Ocular Bioavailability and Tissue Distribution of [^{14}C]Ketorolac Tromethamine in Rabbits

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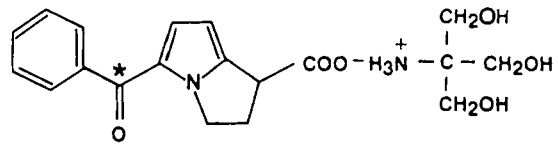
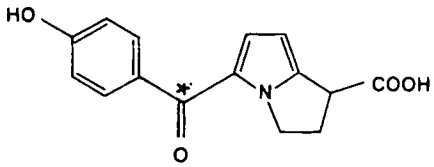
Abstract □ The ocular bioavailability of 0.5% [^{14}C]ketorolac tromethamine administered topically (50 μL) to the eye was determined. The ocular bioavailability of ketorolac was 4% in anesthetized rabbits and was determined by comparing drug concentrations in the aqueous humor after topical application with those obtained after intracameral injection of an equivalent dose of 0.25 mg of ketorolac tromethamine per eye. Although ketorolac administered to the eye was completely absorbed systemically, concentrations of ketorolac (AUC) were, on the average, 13 times higher in the aqueous humor than in plasma after topical administration. In a separate ocular distribution study, peak concentrations of radioactivity were achieved in the ocular tissues and in plasma within 1 h post instillation. Concentrations of total radioactivity were highest in the cornea and sclera and lowest in the lens.

Ketorolac tromethamine (1) is a nonsteroidal anti-inflammatory agent with potent analgesic and anti-inflammatory activity.[1] When applied topically to the eye, 1 significantly inhibited the inflammatory response to silver nitrate-induced cauterization of the corneas of rat eyes at concentrations of 0.25 and 0.5%.[2] Ketorolac tromethamine was also shown to be nonirritating when applied topically to the eyes of dogs, rhesus monkeys, and rats at concentrations up to 0.5%.[2]

The ocular and systemic bioavailabilities of 1 were evaluated after ophthalmic administration in rabbits. Drug concentrations in the aqueous humor were compared after ocular versus intracameral injection in the determination of ocular bioavailability. The systemic bioavailability was determined by evaluating plasma concentrations of drug after ocular versus intravenous administration. The ocular bioavailability of ophthalmic flurbiprofen, determined in a similar manner, was shown to average 7–10%, and the systemic bioavailability averaged 74%.[3]



Ketorolac Tromethamine **(1)**



p-Hydroxy Ketorolac **(2)**

*indicates position of ^{14}C label

Distribution of drug-related radioactivity in various ocular tissues after a single instillation of 1 in the eyes of rabbits was determined to be similar to those described for pilocarpine,[4–6] timolol,[7–9] and clonidine.[10]

Experimental Section

Materials—[^{14}C]Ketorolac (lot no. 8891-GH-44) was synthesized by Syntex Research, Palo Alto, CA. The following reagents were used in dose assays and sample processing and analyses: Biofluor scintillation cocktail (New England Nuclear, Boston, MA); methanol, methylene chloride, and acetonitrile were distilled in glass (Burdick and Jackson Laboratories, Muskegon, MI); ethyl acetate and hexane (J. T. Baker Chemical Co., Phillipsburg, NJ); glacial acetic acid, ACS grade, and sodium citrate, AR grade (Mallinckrodt Inc., Paris, KY); citric acid (Syntex Inc., Palo Alto, CA); potassium hydroxide, N.F. grade (Mallinckrodt); hydrochloric acid, ACS grade (Mallinckrodt); and TLC plates (Silica Gel-GF plates; Analtech Inc., Newark, DE).

