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Modification of ocular and systemic absorption of timolol from ocular inserts by a buffering agent and a vasoconstrictor

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Summary

Alkyl monoesters of poly(vinyl methyl ether-maleic anhydride) (PVM-MA) are promising bioerodible polymers for controlling drug release. The dissolution of these polymers and drug release from them are highly dependent on the pH on the polymer surface. In this study, we examined the effects of disodium phosphate buffer on in vivo release, and on ocular and systemic absorption of timolol given in matrices of monoisopropyl ester of PVM-MA ocularly in rabbits. The vasoconstrictor methaoxedrine was added to some matrices to reduce the systemic absorption of timolol. Timolol concentrations in tear fluid and plasma were measured using a radioreceptor assay. [^{3}H]Timolol-labeled matrices were used to study ocular absorption of timolol. The unbuffered matrix yielded a lower peak concentration of timolol in tear fluid (64 ± 9 (S.E.) μg/ml) and a lower steady-state concentration (1.0 ± 0.1 ng/ml) in plasma 3 h later than the buffered one (C_{max}, tear fluid = 104 ± 8 μg/ml; C_{max}, plasma = 7.3 ± 1.1 ng/ml). Thus, disodium phosphate increases the rate of timolol release from the inserts several fold in tear fluid. Co-administration of methaoxedrine in buffered matrices decreased the peak timolol concentration in plasma about 3-times and increased that in tear fluid about 2-fold. In iris-ciliary body, administration of buffered matrices resulted in timolol concentrations that were comparable with the levels after eye drop instillation. Compared to the unbuffered matrices, disodium phosphate without or with methaoxedrine at least doubled the concentration ratio of iris-ciliary body to plasma. The best iris-ciliary body/plasma concentration ratios were achieved with the buffered matrices containing methaoxedrine. Conjunctival and nasal vasoconstriction by methaoxedrine improves the ocular to systemic concentration ratio of timolol several fold. Compared to eye drops timolol administration in the inserts reduced the systemic β-blocking activity significantly.

Introduction

Topically applied timolol is the most commonly used drug in the treatment of open-angle glaucoma. Several ophthalmic drugs such as timolol (Kaila et al., 1986), betaxolol (Polansky and Alvarado, 1985), adrenaline and dipivalyl adrenaline (Anderson, 1980) and methaoxedrine (Kumar et al., 1986), have been shown to absorb to the systemic circulation after topical application to the eye. As much as 74% of the timolol applied in eye drops has been shown to absorb to the systemic circulation (Chang and Lee, 1987). Systemic absorption of timolol may cause severe respiratory and cardiovascular side-effects, especially in patients with predisposing respiratory or cardiovascular diseases (Nelson et al., 1986). Thus, it is

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necessary to reduce the systemic absorption of topically applied ophthalmic timolol.

The ocular bioavailability of several drugs can be improved by prolonging the ocular contact time or by increasing the corneal drug permeability (Lee and Robinson, 1986). A prolongation of ocular contact time by nasolacrimal occlusion reduced systemic absorption of timolol eye drops and, thus, indirectly enhanced ocular drug absorption (Chang and Lee, 1987). Nasolacrimal occlusion for 5 min nearly doubled the ocular bioavailability of timolol. Systemic absorption of drugs can also be reduced by applying the drug in a polymer matrix instead of eye drops. This has been shown with an *n*-butyl poly(vinyl methyl ether-maleic anhydride) matrix containing pilocarpine (Urtti et al., 1985). Recently, we have shown (Urtti et al., 1990) that the peak timolol activity in plasma was at least 17-fold lower when the drug was administered in an experimental silicone device instead of eye drops. The relative ocular timolol bioavailability after controlled drug delivery was about 2-times greater than from eye drops. The silicone device is, however, a model system not intended for treatment of glaucoma.

Alkyl monoesters of poly(vinyl methyl ether-maleic anhydride) (PVM-MA) are bioerodible polymers that are used to control drug release (Heller et al., 1978; Heller and Trescony, 1979; Urtti, 1985; Urtti et al., 1985). The dissolution of such polymers is highly dependent on the pH of the surrounding medium (Heller et al., 1978). Especially the microclimate pH on the polymer surface tends to decrease if the buffering capacity of the dissolution medium is low (Heller et al., 1978). This is due to the ionization of the carboxylic groups in the polymer. We have shown previously that drug release from the monoesters of PVM-MA in vitro can be modified by adding basic salts to the polymer matrix to increase the microclimate pH on the polymer surface (Finne et al., 1989). Since the buffering capacity of tear fluid is poor (Longwell et al., 1976) dissolution of alkyl esters of PVM-MA is retarded (Urtti et al., 1985). Consequently, the aim of this study was to regulate the dissolution of these polymers and drug release from them in the tear fluid by adding basic buffer to the polymer.

