*International Journal of Pharmaceutics*, 84 (1992) 217–222



IJP 02793

# Pharmacokinetic simulation reveals in vivo deviations from in vitro release of timolol from polymer matrices

Ulla Finne and Arto Urtti

*Department of Pharmaceutical Technology, University of Kuopio, P.O. Box 1627, SF-70211 Kuopio (Finland)*

(Received 2 September 1991)
(Accepted 30 January 1992)

*Key words*: Timolol; Systemic absorption; STELLA® simulation; Monoisopropyl PVM-MA; Sodium acetate

## Summary

In some glaucoma patients timolol eye drops may cause severe cardiovascular and respiratory side-effects due to systemic absorption. Administration of timolol in controlled release devices reduces the harmful systemic absorption of the drug, however, as in the case of many drug delivery systems, the in vivo release rate relative to the in vitro rate is not known. In the present investigation, the concentrations of timolol in lacrimal fluid and plasma after administration into rabbit eyes in different matrices of monoisopropyl ester of poly(vinyl methyl ether-maleic anhydride) (PVM-MA) was analysed using STELLA® simulation software. In the model, in vitro release rates of timolol were used as input values and preocular and systemic kinetic parameters from literature were included. The simulation model calculated predicted drug concentration profiles in tear fluid and plasma. The simulations showed that drug release in vivo in tear fluid was decreased compared to the in vitro experiments in the case of the unbuffered and phosphate buffered matrices. Differences in the conjunctival permeability of timolol after administration of various matrix types were revealed by the simulations where measured concentrations in tear fluid were used as input values. The phosphate buffered matrix showed similar tear fluid - plasma transfer of timolol in simulations and experimentally. Sodium acetate buffer in the matrix reduced the permeability of timolol in the conjunctival epithelium, thus decreasing the systemic absorption of the drug. In the case of the unbuffered matrix, probably a decrease in the pH of tear fluid close to the matrix may reduce the transport of timolol through the conjunctiva. STELLA® simulations proved useful in the in vitro - in vivo comparisons of ophthalmic timolol.

## Introduction

Many topically applied ophthalmic drugs like timolol (Kaila et al., 1986), betaxolol (Polansky and Alvarado, 1985), adrenaline and dipivefrin (Anderson, 1980) as well as phenylephrine (Kumar et al., 1986) have been shown to absorb to the systemic circulation after application to the eye. Several of these drugs may exert systemic side-effects that limit the therapeutic utility of the compounds in some patients. Antiglaucoma drugs, e.g., timolol, may cause severe cardiovascular and respiratory side-effects (Nelson et al., 1986). The systemic absorption of ophthalmic timolol is decreased by administering the drug in polymeric inserts (Finne et al., 1990, 1991) or in

*Correspondence to:* A. Urtti, University of Kuopio, Department of Pharmaceutical Technology, P.O. Box 1627, SF-70211 Kuopio, Finland.

an experimental controlled release device (Urtti et al., 1990) instead of eye drops.

Computer simulations are useful in investigating and predicting the behaviour of delivery systems in vivo, assuming adequate background information about the pharmacokinetics of the drug is available. STELLA® is a mathematical modelling program that can be used to simulate, among other things, the time-dependent behaviour of pharmacokinetic and pharmacodynamic systems (Bogan, 1989; Washington et al., 1990). The program uses the input values and calculates the resultant drug concentrations in every compartment in the model for each time interval. Pharmacokinetic modelling with STELLA® has been used to predict the behaviour of controlled release formulations (Grass and Morehead, 1989; Wilson et al., 1991) and devices (Urtti, 1991). A STELLA® model has also been used to predict timolol concentrations in aqueous humor and plasma after topical eye drop administration (Grass and Lee, 1990).

In the present investigation, a kinetic model for ocular administration of timolol in inserts was developed. In vitro release was used as an input and the predicted values were compared with the previously published timolol concentrations in vivo in rabbit tear fluid and plasma (Finne et al., 1990, 1991). In another set of simulations, measured timolol concentration profiles in tear fluid were used as input values to study drug transport from tear fluid to plasma (Finne et al., 1990, 1991).

