International Journal of Pharmaceutics, 61 (1990) 235–240
Elsevier

IJP 02072

Controlled drug delivery devices for experimental ocular studies with timolol
1. In vitro release studies

Arto Urtti [1,*], James D. Pipkin [1,2], Gerald Rork [1,2] and A.J. Repta [1,2]

[1] *Department of Pharmaceutical Chemistry, The University of Kansas, Lawrence, KS 66045 (U.S.A.) and* [2] *INTERx Research Corp., Merck Sharp & Dohme Research Laboratories, Lawrence, KS 66047 (U.S.A.)*

(Received 20 June 1989)
(Modified version received 22 December 1989)
(Accepted 23 December 1989)

Key words: Timolol; Drug delivery device; Silicone tubing; Ocular drug delivery; Controlled release; Release rate

Summary

Controlled drug delivery devices which can be easily tailored and conveniently prepared to release ocular drugs at specified controlled rates can be useful in studies of ocular drug delivery and drug response. The devices were fabricated by injecting 6.0, 9.0 or 12.5 μl of aqueous borate buffered solutions of timolol (2.5–20.0 mg/ml) into end-plugged pieces of silicone tubing and the release of timolol was studied in vitro. An initial lag in timolol release was avoided by storing the devices for an appropriate time (3–9 h) after filling the devices. The appropriate storage time was dependent on the steady-state flux of timolol, indicating that timolol was binding to the silica filler in the silicone tubing. Timolol was released at a constant rate (0.7–7.2 μg/h) for 8 h from the devices when the initial core pH was 8.64 and when the devices were stored for an appropriate time before an experiment. The steady-state release rate of timolol was increased 2.4-fold when the pH inside the device was increased from 8.34 to 9.24. As expected, the observed release rate increased with increased drug concentration in the device core and with increased length of the device. The permeability of timolol in the silicone membrane walls was $2.48–6.03 \times 10^{-9}$ cm^2 s^{-1} depending on the composition of the inner core solution. Thickness of the end-plugs (0.5 or 3.5 mm) did not affect timolol release from the devices, when the volume of timolol solution in the devices was 12.5 μl.

Introduction

Ocular drugs are usually applied as aqueous eye drops. In many cases less than 1% of the dose is absorbed in the eye. Most of the dose entering the anterior chamber is absorbed transcorneally (Maurice and Mishima, 1984). The poor bioavailability is caused partly by the rapid decrease ($t_{1/2}$ = 30–90 s) of drug concentration in precorneal tear fluid (Lee and Robinson, 1979). The rapid precorneal elimination of drugs given in eye drops is mainly due to conjunctival absorption and solution drainage by gravity, induced lacrimation and normal tear turnover (Lee and Robinson, 1979). Because of poor ocular bioavailability, many ocular drugs are applied in high concentrations. This causes both ocular and systemic side-effects, which

Correspondence: J.D. Pipkin, INTERx Research Corp., Merck Sharp & Dohme Research Laboratories, Lawrence, KS 66047, U.S.A.

* *Present address:* Department of Pharmaceutical Technology, University of Kuopio, P.O. Box 6, 70211 Kuopio, Finland.

are often related to high peak drug concentrations in the eye and in systemic circulation (Havener, 1983).

Ocular bioavailability of many topically applied drugs has been increased by using vehicles that retard precorneal drug loss (Chrai and Robinson, 1974; Grass and Robinson, 1984). These vehicles include ointments, gels, latex systems, liposomes, nanoparticles, polymer matrices and Ocusert™ reservoir device (for references see Shell, 1984). Controlled drug release can also lower peak drug concentrations in the systemic circulation (Urtti et al., 1985).

In spite of numerous studies of ocular bioavailability, which have utilized various vehicles, there are very few studies on the effects of different defined delivery rates and patterns (first-order, zero-order) on ocular and systemic drug absorption and on pharmacological responses in the eye. In most cases the release characteristics of drugs from the vehicles have not been studied or the actual in vivo concentration-time profile in the tear fluid probably differs from that observed in vitro owing to drainage of particles or viscous vehicles from the conjunctival sac. In the conjunctival sac, the erosion properties of water-soluble polymers may also be different from those in in vitro experiments. Nevertheless, knowledge of the relationship between the actual pattern of drug delivery and drug distribution or drug response is important when delivery systems with optimal input rates are to be developed for ocular therapy.