Another goal of the present investigation was to determine whether it is possible to increase further the ocular/systemic absorption ratio of timolol given in polymer matrices by including a vasoconstrictive additive, methaoxedrine, in the matrices. Recently, we have shown that methaoxedrine decreases the rate of systemic timolol absorption when administered as an eye drop (Urtti and Kyyrönen, 1989). Pigmented rabbits were used as the animal model in all studies.

Materials and Methods

Materials

Timolol was used as the maleate salt (INTERx Research Corp., MSDRL, Lawrence, KS, U.S.A.) and methaoxedrine as the hydrochloride (Egachemie, Albuch, F.R.G.). DL-Propranolol was purchased from Sigma (St. Louis, MO, U.S.A.). The monoisopropyl ester of PVM-MA was used as a 50% isopropanol solution of the polymer (Gantrez^{R} ES-335, GAF Europe, Esher, U.K.). The radioligand was (−)-[^{3}H]CGP-12177 (spec. act. 48.8–53.0 Ci/mmol, radiochemical purity 99%; Amersham International, Bucks, U.K.). [^{3}H]-Timolol (spec. act. 2.1–8.5 Ci/mmol, radiochemical purity 98.2–99.0%) was a gift from Merck, Sharp & Dohme, Inc. (Rahway, NJ, U.S.A.). T-61 euthanasia solution was from American Hoechst Corp. (Sommerville, NJ, U.S.A.). Lipoluma^{TM} and Lumasolve^{TM} were purchased from Lumac (Schaesberg, The Netherlands). ACS^{R} scintillation liquid was from Amersham International (Bucks, U.K.).

Mixed-breed pigmented rabbits of both sexes (1.4–4.0 kg) were used in this study. Before experiments, the rabbits were housed singly under standard laboratory conditions: 10 h dark/14 h light cycle, 20–21° C temperature, 40–70% relative air humidity. Food and water were given to the animals ad libitum. During the experiment, the animals were kept in wooden restraint boxes.

Preparation of polymer matrices

Matrices composed of the monoisopropyl ester of PVM-MA were prepared by solvent casting from organic solvents as described earlier (Finne

et al., 1989). The amount of added timolol maleate was 50 mg in 2.5 g of the polymer. Circular matrices (diameter, 5 mm; weight, 6.0–6.9 mg) were cut with a cork bore from the film and were kept in a desiccator until use. Each matrix contained 125 μg of timolol maleate corresponding to 25 μl of commercially available 0.5% timolol solution.

Buffered polymer films containing 12.9% disodium phosphate (2.11 mmol) with or without 10 mg (0.05 mmol) of methaoxedrine were also prepared by solvent casting as described earlier (Finne et al., 1989). The amounts of methaoxedrine and disodium phosphate in inserts were 20 and 752 μg, respectively.

For tissue measurements, tritiated matrices were prepared using [^{3}H]timolol (34.5 or 172.5 nCi/μg timolol base) as a tracer.

Timolol in plasma and tear fluid

Matrices of the monoisopropyl ester of PVM-MA were carefully applied in the lower conjunctival sac of rabbits. Plasma and tear fluid samples were collected at different intervals during a period of 8 h. Blood samples were taken from the cannulated ear artery. Plasma was separated by centrifugation (2000 × *g*, 4 min) and stored at −20 °C until analysis. Tear fluid samples (1 μl) were collected from the lower marginal tear strip using microcapillaries at different times after application of the matrices. When the capillary was not filled completely, the sample volume was calculated by measuring the filled portion of the capillary and the total length of the capillary tube. The capillaries were flushed several times with phosphate buffer. Tear fluid samples were diluted in 5 ml of phosphate buffer. Recovery of timolol from the capillaries was 99% using this procedure.

Timolol concentration in plasma and tear fluid samples was measured according to a modified radioreceptor assay of Wellstein et al. (1984). In the assay, displacement of a β-antagonist, (−)-[^{3}H]CGP-12177, from β_2-receptors of rat reticulocyte membranes by timolol is measured. Rat reticulocyte membranes were obtained as described by Wellstein et al. (1984). In a total volume of 300 μl, 50 μl reticulocyte membranes (500 μg protein) in phosphate buffer were incubated for 60 min at 25 °C with 50 μl (30 nCi) of (−)-[^{3}H]CGP-12177, 180 μl of plasma and 20 μl of 310 mosM sodium phosphate buffer (pH 7.4). Standard curves were run for each experiment separately. To generate the standard curves, blank plasma was used and incubation was performed in the presence of 1–20 nM timolol. Non-specific binding (~ 3%) was determined by incubation in 10^{-5} M propranolol solution. Timolol concentration in tear fluid samples was measured by incubating 200 μl of the diluted tear fluid in phosphate buffer. After incubation, bound and free radioligand were separated as described earlier (Urtti and Kyyrönen, 1989).