## Materials and Methods

*Preparation of polymer matrices*

The monoisopropyl ester of poly(vinyl methyl ether-maleic anhydride) (PVM-MA) was used as a 50% isopropanol solution of the polymer (Gantrez® ES-335, GAF Europe, Esher, U.K.) to prepare the matrices. Buffered polymer films containing 12.9% disodium phosphate (2.11 mmol) or 6.4% sodium acetate (2.11 mmol) and unbuffered polymer films were prepared by solvent casting from organic solvents as described previously (Finne et al., 1989). Circular matrices

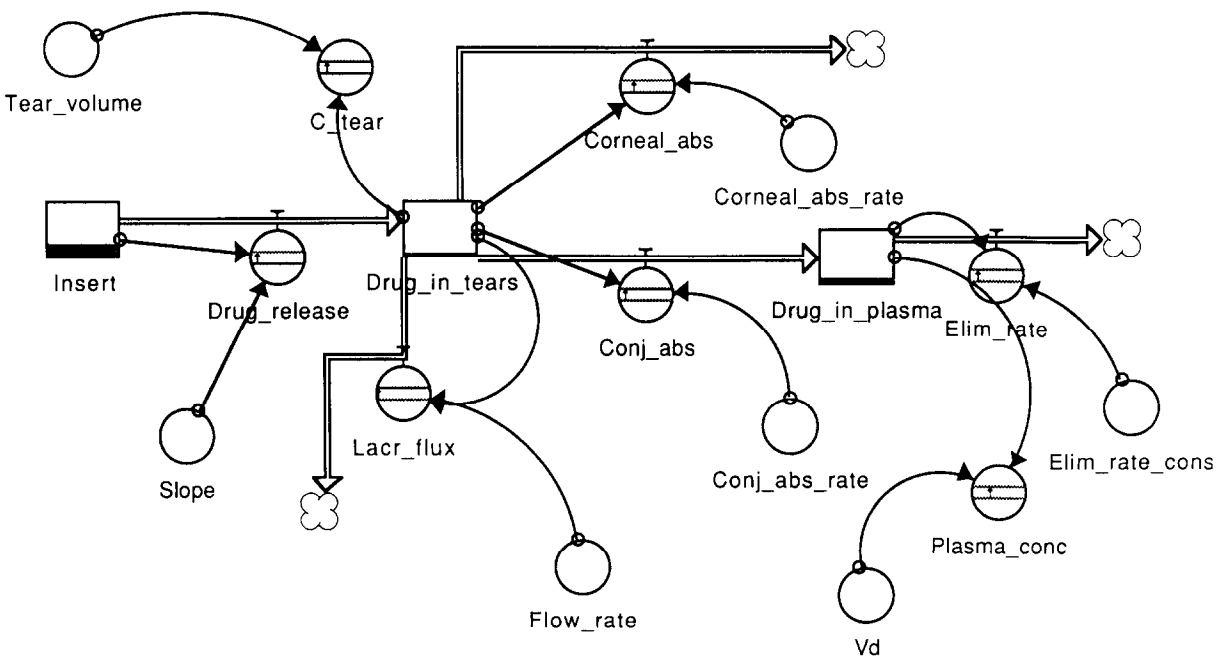


Fig. 1. Diagram of the STELLA® model used for simulation of the pharmacokinetics of ophthalmic timolol.

with diameter 5 mm were cut with a cork bore from the film. Each matrix contained 125 $\mu$g of timolol maleate closely corresponding to 25 $\mu$l of the commercially available 0.5% timolol solution. The polymer matrices were similar to those used in the in vivo studies.

*Drug release in vitro*

Timolol release from the unbuffered monoisopropyl PVM-MA matrices and from those containing disodium phosphate or sodium acetate as a basic additive was studied in diffusion chambers made of plexi glass (Schoenwald and Huang, 1983). The matrix was attached with molten blockform paraffin to a microscope slide coated with Teflon to allow drug release only from one side of the polymer matrix. Microscope slides were then mounted between the two compartments of the diffusion cell. The dissolution medium was 6.5 ml of 2 mM phosphate buffer pH 7.4 ($\mu$ = 0.5 with NaCl) at 32°C. Mixing was provided by nitrogen bubbles (3–4/s) as described earlier (Schoenwald and Huang, 1983). Samples (1.0 ml) were withdrawn at various times and replaced by fresh dissolution medium. Timolol concentrations were determined with a UV spectrophotometer at 294 nm.