Apparatus—A Packard Tri-carb liquid scintillation spectrometer (model 3330) was used for liquid scintillation counting. The TLC plates were scanned for radioactivity using a Berthold TLC-scanner (model LB-2760). A Perkin-Elmer UV visible spectrophotometer (model 559) was used for the dose assay. The following apparatus was used for the HPLC assay: a Varian HPLC (model 5060), a C-18 reversed phase column (Altex-Ultrasphere 5-μm particle size, 250 mm × 4.6 mm I.D., Berkeley, CA), a UV detector (model UV-100, Varian), an HPLC radioactivity detector (model HP Flo-One, Radiomatics), a fraction collector (model FRAC-100, Pharmacia), and a recorder (Kipp-zonen) with a 1-mV full scale response and a chart speed of 0.5 cm/min.

Dose Assay—The dose solutions were analyzed for [^{14}C]ketorolac tromethamine content by mixing with Biofluor scintillation cocktail, followed by scintillation counting. The stability of the radiolabeled solution was determined by TLC using methylene chloride:methanol:acetic acid (95:5:0.1) as the developing solvent system. The TLC plates were subsequently scanned for radioactivity. The total ketorolac tromethamine (1) content was determined by UV spectroscopy.

Sample Analyses—Samples of aqueous humor (12–15 μL) or plasma (150 μL) were mixed with 15 mL of Biofluor and analyzed for total radioactivity. Dissected eye tissues were accurately weighed and then hydrolyzed with 3 N KOH. Aliquots (200 μL) of these digests were neutralized with 3 N HCl prior to the addition of 15 mL of Biofluor scintillation cocktail and liquid scintillation counting. All samples were corrected for quenching by automatic external standardization. As soon as possible after sample collection, plasma was analyzed for intact 1. One milliliter of plasma was mixed with an equal volume of 0.1 M citrate buffer (pH 3.0) and the pH was adjusted to 3 with 1 N HCl. The sample was extracted twice with 5 mL of an ethyl acetate:hexane mixture (30:70). The combined extracts were evaporated under nitrogen and the residue was reconstituted in 1 mL of methanol. Two 0.1-mL aliquots of the extract and the entire aqueous (plasma) residue were each mixed with 15 mL of Biofluor for analysis of total radioactivity. The remaining extract was evaporated under nitrogen and stored at −15 to −20 °C until the time of HPLC analysis.

For the HPLC assay, the sample extract was reconstituted in 250 μL of water:acetonitrile (70:30), and 80 μL were injected onto the HPLC column. The flow rate of the mobile phase, which consisted of 0.2% acetic acid:acetonitrile (60:40), was 1.0 mL/min. The absor-

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bance of the column effluent was monitored at 320 nm (0.005 absorbance unit per mV). Detection and quantitation were accomplished with either an HPLC radioactivity detector or a fraction collector followed by liquid scintillation counting. In the case of fractionation, fifteen 0.8-min fractions were collected for each sample. The retention times of 1 and the *p*-hydroxy metabolite (2) were ~8 and 4 min, respectively. Plasma concentrations of ketorolac were estimated as follows:

$$C_p = \text{Total Radioactivity} \times f_{ext} \times f_{HPLC} \quad (1)$$

where f_{ext} is the fraction of radioactivity extractable into the organic solvent and f_{HPLC} is the fraction of extractable radioactivity eluting with the same retention time as an authentic ketorolac reference standard on HPLC. The extraction efficiency of ketorolac in plasma samples spiked with the drug was ~97%, and the drug was not degraded by this procedure.

Plasma extracts from some samples which had poor resolution due to a contaminating peak were re-assayed for 1 by silica gel TLC, using the solvent system benzene:tetrahydrofuran:acetic acid (9:1:1). Using this solvent system, the R_f values of 1 and 2 were 0.51 and 0.34, respectively. The TLC plates were scraped into 19 fractions. One milliliter of methanol, followed by 15 mL of Biofluor, was added to each fraction and the fractions were counted for radioactivity.