Timolol in ocular tissues

Rabbits were killed by injection of T-61 euthanasia solution (i.v.) at 0.5, 2.5, 4.0 or 8.0 h after application of [^{3}H]timolol-labeled matrices of monoisopropyl PVM-MA in the lower conjunctival sac of rabbits. Aqueous humor was withdrawn from proptosed eyes with a needle and syringe and placed in preweighed polyethylene liquid scintillation vials. After weighing, ACS scintillation liquid (5 ml) was added. The proptosed eyes were enucleated, frozen in liquid nitrogen and stored at −20 °C until dissection. Cornea, iris-ciliary body, conjunctiva, and sclera were dissected. To each tissue 100 μl of distilled water was added and the tissues were solubilized in 1 ml of Lumasolve™ at 40–50 °C.

10 ml of an isopropanol-Lipoluma™ mixture (0.3 : 10) was added to the solubilized tissue samples after cooling. The iris-ciliary body samples were first bleached with 0.3 ml of hydrogen peroxide (35%) for 15–30 min. To eliminate chemiluminescence, 100 μl of 0.1 N HCl was added to the iris-ciliary body samples after addition of the scintillation liquid. The samples were stored in darkness overnight before determination of radioactivity.

Analysis of the results

Areas under the curves (AUC_{0-8h}) of timolol activity in plasma and tear fluid were calculated using the trapezoidal method (Gibaldi and Perrier, 1982). Peak β-blocking activity (C_{max}) in plasma and tear fluid was determined from the data points.

Statistical significance of the differences was assessed by carrying out the Mann-Whitney U-test or variance analysis. $P < 0.05$ was considered to be a statistically significant difference.

Results and Discussion

Timolol in tear fluid

The surface of monoisopropyl PVM-MA softened within a few minutes after application in the eye. In contrast to unbuffered matrices, the buffered matrices with or without methaoxedrine dissolved in tear fluid in 8 h. Inserts did not cause any irritation in rabbit eyes.

Compared with unbuffered matrix ($C_{max} = 64 \pm 9$ μg/ml at 4 h), administration of timolol in polymer matrix with disodium phosphate already resulted in a 61% higher peak in tear fluid at 30 min (Fig. 1A). Peak timolol concentration in tear fluid was further increased by adding methaoxedrine in the buffered matrices (Fig. 1A, Table 1). Methaoxedrine did not affect the time of peak appearance. The AUC_{0-8h} values of timolol in tear fluid were not significantly different between the groups (Table 1).

Timolol clearance from the tear fluid was calculated by dividing the dose (125 μg) by the mean AUC_{0-8h} in tear fluid after administration of buffered matrices. Timolol clearance from tear fluid for the buffered matrices without or with methaoxedrine was 8.2 and 6.5 μl/min, respectively. These values are fairly close to those determined earlier (Francouer, 1987). The data from administration of the unbuffered matrices could not be used in the determinations of clearance due to their incomplete dissolution and drug release in the tear fluid during the experiment.

During the experiment, the matrices containing disodium phosphate dissolved more rapidly in the tear fluid than the unbuffered inserts. This has been demonstrated previously in vitro (Finne et al., 1989), where the rate of timolol release from matrices with 2.11 mmol of disodium phosphate was more than twice as fast as that from unbuffered matrices. Addition of methaoxedrine to the buffered matrices did not change timolol release in vitro (unpublished results). The difference between timolol release from buffered and unbuffered matrices appears to be greater in vivo than in vitro. This is suggested by the large difference in the levels and times of peak timolol concentrations in the tear fluid (Fig. 1A).