*Computer simulations*

A model of timolol kinetics (Fig. 1) in tear fluid and plasma was constructed using the STELLA® program (High Performance Systems, Inc., Lyme, NH, U.S.A.) on a Macintosh Plus computer (Apple Computer, Inc., Cupertino, CA, U.S.A.). In vitro release rates of timolol from the three matrix types were used as input values in these simulations. The parameters and equations used in the simulations are listed in Table 1. We have shown previously (Urtti et al., 1990) that systemic absorption of timolol after device administration takes place mainly via the conjunctiva, because timolol clearance via normal tear fluid is negligible compared to conjunctival clearance. Thus, in contrast to the case of eye drops, nasal mucosa was considered to be of minor importance in the systemic absorption of timolol from the matrices and, accordingly, nasal absorption was omitted from the model. Simulations were conducted using a fourth-order Runge-Kutta algorithm with a d$t$ value of 0.015 h. The predicted

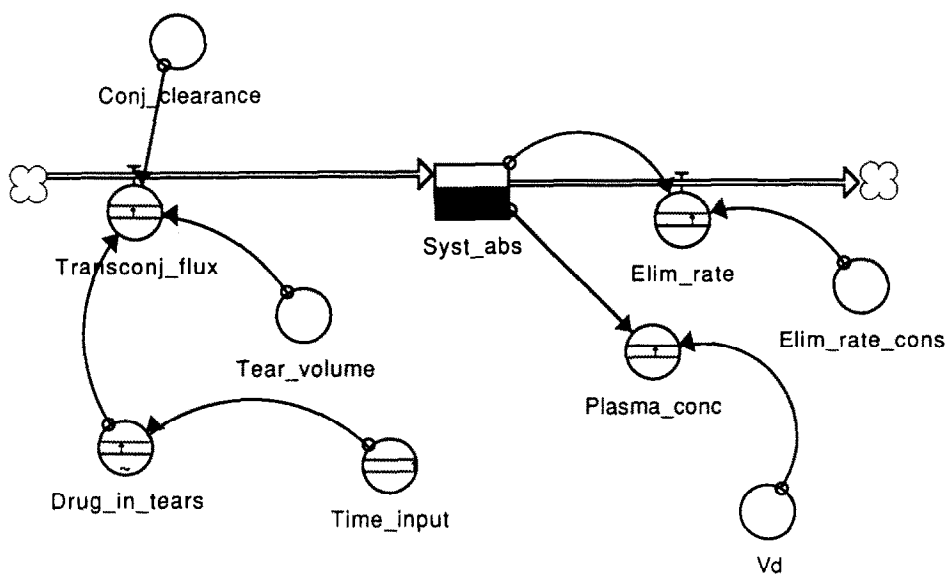


Fig. 2. Diagram of the STELLA® model used for simulation of the systemic concentrations of timolol on the basis of the in vivo tear fluid data.

TABLE 1

*Equations and parameters used in the in vitro-in vivo simulation (for references see Materials and Methods)*

| |
|---|
| Drug in tears = Drug in tears + d$t$ · (Drug release − Conj. abs − Corneal abs − Lacr. flux) |
| INIT (Drug in tears) = 0 |
| Insert = Insert + d$t$ · ( − Drug release) |
| INIT (Insert) = 125 $\mu$g |
| Drug in plasma = Drug in plasma + d$t$ · (Conj. abs − Elim. rate) |
| INIT (Drug in plasma) = 0 |
| Conj. abs. − Drug in tears · Conj. abs. rate |
| Conj. abs. rate = 89 $h^{-1}$ |
| Corneal abs. = Drug in tears · Corneal abs. rate |
| Corneal abs. rate = 11.6 $h^{-1}$ |
| $C_{tear}$ = Drug in tears/Tear volume |
| Lacr. flux = Drug in tears · Flow rate |
| Drug release = Insert · Slope |
| Elim. rate = Elim. rate const. · Drug in plasma |
| Elim. rate const. = 1.6 $h^{-1}$ |
| Flow rate = 4.2 $h^{-1}$ |
| Plasma conc. = 1000 · 2 · Drug in plasma/ $V_d$ |
| Slope = [a,b,c] |
| Tear volume = 0.007 ml |
| $V_d$ = 8800 ml |

[a] 0.28 $\mu$g/h for the unbuffered matrix.
[b] 0.37 $\mu$g/h for the acetate buffered matrix.
[c] 1.08 $\mu$g/h for the phosphate buffered matrix.

values were compared with the previously reported timolol concentrations in lacrimal fluid and plasma (Finne et al., 1990, 1991).