This study is an extension of the work initiated by Baustian and Mikkelson (1985) and its goals were to develop non-erodible devices from safe materials that could be conveniently constructed, easily altered and used to determine the effects of different rates and patterns of drug delivery on drug distribution in rabbit eyes and on drug responses in both animals and humans. Timolol maleate was used to demonstrate the release characteristics afforded by the devices.

Materials and Methods

Preparation of the devices

Medical grade silicone tubing (Silastic™, Dow Corning, Midland, MI), with the dimensions 1.46 × 1.94 × 0.24 mm (inner diameter × outer diameter × wall thickness), was cut to give lengths of 15 mm. Silastic ™ Adhesive A (Dow Corning, Midland, MI) was used to form a plug at one end of the tubing. The adhesive was allowed to cure overnight. The other end was plugged with the adhesive to leave a 5.0, 6.5 or 8.5 mm length of empty tubing inside the device. When forming the second plug, a 27-gauge needle was inserted through the first plug providing pressure relief. When the adhesive had cured, the ends of the devices were cut to leave 0.5 mm of adhesive in both ends of each device.

End-plugged silicone tubings were filled with 6.0, 9.0 or 12.5 μl of aqueous timolol solutions using a syringe with a fixed needle (Precision Sampling B-110-FN, Supelco, Belleforte, PA) inserted through one end. A 27-gauge needle was inserted through the other end providing pressure relief. The cured adhesive end-plugs self-sealed after removal of the needles. The solutions contained 0.15 M sodium borate (0.10 M at pH 9.24) and 2.5, 5.0 or 20.0 mg/ml of L-timolol equivalents. Timolol maleate was used (Merck, Sharp & Dohme Research Laboratories, Rahway, NJ) and the pH values of the solutions were adjusted to 8.34, 8.64, 8.84 or 9.24 at 32° C with 10% HCl or 5 N NaOH. After filling, the devices were stored in 98% relative humidity at room temperature (above lead nitrate) to minimize evaporation of water through the wall membranes. The loss of water in 14 h of storage was 2.9 ± 0.9% (mean ± S.D.).

Studies of in vitro release

Release of timolol from the devices was studied using the rotating bottle method (NF XIV). Speed of rotation was 30 rpm. The dissolution medium was 2.0 ml of 10 mM isotonic phosphate buffer (pH 7.40) at 32° C. Samples of 100–125 μl were withdrawn and replaced by dissolution medium. The samples were analyzed at 294 nm using reverse-phase HPLC with a 3 μm C18 column (50 × 4.6 mm) (Analytichem International, Harbor City, CA). The mobile phase contained 12.5% acetonitrile. The aqueous phase was 2.8 ml triethylamine and 10 ml glacial acetic acid per liter of water. The pH was adjusted to 4.0 using NaOH. The reten-

tion time of timolol in this system was about 4 min.

The release rate of timolol from the devices was calculated as the slope of the least-squares fit line of a plot of μg released vs time. Fraction of initial burst or lag in timolol release was quantitated as the amount released at time zero (y-intercept of the least-squares fit line) divided by the cumulative amount released after 8 h.Lag in timolol release was indicated by a negative y-intercept.

Results and Discussion

The storage time of the devices affected timolol release; i.e. short storage times resulted in lag times before steady-state timolol release was achieved and long storage times caused an initial burst of timolol release (Fig. 1). The lag time is due to inadequate equilibration of the drug between the solution core of the device and the rate-limiting membranes (Baker and Lonsdale, 1974; Flynn et al., 1974). Steady-state timolol release is achieved only after a lag time, during which the drug partitioned into the membrane. A burst effect, on the other hand, is caused by the rapid initial release of timolol that has accumulated in the silicone membranes during the storage time. If the diffusing drug molecules are not absorbed or adsorbed to the membrane constituents, lag time will not be dependent on the drug flux through the membranes (Baker and Lonsdale, 1974). However, in our experiments, lag times were influenced by the flux through the membranes (Fig. 2), which indicates that a constant amount of timolol is bound to be membrane regardless of the flux rate (Higuchi and Higuchi, 1960). Probably, a fixed amount of timolol is adsorbed per unit of silica filler in the Silastic™ membranes (Most, 1970). For each flux rate of timolol there was an ideal storage time after which timolol is released without a lag or burst (Fig. 1). After the ideal storage time the amount of timolol that has partitioned into the membranes is the same as that in the membranes at pseudo-equilibrium during drug release. Approximate ideal storage times (t_s) were determined empirically for several formulations with different rates of timolol release. Thereafter, the various formulations were equilibrated for the appropriate ideal storage times to avoid a burst or lag during timolol release.