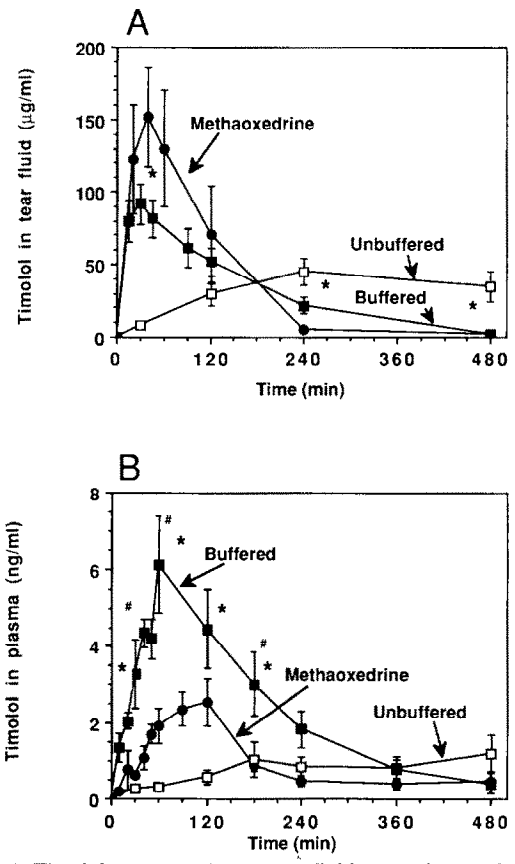

Fig. 1. Timolol concentration in tear fluid (μg/ml) ($n = 6–18$) (A) and in plasma (ng/ml) ($n = 6–12$) (B) after application of 125 μg timolol maleate in monoisopropyl PVM-MA matrices in both eyes of rabbit. Means ± S.E. are shown. (*) Buffered vs unbuffered, $p < 0.05$ (tear fluid, variance analysis; plasma, Mann-Whitney U-test); (#) buffered vs methaoxedrine, $p < 0.05$ (tear fluid, variance analysis; plasma, Mann-Whitney U-test).

Timolol in plasma

The concentration profiles of timolol after administration of unbuffered and buffered matrices in plasma (Fig. 1B) resemble those in tear fluid (Fig. 1A). The peak timolol concentration (7.3 ± 1.1 ng/ml) in plasma occurred at 60 min with the buffered matrices (Fig. 1B, Table 1). Methaox-

TABLE 1

Pharmacokinetic parameters in tear fluid and plasma after application of 125 μg timolol maleate in monoisopropyl PVM-MA matrices in both eyes of rabbits

Treatment	Tear fluid			Plasma		
	C_{max} (μg/ml)	t_{max} (min)	AUC_{0-8h} (h ng ml^{-1})	C_{max} (ng/ml)	t_{max} (min)	AUC_{0-8h} (h ng ml^{-1})
Buffered	104 ± 8 [a] (34)	30	255 ± 32 (18)	7.3 ± 1.1 [a] (12)	60	21.9 ± 3.9 [a] (9)
Buffered + methaoxedrine	192 ± 35 (8)	45	319 ± 121 (6)	2.8 ± 0.6 (7)	120	7.2 ± 1.8 (3)
Unbuffered	64 ± 9 [b] (14)	240	263 ± 48 (12)	2.4 ± 0.7 [b] (7)	480	5.3 ± 1.4 [b] (5)

The matrices were unbuffered or buffered with 752 μg disodium phosphate without or with methaoxedrine (20 μg). Means ± S.E. (n) are presented. C_{max}, peak timolol concentration in tear fluid or plasma; t_{max}, time of timolol peak concentration (from Fig. 1); AUC_{0-8h}, area under the timolol concentration in tear fluid or plasma vs time curve. [a] $p < 0.05$ buffered vs methaoxedrine (variance analysis); [b] $p < 0.05$ buffered vs unbuffered (variance analysis).

edrine lowered the peak timolol concentration about 3-times and with methaoxedrine, the peak levels were observed at 2 h (Fig. 1B, Table 1). After administration of unbuffered matrix, a steady-state level of 1.0 ± 0.1 ng/ml in plasma was achieved at 3 h (Fig. 1B). When timolol was previously given in eye drops (5 mg/ml, 25 μl) in one eye of the rabbit, C_{max} amounted to 17.0 ± 2.0 ng/ml (Urtti and Kyyrönen, 1989). Relative to the dose, administration of timolol in monoisopropyl PVM-MA matrices decreased the peak in plasma to 1/34, 1/4, and 1/12 compared to timolol eye drops (Urtti and Kyyrönen, 1989) with the unbuffered matrices and with the buffered matrices without and with methaoxedrine, respectively.