TABLE 2

*Equations and parameters used in the tear fluid - plasma simulation (for references see Materials and Methods)*

| |
|---|
| Syst. abs. = Syst. abs. + d$t$ · (Transconj. flux − Elim. rate) |
| INIT (Syst. abs.) = 0 |
| Conj. clearance = 0.62 ml/h |
| Transconj. flux = (Drug in tears/Tear volume) · Conj. clearance |
| Elim. rate = Elim. rate const. · Syst. abs. |
| Elim. rate const. = 1.6 $h^{-1}$ |
| Plasma conc. = 1000 · 2 · Syst. abs./ $V_d$ |
| Tear volume = 0.007 ml |
| Time input = Time |
| $V_d$ = 8800 ml |
| Drug in tears = graph (Time input) |

In another model (Fig. 2), measured timolol concentrations in tear fluid were used as a graphical drug input and the predicted values were compared with the measured timolol concentrations in plasma (Finne et al., 1990, 1991). Table 2 shows the parameters and equations used in these simulations that were conducted as above with a d$t$ value of 0.05 h.

*Pharmacokinetic parameters used in the simulations*

The following pharmacokinetic parameters were used in simulations: conjunctival absorption

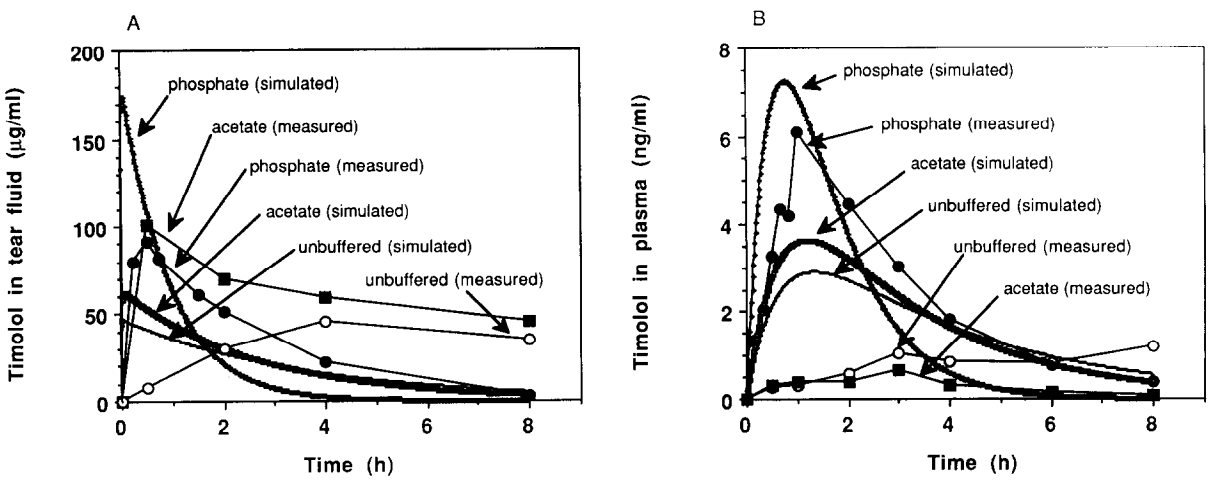


Fig. 3. Comparison of the measured and simulated timolol concentrations (A) in tear fluid ($\mu$g/ml) and (B) in plasma (ng/ml) using the STELLA® model shown in Fig. 1. In vitro release rates of timolol from the unbuffered, acetate and phosphate buffered matrices of monoisopropyl PVM-MA were used as input values.

