is quite small when compared to the period of diffusional release (t_{DRR}) due to the initial burst release caused mainly by drug located at/near the surface.

Table 1.Correlated drug release kinetic parameters for contact lenses impregnated by SSI (CO_2+H_2O/EtOH)

Kinetic parameters	**Timolol Maleate**						**Acetazolamide**	
	H_2O 5%	H_2O 10%	H_2O 15%	EtOH 5%	EtOH 10%	EtOH 15%	H_2O 5%	EtOH 5%
M_{CRR}, µg/h	93.5	87.0	154.8	2738.5	10329.4	3243.8	106.2	253.8
t_{CRR}, h	0.150	0.224	0.135	0.130	0.020	0.090	0.055	0.038
M_{DRR}, µg/h	0.41	0.14	0.18	0.92	1.01	0.83	1.20	0.39
t_{DRR}, h	7.59	7.86	7.82	7.08	6.99	7.17	1.83	1.94
SSE	0.0758	0.0347	0.0316	0.0017	0.0024	0.0015	0.0476	0.0252

By comparison to control lenses (non-impregnated, non-processed in $scCO_2$ and non-released), oxygen permeability experiments showed that contact lenses O_2 permeability was kept for all employed experimental conditions (**Figure 4**). Despite it seems there is a slight permeability increase for some of the employed experimental conditions, this variation can be included in the measurement error. These values are in accordance with literature, considering the same contact lens (Balafilcon A) and the same lens power (-8) [19].

DSC analysis was performed for processed and non-processed freeze-dried lenses. Results showed that glass transition temperatures were not altered after the impregnation process (90.9 ± 2.11 ºC) and after drug release experiments (90.65 ± 2.05 ºC). Moreover, obtained glass transition results are in good agreement with literature values for Balafilcon A [20].

Therefore, we may conclude that Balafilcon A contact lenses O_2 permeability and glass temperature transition are kept after the impregnation and drug release processes, and that the

operational parameters, $scCO_2$ and employed cosolvents/drugs did not alter these extremely important contact lenses product parameters.

CONCLUSIONS

Results demonstrated the feasibility of preparing acetazolamide and timolol maleate impregnated therapeutic Balafilcon A contact lenses using $scCO_2$+EtOH and $scCO_2$+H_2O solvent mixtures. Different cosolvents and different cosolvent concentrations can lead to different impregnated drug amounts, and this knowledge can be a helpful tool to impregnate and to control the impregnated amounts of drugs of different hydrophilic/hydrophobic characters, besides the use of other operational conditions such as temperature and pressure.

Furthermore, Balafilcon A contact lenses O_2 permeability and glass temperature transition were kept after the impregnation and drug release processes, and the operational parameters, $scCO_2$ and employed cosolvents/drugs did not alter these extremely important contact lenses product functionalities/parameters as ophthalmic devices.

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