The area under the plasma curve (AUC_{0-8h}) of the buffered matrix was 3- or 2-times larger than that of the unbuffered matrix or buffered matrix with methaoxedrine, respectively (Table 1). Administration of timolol in the buffered matrix with methaoxedrine and the unbuffered matrix decreased the plasma AUC_{0-8h} to 1/3 of the plasma AUC_{0-3h} of timolol eye drops (Urtti and Kyyrönen, 1989). After eye drop administration the β-blocking activity was essentially zero at 3 h. Since the unbuffered matrices were not completely dissolved at 8 h, the dose delivered during this time remains unknown and the AUC values cannot be compared with those of other groups. The buffered matrices without methaoxedrine resulted in an AUC value of timolol in plasma identical to that seen with eye drops (Urtti and Kyyrönen, 1989). The results suggest that with the inserts containing methaoxedrine, both the rate and extent of systemic timolol absorption were decreased. Without methaoxedrine only the rate of timolol absorption was affected.

Methaoxedrine is an α_1-receptor agonist and resulted in a decrease in systemic timolol absorption, probably by constricting the conjunctival vessels (Meyer and Fraunfelder, 1980). This effect is also reflected in the form of elevated timolol concentrations in the tear fluid (Fig. 1A). The in vivo results correspond quite well with those of the in vitro experiments (Finne et al., 1989), although the effect of a basic additive on drug release appears to be greater in vivo than in vitro. This might be due to the differences in mixing conditions. The relative significance of the neutralizing effect of the buffer on drug release may be greater in tear fluid due to the small convective transport of hydrogen ions from the polymer surface.

Timolol in ocular tissues

Since timolol is not metabolized in rabbit eyes (Putterman et al., 1985) the radioactivity measured in the tissues represents unmetabolized timolol.

In all ocular tissues, peak concentrations of timolol after application of the buffered matrices with or without methaoxedrine were observed much earlier than after application of the unbuffered ones (Figs 2 and 3). This is due to the faster release of timolol from the buffered matrices

in the tear fluid. The peaks in timolol concentrations in cornea (Fig. 2A), conjunctiva (Fig. 2B) and sclera (Fig. 2C) were observed at 0.5 h after application of the buffered matrices with or without methaoxedrine. With the unbuffered matrices, however, peak levels were achieved at 4.0 h in conjunctiva (Fig. 2B) and at 8.0 h in cornea

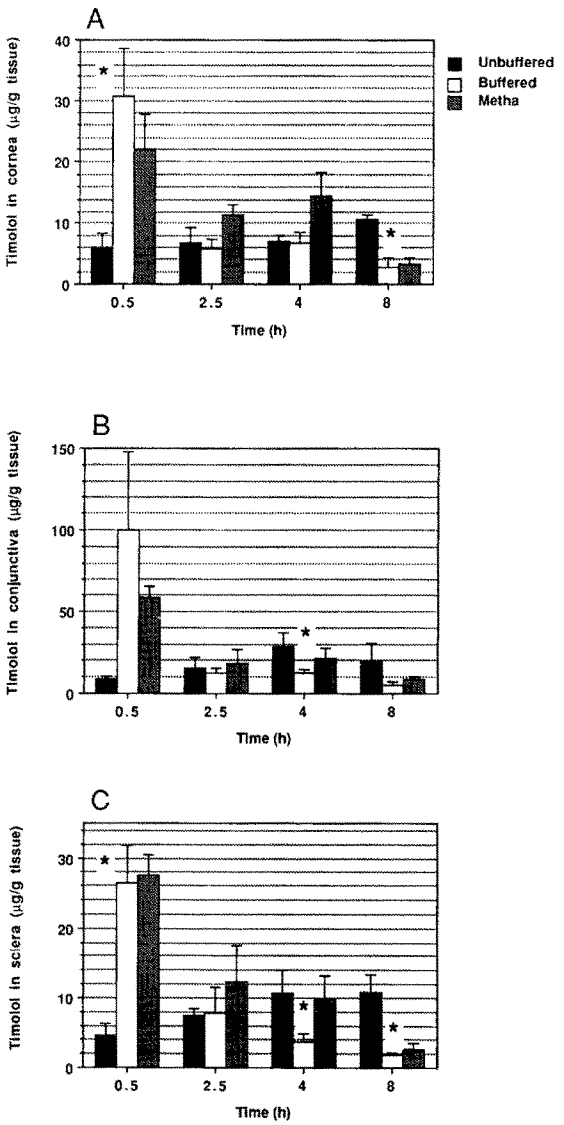

Fig. 2. Timolol concentration (μg/g tissue) in cornea (A), conjunctiva (B), and sclera (C) after application of 125 μg timolol maleate in monoisopropyl PVM-MA matrices in both eyes ($n = 4$) of rabbit. Means $\pm$ S.E. are shown. (*) Buffered vs unbuffered, $p < 0.05$ (Mann Whitney U-test).