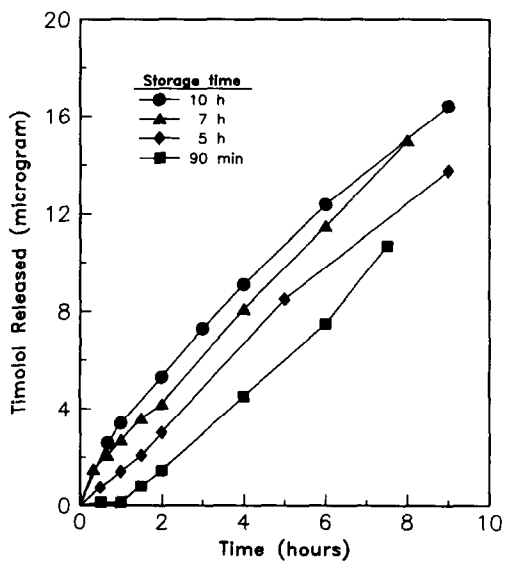

Fig. 1. Timolol release from silicone tubing devices containing 12.5 μl of 5 mg/ml timolol solution at pH 8.64. The devices were stored 90 min, 5 h, 7 h and 10 h before release study.

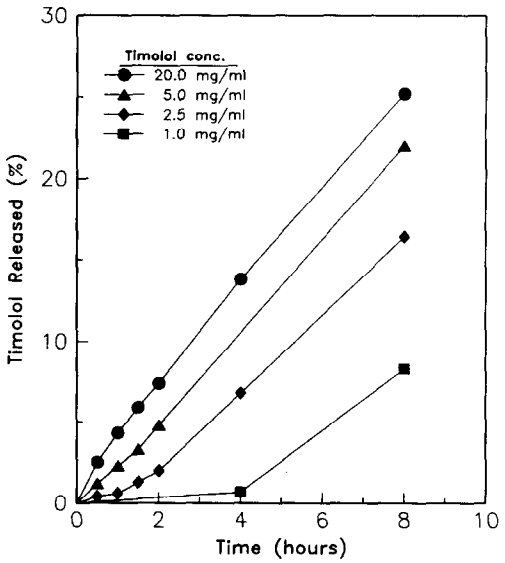

Fig. 2. Timolol release from silicone tubing devices containing 1.0, 2.5, 5.0 or 20.0 mg/ml of timolol in 0.15 M sodium borate at pH 8.64. Storage time for the devices was 5 h.

TABLE 1

Release characteristics of timolol from silicone tubing devices

V (μl)	t_s (h)	pH	C_i (mg/ml)	dM_t/dt		Burst (%)	P ($\times 10^9$) ($cm^2\ s^{-1}$)	n
				$\mu g/h$	%/h			
12.5	8	8.34	5.0	1.21 ± 0.04 [a]	2.02 ± 0.04	6.4 ± 0.4	2.48 ± 0.08	5
12.5	9	8.64	2.5	0.71 ± 0.02	2.32 ± 0.03	7.8 ± 1.4	2.91 ± 0.08	5
12.5	5	8.64	5.0	1.75 ± 0.04	2.82 ± 0.05	−2.0 ± 0.9	3.59 ± 0.08	14
6.0	3	8.64	20.0	2.69 ± 0.05	2.21 ± 0.07	1.6 ± 0.5	2.89 ± 0.05	5
9.0	3	8.64	20.0	4.40 ± 0.09	2.37 ± 0.05	1.1 ± 0.3	3.13 ± 0.06	6
12.5	3	8.64	20.0	7.21 ± 0.26	2.98 ± 0.18	2.9 ± 1.0	3.70 ± 0.13	7
12.5	4	8.84	5.0	2.23 ± 0.05	3.80 ± 0.10	2.6 ± 0.8	4.58 ± 0.10	5
12.5	3	9.24	5.0	2.94 ± 0.11	4.79 ± 0.13	6.9 ± 1.0	6.03 ± 0.23	5

[a] Means ± S.E. of n determinations.