rate $k_{cj} = 89\ h^{-1}$ was calculated from conjunctival clearance $Cl_{cj} = 10.38\ \mu l/min$ (Ahmed et al., 1987) and tear fluid volume $V_{dtf} = 7\ \mu l$ (Maurice and Mishima, 1984) by $k_{cj} = Cl_{cj}/V_{dtf}$; corneal absorption rate $k_c = 11.6\ h^{-1}$ was calculated similarly with $Cl_c = 1.35\ \mu l/min$ (Urtti et al., 1990) and $V_{dtf} = 7\ \mu l$ (Maurice and Mishima, 1984); tear fluid flow rate $K_{tf}$ was $4.2\ h^{-1}$ (Maurice and Mishima, 1984). Elimination rate of timolol from plasma $k_{el}$ and volume of distribution $V_d$ were calculated in the following way from data from Järvinen et al. (1991): mean residence time of timolol in rabbit plasma after 125 $\mu$g i.v. injection $MRT_{iv} = AUMC/AUC = 37.6$ min; $t_{1/2} = \ln 2 \times MRT = 0.43$ h; $k_{el} = \ln 2/t_{1/2} = 1.6\ h^{-1}$; plasma clearance of timolol $Cl_{pla} = dose_{iv}/AUC = 125\ \mu g/8.92\ h \times ng/ml = 14.0$ l/h; distribution volume $V_d = Cl_{pla}/k_{el} = 8.8$ l.

The release rate of timolol from the matrices was evaluated from the slope of $\ln Q/t$, where $Q$ is the amount of timolol remaining in the matrix at time $t$. The slope (a mean of four release experiments) was 0.28, 0.37, or 1.08 $\mu$g/h for the unbuffered, acetate, and phosphate buffered matrix, respectively. In model II (Fig. 2), the input value of timolol in tears ($\mu$g) is given in graphical form. The values were obtained directly from experimental data (Finne et al., 1990, 1991).

## Results and Conclusions

The simulated and measured timolol concentrations in tear fluid and plasma are shown in Fig. 3A and B, respectively. Simulations using the model in Fig. 1 show that the timolol concentrations in tear fluid and plasma cannot be predicted from the in vitro release experiments. Simulation revealed that drug release in vivo in tear fluid was decreased compared to the in vitro release (Fig. 3A). Comparison of the measured and simulated lacrimal timolol concentrations after the acetate buffered matrices (real concentrations larger than simulated, Fig. 3A) was complicated by the possible acetate induced change in $Cl_{cj}$. Only in the case of the phosphate buffered matrix were the predicted and measured timolol concentrations in plasma close to one another (Fig. 3B). In vivo,

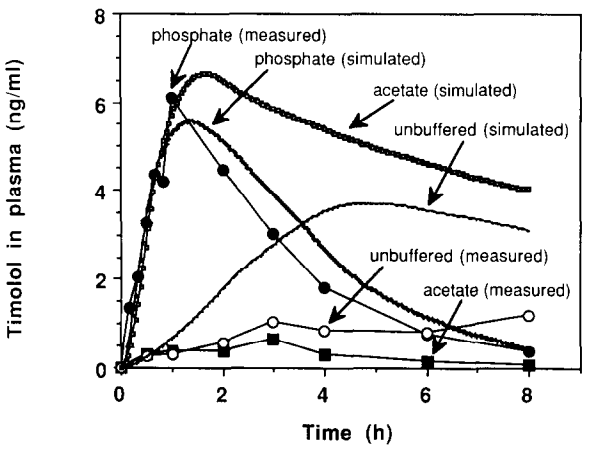


Fig. 4. Comparison of the measured and simulated timolol concentrations in plasma (ng/ml) after administration of unbuffered, acetate and phosphate buffered matrices of monoisopropyl PVM-MA using the STELLA® model shown in Fig. 2. Measured timolol concentrations in tear fluid were used as input values.

timolol release from the unbuffered and phosphate buffered matrices was retarded possibly due to the poor mixing in lacrimal fluid. The conjunctival clearance of timolol may be reduced when administered in the acetate buffered matrix. This might explain the increased lacrimal and decreased systemic timolol concentrations in this case (Fig. 3A and B).