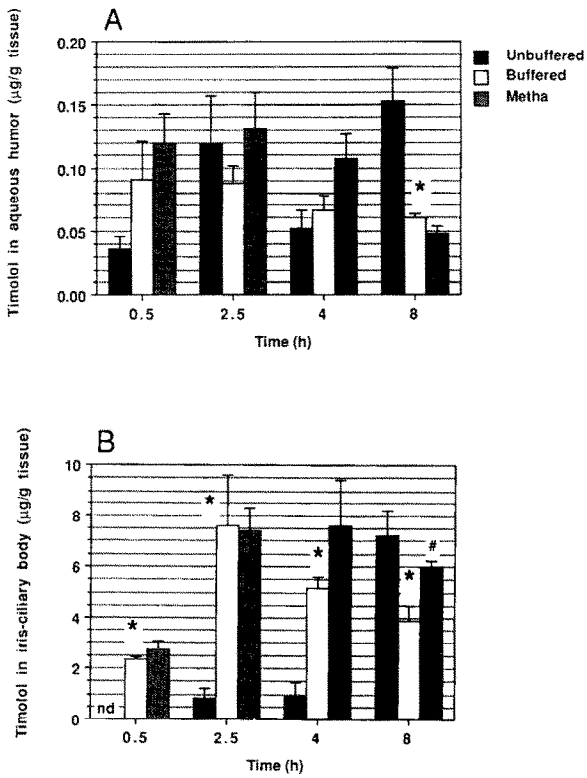

Fig. 3. Timolol concentration (μg/g tissue) in aqueous humor (A) and iris-ciliary body after application of 125 μg timolol maleate in monoisopropyl PVM-MA matrices in both eyes ($n = 4$) of rabbit. Means $\pm$ S.E. are shown. (*) Buffered vs unbuffered, $p < 0.05$ (Mann Whitney U-test); (#) buffered vs methaoxedrine, $p < 0.05$ (Mann Whitney U-test); nd, timolol concentration not detectable.

(Fig. 2A), sclera (Fig. 2C) and iris-ciliary body (Fig. 3B). In aqueous humor, the peak in timolol concentration occurred at 0.5 and 2.5 h after application of the buffered matrices without (91.3 ± 29.8 ng/g) and with methaoxedrine (132.4 ± 28.5 ng/g), respectively (Fig. 3A). With the unbuffered matrices, the peak timolol levels (153.1 ± 26.0 ng/g) in aqueous humor appeared only at 8.0 h (Fig. 3A). In addition, timolol release from unbuffered matrices is slow at first which is indicated by the flat tear fluid and plasma curves and is also reflected in the timolol concentrations in ocular tissues.

In iris-ciliary body, the target tissue of timolol, the highest timolol concentrations were achieved with the buffered matrices (7.6 ± 2.9 μg/g) at 2.5 h and with buffered timolol-methaoxedrine

TABLE 2

Area under timolol concentration vs time curve (AUC_{0-8h}) (h μg g^{-1}) and ocular tissue/plasma (pl) AUC_{0-8h} ratio in cornea (CO), conjunctiva (CJ), sclera (SC), aqueous humor (AH) and iris-ciliary body (ICB) after application of 125 μg timolol maleate in monoisopropyl PVM-MA matrices in both eyes (n = 4) of rabbits

Treatment	CO	CO/pl	CJ	CJ/pl	SC	SC/pl	AH	AH/pl	ICB	ICB/pl
Buffered	72.7	3320	190.5	8698	60.1	2745	0.57	26	38.3	1747
Buffered + methaoxedrine	93.7	13106	180.2	25200	88.3	12352	0.77	108	49.3	6894

matrices at 4.0 h (7.6 ± 1.8 μg/g) after application. With methaoxedrine timolol concentration in iris-ciliary body remained higher than with the buffered matrices without methaoxedrine. This may be due to the methaoxedrine-induced decrease in blood flow in iris-ciliary body (Meyer and Fraunfelder, 1980). The difference was, however, statistically significant only at 8.0 h (Fig. 3B). Compared with timolol eye drops (125 μg timolol) and a silicone device releasing timolol at a rate of 7.2 μg/h, the buffered matrices and those with methaoxedrine resulted in a timolol concentration of only one half of the value for the 5 mg/ml eye drops and about the same as that of the silicone device in iris-ciliary body during the experiment (Urtti et al., 1990). Since timolol is also an effective antiglaucomatous drug at instilled concentrations of 2.5 mg/ml, it is obvious that both types of buffered matrices yielded therapeutic timolol concentrations in iris-ciliary body.