V, volume of timolol solution in the device; t_s, ideal storage time; C_i, timolol concentration in the device; dM_t/dt, release rate; P, permeability (DK from Eqn. 1).

Without the initial lag or burst, timolol release from a cylindrical reservoir device should obey Eqn 1 (Baker and Lonsdale, 1974):

$$\frac{dM_t}{dt} = \frac{2\pi hDK\Delta C}{\ln(r_0/r_i)} \quad (1)$$

where dM_t/dt is the steady-state release rate at time t, r_0 and r_i are the outer and inner radii of the cylinder, respectively, h is the length of the cylinder, D is the diffusion coefficient of the drug in the membranes, K is the partition coefficient of drug between membrane and core of the device and ΔC is the difference between the internal (C_i) and external (C_e) drug concentrations. When $C_e < C_i$, the difference ΔC can be considered equal to the internal drug concentration. In all cases studied, $C_i < 250C_e$, and therefore ΔC was considered to be equal to C_i. Also, the permeability (P) of timolol in silicone membranes is defined as DK, the product of the diffusion (D) and partition (K) coefficients.

Zero-order drug release is achieved when the diffusable timolol concentration (ΔC) in the device remains constant (Eqn 1). When drug is in solution in the device core, as in these experiments, the diffusable concentration of timolol in the core of the device will decrease during drug release. When the fraction of timolol released during the experiment is small (< 20%), the drug is released at an approximately constant rate, i.e., the internal drug concentration or ΔC decreases only slightly. In that case, approximately zero-order drug release is obtained (Peterlin, 1983).

Release of timolol from the devices after the ideal storage time is presented in Table 1 and Figs. 3 and 4. Timolol is an amine with a pK_a of 9.2 (Huang et al., 1983) and consequently, as pH and therefore the fraction of nonionized timolol

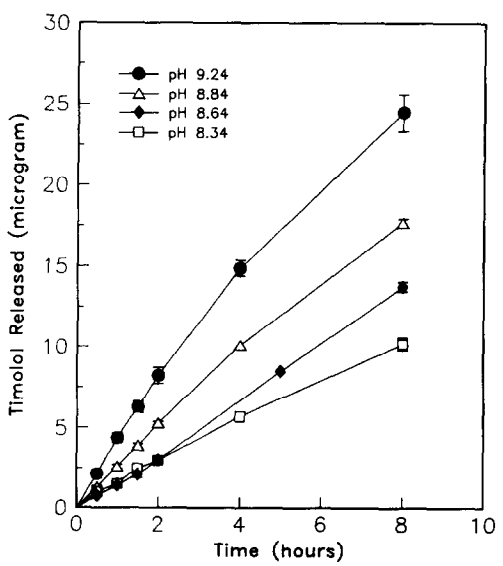

Fig. 3. Timolol release from silicone tubing devices containing 5 mg/ml of timolol at pH 8.34, 8.64, 8.84, and 9.24. The storage times of the devices prior to experiments are shown in Table 1. Means ± S.E. of 14 (pH 8.64) or 5 (pH 8.34, 8.84 and 9.24) experiments are presented.

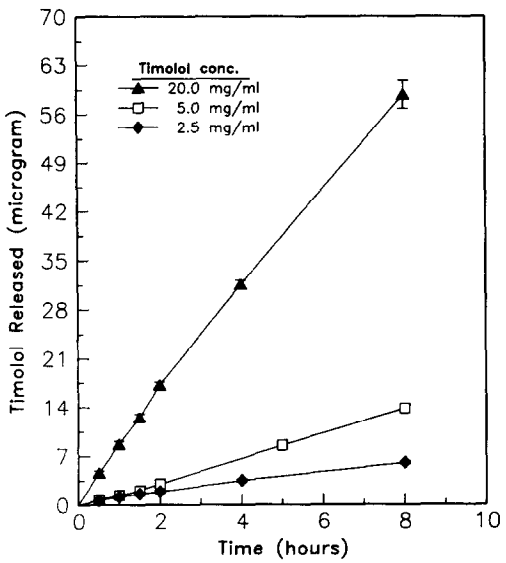

Fig. 4. Timolol release from silicone tubing devices with core pH 8.64 after ideal storage time (t_s; Table 1). Volume of timolol solution in the devices was 12.5 μl and concentrations were 2.5, 5.0 and 20.0 mg/ml. Means ± S.E. of 5 (2.5 mg/ml), 14 (5 mg/ml) and 7 (20.0 mg/ml) experiments are shown.