Bioavailability Studies in Rabbits—Fifteen female New Zealand white rabbits (2.9–4.2 kg) were used. Three rabbits were used for the iv administration and 12 rabbits were used for the ocular studies. The rabbits were drug free for at least two weeks prior to the study. The rabbits were also fasted for 12 h prior to dosing. Food (Purina Rabbit Chow) was withheld until 4 h after the dose; water was allowed ad libitum.

The intravenous dose solution [1 mL of phosphate buffer solution containing 1 mg (20 μCi) of [^{14}C]ketorolac tromethamine, pH 8] was delivered via the marginal ear vein in each of three rabbits. Six rabbits each received 50 μL of 0.5% [^{14}C]ketorolac tromethamine ophthalmic solution (containing benzalkonium chloride) per eye (0.5 mg, 9–10 μCi per rabbit) by topical instillation. An additional six rabbits each received 20 μL of 1.27% [^{14}C]ketorolac tromethamine aqueous solution per eye (0.5 mg, 3.4 μCi per rabbit) by intracameral injection.

In the ocular studies, the rabbits were anesthetized prior to collection of aqueous humor and during topical instillations or intracameral injections. The anesthetic cocktail consisted of ketamine (50 mg/mL), xylazine (5 mg/mL), and acepromazine (1 mg/mL) in saline, and was injected im at a dose rate of 0.85 mL/kg. Topical doses were delivered via microliter syringe in a dropwise manner onto the eye. The intracameral dose was injected directly into the anterior chamber via puncture at the limbus using a ½-inch, 28-gauge syringe (#2 point, Hamilton Co.).

Blood (2–3 mL) was sampled from the marginal ear vein of each rabbit at 5, 15, and 30 min and at 1, 2, 3, 5, 7, 10, and 24 h after the iv dose. After ocular doses, blood sampling times were at 5 (intracameral only), 15, and 30 min, and 1, 2, 4, 6, and 8 h. After ocular doses, aqueous humor (10–20 μL) was sampled from each eye of the rabbit at 0.5, 1, 2, 4, 6, and 8 h using a 28-gauge needle and a Hamilton syringe. In the ocular studies, the rabbits were kept anesthetized throughout the study and were sacrificed by intravenous injection of T-61 Euthanasia solution (Taylor Pharmacal Co., Decatur, IL) before recovery from anesthesia.

Tissue Distribution Study in Rabbits—Twenty-four female New Zealand white rabbits (2.3–3.0 kg) were used in the ocular and intravenous studies. Rabbits were fasted in a similar manner to those used in the bioavailability studies. Twenty-one rabbits each received a topical ocular and intravenous dose of 50 μL per eye of 0.5% [^{14}C]ketorolac tromethamine ophthalmic solution. Following application of the dose, animals were sacrificed in groups of three at 0.5, 1, 2, 4, 6, 8, and 24 h. Shortly prior to sacrifice, animals were anesthetized with the anesthetic cocktail and aqueous humor was sampled via puncture at the limbus. At sacrifice, a blood sample was obtained from each rabbit by cardiac puncture. After sacrifice, both eyes were enucleated and thoroughly rinsed with saline to remove any residual dose solution. Each eye was further dissected in order to collect the cornea, lens, vitreous humor, iris-ciliary body, retina-choroid, and sclera.

Following the intravenous administration of [^{14}C]ketorolac tromethamine (1 mg, 20 μCi) in three rabbits, the rabbits were sacrificed at 0.5, 1, or 5 h. The following tissues were collected from both eyes of each rabbit: cornea, iris-ciliary body, lens, retina-choroid, aqueous humor, and vitreous humor. Baseline plasma and tissue samples were obtained from a nondosed rabbit.

Results and Discussion

Dose Assay—Dose assays performed for the intravenous and ocular doses indicated values between 98 and 102% of target concentrations of 1. The R_f of [^{14}C]ketorolac tromethamine on TLC plates averaged 0.28, and the purity of the ^{14}C label was >95%. The specific activity of [^{14}C]ketorolac tromethamine averaged 2.9×10^{-2} μCi/μg in the topical and intravenous studies and 9.8×10^{-3} μCi/μg in the intracameral study.