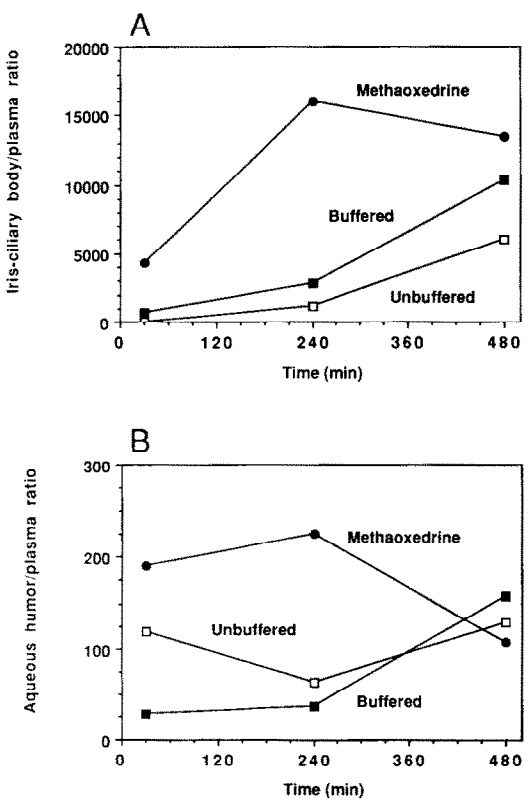

Fig. 4. Timolol concentration ratio of iris-ciliary body to plasma (A) and aqueous humor to plasma (B).

Application of timolol in the buffered matrices with methaoxedrine yielded higher timolol concentrations in cornea (Fig. 2A), conjunctiva (Fig. 2B), sclera (Fig. 2C), and iris-ciliary body (Fig. 3B) at 8 h than after the buffered matrices without methaoxedrine. This is probably due to the vasoconstrictor effect of methaoxedrine.

Table 2 lists the values of AUC_{0-8h} in ocular tissues and AUC_{0-8h} ratios between ocular tissues and plasma after application of timolol maleate in buffered PVM-MA matrices with and without methaoxedrine. The data for the unbuffered matrix have been omitted from Table 2 because of differences in the kinetics of timolol release between the unbuffered and buffered matrices both in vitro and in vivo. Addition of methaoxedrine to the matrices increased the ocular tissue/plasma AUC_{0-8h} ratios 3–5-fold compared to the buffered matrices without methaoxedrine. With the exception of conjunctiva the AUC_{0-8h} values were 28–47% higher in the case of matrices containing methaoxedrine as compared to those without the vasoconstrictor.

The relative safety of drug delivery systems in terms of their systemic side-effects can be described by the ratio between the ocular and sys-

temic drug concentrations (Chang et al., 1987). Compared to the unbuffered matrices, matrices with disodium phosphate increased the concentration ratio of timolol between iris-ciliary body and plasma about 2-fold at 4–8 h (Fig. 4A). The effect of methaoxedrine-containing matrices was at least 10-fold at 0–4 h and 1.3–2.2-fold at 8 h as compared to the other matrices (Fig. 4A). Similar, albeit smaller, effects of methaoxedrine can be observed in the aqueous humor/plasma timolol concentration ratio (Fig. 4B). The results show that conjunctival vasoconstriction by methaoxedrine improves the ocular to systemic concentration ratio of timolol several fold.

Conclusions

Disodium phosphate, a basic additive, in the matrices of monoisopropyl PVM-MA increased the release of timolol significantly in vivo. This effect appears to be greater in vivo than in vitro. Basic additives offer a means of modifying in vivo drug release from esters of PVM-MA and possibly also from other bioerodible poly(carboxylic acids).

The buffered matrices with or without methaoxedrine yielded a therapeutic timolol concentration in iris-ciliary body, but decreased the peak timolol concentration in plasma 4.6–12.1-fold compared to eye drop instillation. Thus, systemic peak concentrations of timolol can be reduced significantly by administering the drug in a controlled release device. Methaoxedrine, a vasoconstrictor, further decreases the systemic absorption of timolol.

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References

Anderson, J.A., Systemic absorption of topical ocularly applied epinephrine and dipivefrin. *Arch. Ophthalmol.*, 98 (1980) 350–353.

Chang, S.C., Bundgaard, H., Buur, A. and Lee, V.H.L., Improved corneal penetration of timolol prodrugs as a means to reduce systemic drug load. *Invest. Ophthalmol. Vis. Sci.*, 28 (1987) 487–491.