When measured timolol concentrations in tear fluid are used as input values in the simulations according to the model in Fig. 2 a good correlation between the measured and simulated concentrations in plasma (Fig. 4) can be seen in the case of the phosphate buffered matrix, indicating that the conjunctival clearance is close to the in vitro value used in the simulation. Timolol concentrations in plasma from the acetate buffered matrix, on the other hand, are substantially below the predicted values. After administration in acetate buffered matrix, the measured timolol concentrations in plasma are only 1/10 of the predicted values (Fig. 4), which is at least partly explained by the reduction of the conjunctival clearance due to acetate ions. According to our preliminary in vitro studies with rabbit conjunctiva, addition of acetate to constant pH medium

reduces the permeability of timolol in conjunctiva via an unknown mechanism.

The simulations of tear fluid data using the model in Fig. 2 reveal that the predicted timolol concentrations in plasma are also several fold greater than the actual values in the case of the unbuffered matrix (Fig. 4). The difference can be explained on the basis that, during timolol release from the unbuffered monoisopropyl PVM-MA matrix, the pH in the lacrimal fluid falls due to accumulation of hydrogen ions that are released from the dissolving polymer matrix. For example, pH on the polymer surface is expected to decrease to 5.7 in 2 mM phosphate buffer in vitro (Finne et al., 1991). In vivo, the hydrogen ions are not efficiently neutralized owing to the poor buffering capacity of tear fluid. Thus, a possible decrease of pH close to the matrix may increase ionization of timolol and, accordingly, reduce its transport through the conjunctiva.

In conclusion, due to changes in drug release and conjunctival permeability, timolol concentrations in plasma can be predicted from the in vitro studies only in the case of the phosphate buffered matrices. STELLA® simulation is useful when in vivo drug release properties from different controlled release devices are compared.

## Acknowledgement

This work was supported by a grant from Academy of Finland.

## References

Ahmed, I., Gokhale, R.J., Shah, M.V. and Patton, T.F., Physicochemical determinants of drug diffusion across the conjunctiva, sclera and cornea. *J. Pharm. Sci.*, 76 (1987) 583–586.

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

Bogan, D.K., Simulation software for the Macintosh. *Science*, 246 (1989) 138–142.

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. Controlled Release*, 10 (1989) 189–194.

Finne, U., Salivirta, J. and Urtti, A., Sodium acetate improves the ocular/systemic absorption ratio of timolol applied ocularly in monoisopropyl PVM-MA matrices. *Int. J. Pharm.*, 75 (1991) R1–R4.

Finne, U., Väisänen, V. and Urtti, A., Modification of ocular and systemic absorption of timolol from ocular inserts by a buffering agent and a vasoconstrictor. *Int. J. Pharm.*, 65 (1990) 19–27.

Grass, G.M. and Lee, V.H.L., A STELLA® model to predict aqueous humor and plasma drug concentrations from topical administration to the eye. *Proc. Int. Symp. Controlled Release Bioact. Mater.*, 17 (1990) 299–300.

Grass, M. and Morehead, W.T., Evidence for site-specific absorption of a novel ACE inhibitor. *Pharm. Res.*, 9 (1989) 759–765.

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.

Järvinen, K., Vartiainen, E. and Urtti, A. Optimizing the systemic and ocular absorption of timolol from eyedrops. *STP Pharm. Sci.*, 2 (1992) 105–110.

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

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.

Schoenwald, R.D. and Huang, H.-S., Corneal penetration behavior of β-blocking agents I: physicochemical factors. *J. Pharm. Sci.*, 72 (1983) 1266–1272.

Urtti, A., Simulation of the absorptive membrane permeability effects on in vivo drug release from controlled release systems. *Proc. Int. Symp. Controlled Release Bioact. Mater.*, 18 (1991) 431–432.

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

Washington, C., Washington, N. and Wilson, C., *Pharmacokinetic Modelling using STELLA on the Apple Macintosh*, 1990, Ellis Horwood, Chichester.

Wilson, C.G., Washington, N., Greaves, J.L., Washington, C., Wilding, I.R., Hoadley, T. and Sims, E.E., Predictive modelling of the behaviour of a controlled release buflomedil HCl formulation using scintigraphic and pharmacokinetic data. *Int. J. Pharm.*, 72 (1991) 79–86.