rise, the rate of timolol release through a hydrophobic silicone membrane increases (Table 1, Fig. 3). This is due to enhanced partitioning (Eqn 1) of timolol into the silicone membrane (Urtti et al., 1987), and as a consequence, an increase also in timolol permeability through the silicone membrane (Table 1). Increasing the permeability (P) accelerates both the absolute (μg/h) and fractional (%/h) release rates (Table 1). A higher fractional release rate causes deviations from a constant rate of drug release, because the concentration gradient of timolol across the silicone membranes (ΔC) decreases more rapidly. This is clearly observed as a more curved release profile as in the case when the pH in the core of the device was 9.24 (Fig. 3).

When the pH was adjusted to 8.64, the absolute rate of timolol release increased with timolol core concentration (C_i) and zero-order release kinetics were still obtained for 8 h (Fig. 4) with minimal burst effects (Table 1, Fig. 4). The rate of timolol release increased almost linearly when timolol concentration in the devices was changed from 2.5 to 20.0 mg/ml at pH 8.64 (Table 1, Fig. 5). When the injected volume of timolol solution in the devices was decreased from 12.5 μl to 9 or 6 μl, the fractional release rate, expressed as percent of timolol in the device released per hour, decreased slightly (less than 25%) leading to an apparent decrease in the calculated permeability coefficients (Table 1). This may be due to an increased proportion of timolol partitioning into the adhesive end-plugs when the diffusional surface area of the walls is decreased in the shorter devices.

When the injected volume of timolol solution in the devices was 12.5 μl, the release of timolol remained unchanged when the thickness of the end-plugs was changed from 0.5 to 3.5 mm. The release rates of timolol from the devices containing 12.5 μl of a solution (pH 8.64) of timolol (5 mg/ml) were 1.75 ± 0.04 μg/h ($n = 10$) and 1.71 ± 0.09 μg/h ($n = 4$) for 0.5 and 3.5 mm end-plugs, respectively. The corresponding fractions of burst/lag were −1.8 ± 1.2 and −2.7 ± 1.2%, respectively. Accordingly, it appears that timolol is released almost entirely through the side-walls and not through the end-plugs of the devices when the end-plugs are at least 0.5 mm thick. This is due to the small diffusional area of the ends (3.3 mm^2)

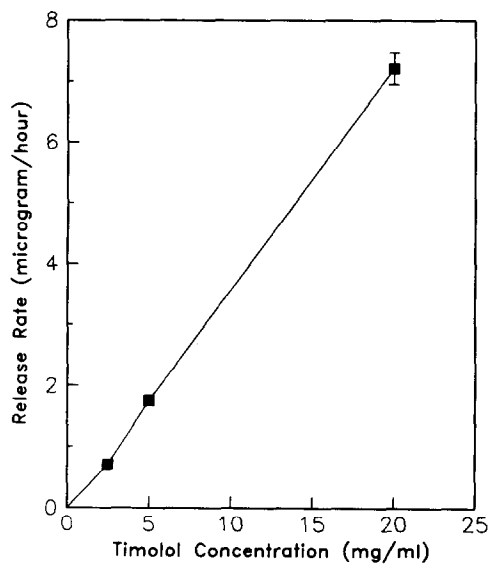

Fig. 5. Effect of timolol concentration in the silicone tubing devices on release rate of timolol from the devices at pH 8.64 after ideal storage time (Table 1). Means ± S.E. of n (see Table 1) experiments are shown.

compared with the side-walls (16.5–34.3 mm^2). The diffusional pathway through the walls (0.24 mm) is also shorter than via the ends (0.5–3.5 mm). In addition, timolol partitions more favorably from the core solution into the walls than into the adhesive end-plugs (Urtti et al., 1987).