Chang, S.C. and Lee, V.H.L., Nasal and conjunctival contributions to the systemic absorption of topical timolol in the pigmented rabbit: implications in the design of strategies to maximize the ratio of ocular to systemic adsorption. *J. Ocul. Pharmacol.*, 3 (1987) 159–167.

Finne, U., Kyyrönen, K. and Urtti, A., Drug release from monoisopropyl ester of poly(vinyl methyl ether-maleic anhydride) can be modified by basic salts in the polymer matrix. *J. Control. Rel.*, 10 (1989) 189–194.

Francouer, M.L., PhD Thesis, Department of Pharmaceutical Chemistry, University of Kansas, 1987.

Gibaldi, M. and Perrier, D., *Pharmacokinetics*, 2nd edn, Dekker, New York, 1982, pp. 445–449.

Heller, J., Baker, R.W., Gale, R.M. and Rodin, J.O., Controlled release by polymer dissolution. I. Partial esters of maleic anhydride copolymers-properties and theory. *J. Appl. Polym. Sci.*, 22 (1978) 1991–2009.

Heller, J. and Trescony, P.V., Controlled drug release by polymer dissolution. II. Enzyme-mediated delivery device. *J. Pharm. Sci.*, 68 (1979) 919–921.

Kaila, T., Huupponen, R. and Salminen, L., Effects of eyelid closure and nasolacrimal duct occlusion on the systemic absorption of ocular timolol in human subjects. *J. Ocul. Pharmacol.*, 2 (1986) 365–369.

Kumar, V., Schoenwald, R.O., Barcellos, W.A., Chien, D.S., Folk, J.C. and Weingeist, T.A. Aqueous vs. viscous phenylephrine. I. Systemic absorption and cardiovascular effects. *Arch. Ophthalmol.*, 104 (1986) 1189–1191.

Lee, V.H.L. and Robinson, J.R., Topical ocular drug delivery: recent developments and future challenges. *J. Ocul. Pharmacol.*, 2 (1986) 67–108.

Longwell, A., Birss, S., Keller, N. and Moore, D., Effect of topically applied pilocarpine on tear film pH. *J. Pharm. Sci.*, 65 (1976) 1654–1657.

Meyer, S.M. and Fraunfelder, F.T., Pharmacology of ocular drugs. 3. Phenylephrine hydrochloride. *Ophthalmology*, 87 (1980) 1177–1180.

Nelson, W.L., Fraunfelder, F.T., Sills, J.M., Arrowsmith, J.B. and Kuritsky, J.N., Adverse respiratory and cardiovascular events attributed to timolol ophthalmic solution 1978-1985. *Am. J. Ophthalmol.*, 102 (1986) 606–611.

Polansky, J.R. and Alvarado, J.A., Isolation and evaluation of target cells in glaucoma research: hormone receptors and drug responses. *Curr. Eye Res.*, 4 (1985) 267–279.

Putterman, G.J., Davidson, J. and Albert, J., Lack of metabolism of timolol by ocular tissues. *J. Ocul. Pharmacol.*, 1 (1985) 287–291.

Urtti, A., Pilocarpine release from matrices of alkyl halfesters

of poly(vinyl methyl ether/maleic anhydride). *Int. J. Pharm.*, 26 (1985) 45–55.

Urtti, A. and Kyyrönen, K., Ophthalmic epinephrine, phenylephrine, and pilocarpine affect the systemic absorption of ocularly applied timolol. *J. Ocul. Pharmacol.*, 5 (1989) 127–132.

Urtti, A., Pipkin, J.D., Rork, G., Sendo, T., Finne, U. and Repta, A.J., Controlled drug delivery devices for experimental ocular studies with timolol 2. Ocular and systemic absorption in rabbits. *Int. J. Pharm.*, 61 (1990) 241–249.

Urtti, A., Salminen, L. and Miinalainen, O., Systemic absorption of ocular pilocarpine is modified by polymer matrices. *Int. J. Pharm.*, 23 (1985) 147–161.

Urtti, A., Sendo, T., Pipkin, J.D., Rork, G. and Repta, A.J., Application site dependent ocular absorption of timolol. *J. Ocul. Pharmacol.*, 4 (1988) 335–343.

Wellstein, A., Palm, D., Wiemer, G., Schafer-Korting, M. and Mutschler, E., Simple and reliable radioreceptor assay for beta-adrenoreceptor antagonists and active metabolites in native human plasma. *Eur. J. Clin. Pharmacol.*, 27 (1984) 545–553.