Ocular Bioavailability—Drug absorption into the eye was determined using the anterior chamber as the sampling compartment, assuming that noncorneal absorption does not contribute significantly to intraocular drug levels after topical dosing.[11] A previous study indicated that no metabolites of 1 were formed in the aqueous humor after topical administration.[12] Hence, concentrations of total radioactivity in aqueous humor after ophthalmic administration of [^{14}C]ketorolac tromethamine were representative of concentrations for intact [^{14}C]ketorolac. The ophthalmic solution was applied to both eyes of each rabbit since a control study showed negligible contralateral contamination between eyes after topical administration.[12]

After topical administration, peak concentrations of [^{14}C]ketorolac in the aqueous humor of anesthetized rabbits were achieved relatively slowly, and the time to peak aqueous humor concentrations (T_{max}) of [^{14}C]ketorolac averaged 3.4 h (Figure 1, Table I). The extent of ocular availability was determined by comparisons of aqueous humor concentrations after topical versus intracameral administration, and averaged 4% (Table I). The low ocular availability resulted partly because the topically applied drug in the precorneal area is subject to drug loss via conjunctival absorption. The conjunctival area is ~6 times larger than the corneal area and is

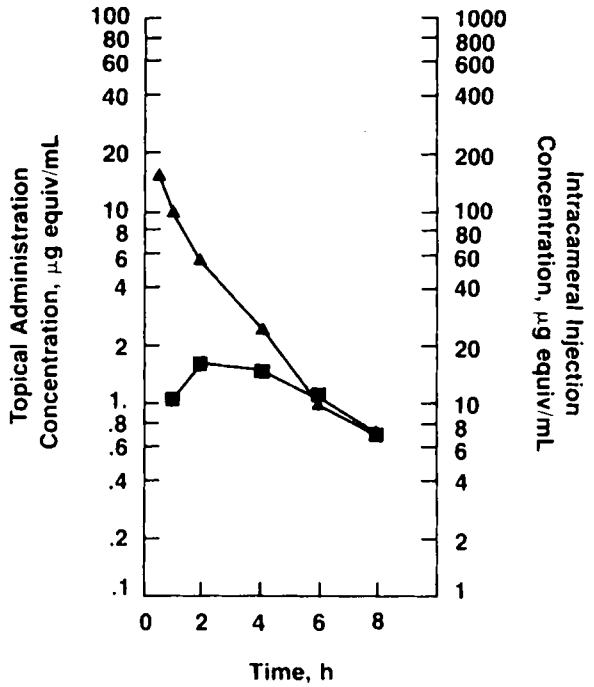

Figure 1—*Mean concentrations of total radioactivity in aqueous humor after topical administration of 50 μL of 0.5% ketorolac tromethamine solution (■) and intracameral injection of 20 μL of 1.27% ketorolac tromethamine solution (▲).*

Table I—Bioavailability and Pharmacokinetic Parameters of [^{14}C]Ketorolac in the Anterior Chamber after Ocular Administration of [^{14}C]Ketorolac Tromethamine[a]

Parameter	Topical	Intracameral
T_{max}, h	3.38 (1.69)	0.50 (0.00)
C_{max}, μg eq/mL	1.905 (0.942)	188.0 (97.7)
AUC, μg eq·h/mL	13.53 (5.72)	335.3 (184.9)
Half-life, h	3.8 (1.6)	2.11 (0.98)
F_{aq}, %[b]	3.7	—
CL_{aq}, μL/min[c]	—	11 (7)
VD_{aq}, mL[d]	—	1.93 (1.55)