Silicone controlled drug delivery devices of the type described are inexpensive and convenient to prepare and may prove to be versatile tools for in vivo evaluation of drug input rate vs drug distribution/response relationships. With these devices both the rate and pattern of timolol release can be easily modified. The release patterns can be modified by changing the pH of the device core (Fig. 3) or the storage time of the devices prior to use (Fig. 1). The release rate is modified by changing the concentration (Fig. 4) and pH (Fig. 3) of the core solution in the devices. Encouraging results were obtained in rabbits (Urrti et al., 1990) using these timolol ocular delivery devices. Devices of this type may also prove suitable for many other drugs.

Acknowledgement

This work was supported by INTERx Research Corporation/Merck, Sharp & Dohme Research Laboratories.

References

Baker, R.W. and Lonsdale, H.K., Controlled release: mechanisms and rates. In Tanquary, A.C. and Lacey, R.E. (Eds), *Controlled Release of Biologically Active agents,* Plenum, New York, 1974, pp. 15–71.

Baustian, C. and Mikkelson, T.J., Steady-state distribution of β-blockers in rabbit eye tissues following topical zero-order input. APhA/APS *39th Natl. Meet.,* 15 (1985) 173.

Chrai, S.S. and Robinson, J.R., Ocular evaluation of methylcellulose vehicle in albino rabbits. *J. Pharm. Sci.,* 63 (1974) 1218–1223.

Flynn, G.L., Yalkowsky, S.H. and Roseman, T.J., Mass transport phenomena and models: theoretical concepts. *J. Pharm. Sci.,* 63 (1974) 479–510.

Grass, G.M. and Robinson, J.R., Relationship of chemical structure to corneal penetration and influence of low-viscosity solution on ocular bioavailability. *J. Pharm. Sci.,* 73 (1984) 1021–1027.

Havener, W.H., *Ocular Pharmacology,* C.V. Mosby, Saint Louis, 1978.

Higuchi, W.I. and Higuchi, T., Theoretical analysis of diffusional movement through heterogenous barriers. *J. Am. Pharm. Assoc. Sci. Ed.,* 49 (1960) 598–606.

Huang, H.S., Schoenwald, R.D., and Lach, J.L., Corneal penetration behavior of beta-blocking agents, II. Assessments of barrier contributions. *J. Pharm. Sci.,* 72 (1983) 1272–1279.

Karlin, K.M., Zimmerman, T.J. and Nardin, G., Beta blockers in ophthalmology. *J. Toxicol. Cutan. Ocular. Toxicol.* 1 (1982) 155–168.

Lee, V.H.L. and Robinson, J.R., Mechanistic and quantitative evaluation of precorneal pilocarpine disposition in albino rabbits. *J. Pharm. Sci.,* 68 (1979) 673–684.

Maurice, E.M. and Mishima, S., Ocular pharmacokinetics. In Sears, M.L. (Ed.), *Handbook of Experimental Pharmacology,* vol. 69, Springer, Berlin, 1984, pp. 19–116.

Most, C.F. Jr, Some filler effects on diffusion in silicone rubber. *J. Appl. Polym. Sci.,* 14 (1970) 1019–1024.

Peterlin, A., Transport of small molecules in polymers. In Bruck, S.D. (Ed.), *Controlled Drug Delivery,* CRC Press, Boca Raton, FL 1983, pp. 16–51.

Schmitt, C.J., Lotti, V.J. and LeDouarec, J.C., Penetration of timolol into the rabbit eye. *Arch. Ophthalmol.,* 98 (1980) 547–551.

Shell, J.W., Ophthalmic drug delivery systems. *Surv. Ophthalmol.,* 29 (1984) 117–128.

Urtti, A., Pipkin, J.D., Rork, G. and Repta, A.J., Experimental surrogate devices for ocular biopharmaceutical studies: in vitro studies. *Proc. Int. Symp. Control. Rel. Bioact. Mater.,* 14 (1987) 295–296.

Urtti, A., Pipkin, J.D., Rork, G. and Repta, A.J., Controlled drug delivery devices for experimental ocular studies with timolol. 2. Tissue distribution and systemic absorption in the pigmented rabbit. *Int. J. Pharm.,* 61 (1990) 241–249.

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