[a] Values represent mean (SD); n = 12; 50 μL of 0.5% ketorolac tromethamine ophthalmic solution was administered topically; 20 μL of 1.27% ketorolac tromethamine solution was injected intracamerally. [b] F_{aq} = AUC_{aq} (topical)/AUC_{aq} (intracameral) × Dose (intracameral)/Dose (topical) × 100. [c] CL_{aq} = Dose (intracameral)/AUC_{aq} (intracameral). [d] VD_{aq} = CL_{aq}/β where β is the terminal decay rate constant of the logarithm of aqueous humor concentration–time curve.

highly vascularized.[13] Hence, the conjunctiva can be a major route for entry of drug into the systemic circulation. Drainage of an instilled drug solution and tear turnover may also contribute to a considerable loss of drug. For example, tear turnover would dilute the drug and, hence, reduce the gradient for transport through the cornea. However, rabbits under systemic or topical anesthesia, such as those in this study, were shown to have a considerably slower drainage rate and lacrimal fluid turnover compared with unanesthetized animals.[14,15] Other factors which may decrease ocular drug bioavailability are nonproductive scleral absorption and possible adsorption of drug on the lids and other eye tissues.[15]

The mean half-life of [^{14}C]ketorolac in the anterior chamber after intracameral injection (2.1 h) was shorter than the average half-lives after topical administration (Table I). This suggests that topical dosing may lead to a reservoir effect in the corneal epithelium, similar to that reported for pilocarpine.[16] Thus, the longer half-lives observed after topical doses were probably due to a continued flux of drug into the aqueous humor from the reservoir in the cornea. This was further substantiated by results from the eye tissue distribution study in rabbits, which indicated that the major fraction of [^{14}C]ketorolac was retained in the cornea and sclera during the 24-h period after topical doses (Figure 2).

The clearance of [^{14}C]ketorolac from the anterior chamber averaged 11 ± 7 μL/min (Table I). Normal aqueous humor turnover can only account for ~3 μL/min of the total ocular clearance, and there was no evidence to suggest that this turnover would be altered by 1. Thus, other possible pathways of drug elimination include precorneal loss, vitreous humor turnover, and drug removal through the intraocular venous circulation.[17] Precorneal loss is not considered to contribute substantially to drug elimination because movement of drug into the anterior segment after topical ocular doses is a rate-limiting process. Vitreous humor turnover is probably a minor pathway of drug elimination since drug concentrations in the vitreous humor were low relative to concentrations in other ocular tissues after topical doses (Figure 2). Since drug present in aqueous humor may distribute to the iris-ciliary body and be removed through the intraocular venous circulation, drug loss via intraocular blood flow possibly represented the major route of drug elimination from the eye.

The apparent volume of distribution of [^{14}C]ketorolac in the anterior chamber averaged 1.93 ± 1.55 mL (Table I) and was 6.7 times the reported value for the physiological aqueous volume (287 μL) determined via anterior chamber injection of inulin.[18] This suggests that ketorolac present in the aqueous humor distributes extensively into tissues surrounding the anterior chamber (i.e., cornea, ciliary body, iris, vitreous humor, and lens).

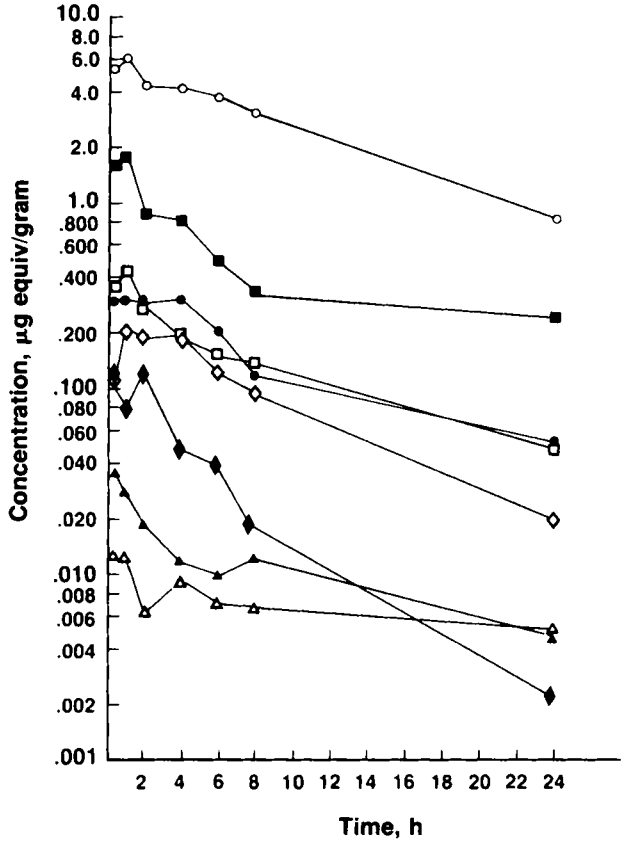

Figure 2—*Ocular distribution of [^{14}C]ketorolac in μg equivalents per g after topical administration of 50 μL per eye of 0.5% ketorolac tromethamine solution. Key: cornea (○); sclera (■); iris-ciliary body (●); retina-choroid (□); aqueous humor (◇); lens (△); vitreous humor (▲); plasma (◆).*

Systemic Bioavailability—Mean maximal concentrations of total radioactivity or intact ketorolac were maximal as early as 15 min after topical doses (Figures 3 and 4). After topical administration, the peak plasma concentration of ketorolac averaged 185 ng/mL (Table II). When [^{14}C]ketorolac tromethamine was injected directly into the anterior

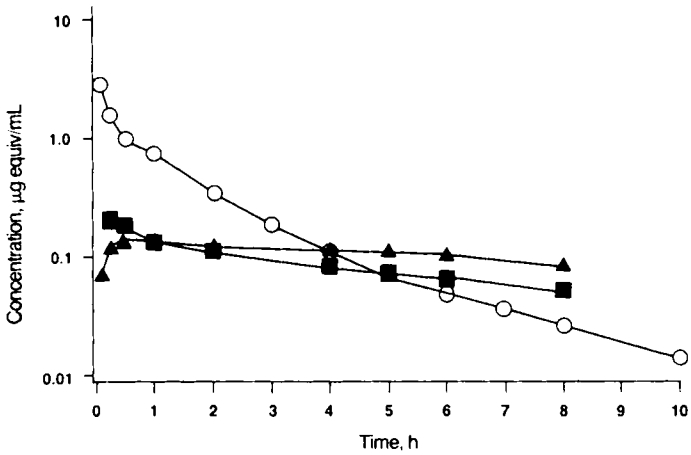

Figure 3—*Mean plasma concentrations of [^{14}C]ketorolac radioequivalents after topical administration of 50 μL of 0.5% ketorolac tromethamine solution (■), intracameral injection of 20 μL of 1.27% ketorolac tromethamine solution (▲), or intravenous administration of 1 mg of ketorolac tromethamine in solution (○).*

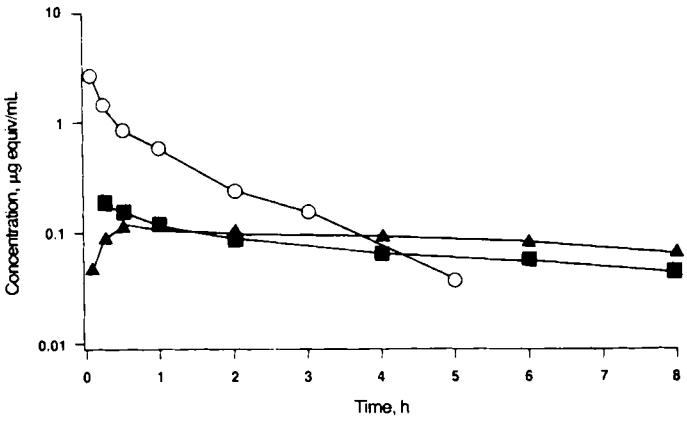

Figure 4—*Mean plasma concentrations of ketorolac after topical administration of 50 μL of 0.5% ketorolac tromethamine solution (■), intracameral injection of 20 μL of 1.27% ketorolac tromethamine solution (▲), or intravenous administration of 1 mg of ketorolac tromethamine in solution (○).*

chamber, peak plasma concentrations of total radioactivity of ketorolac were achieved later as compared with topical doses (Figures 3 and 4). Thus, the peak plasma concentration of ketorolac averaged 116 ng/mL after intracameral injection and was achieved 36 min after dosing (Table II). This suggests that after topical doses entry into the systemic circulation via conjunctival absorption was extremely rapid. When [^{14}C]ketorolac tromethamine was injected into the anterior chamber, however, plasma drug concentrations were perhaps dependent on removal of drug through the intraocular venous circulation.

After ocular or intravenous administration, it was shown that ketorolac represented the major component in plasma while *p*-hydroxy ketorolac (2) was present in small amounts.[12] The extent to which ketorolac was present in plasma in this study was similar after ocular and intravenous administration (Table II). The AUC of ketorolac averaged 91% and 78% of the total radioactivity AUC after topical administration and intracameral injection, respectively. After intravenous administration, 80% of the total radioactivity in plasma was in the form of intact ketorolac (Table II).

The systemic availability estimated, based on plasma concentrations of ketorolac, averaged 106% after topical administration (Table II). The systemic availability after intracameral injection (131%) may be overestimated since a different group of animals was used in each study. The complete systemic availability of 1 suggests that ketorolac does not bind irreversibly to eye tissues after ocular doses, as confirmed by results from the eye distribution study in rabbits (Figure 2). Although 1 was completely absorbed into the systemic circulation, concentrations of ketorolac (as reflected in AUC) were on the average 13 times higher in the aqueous humor than in plasma after topical administration (Tables I and II).

Plasma half-lives of total radioactivity and ketorolac were similar after the ocular doses and exceeded those determined after intravenous administration. The mean plasma half-lives of ketorolac were 6.9 and 7.4 h after the topical and intracameral doses, respectively, but the half-life was 1.1 h after intravenous administration (Table II). The longer plasma half-lives of ketorolac observed after the ocular doses suggest that removal of drug from the eye into the venous circulation may be rate-limiting.

The plasma clearance of ketorolac after intravenous administration averaged 6 ± 2 mL/min (Table II). Thus, after ocular doses, ketorolac was cleared at a relatively slower rate from the anterior chamber (11 μL/min), but was cleared ~500 times more rapidly once it entered the systemic circulation. The apparent volume of distribution of ketorolac in the plasma of rabbits averaged 0.52 L, indicating distribution of ketorolac from the systemic circulation into the extracellular fluid.

Ocular Tissue Distribution—The maximum tissue concentrations occurred at 0.5 to 1.0 h after topical application with the exception of the iris-ciliary body (Figure 1, Table III). However, the concentration of total radioactivity in the iris-ciliary body at 0.5 h (301 ng eq/g) was not very different from its peak value (306 ng eq/g) at 4 h. The maximum concentrations were highest in the cornea (6.06 μg eq/g) and sclera (1.73 μg eq/g), followed by the retina-choroid, iris-ciliary body, aqueous humor, vitreous humor, and lens.

The time to peak aqueous humor concentration in the unanesthetized rabbits in the eye distribution study occurred at 1 h (Table III) as compared with 3.4 h observed in anesthetized rabbits (Table I).

The average peak concentration of total radioactivity in the aqueous humor of unanesthetized rabbits (0.217 μg eq/mL) was 11% of that observed in anesthetized rabbits (1.91 μg eq/mL; Table I). The aqueous humor AUC values in unanesthetized rabbits (2.33 μg eq·h/mL) was 17% of the mean value obtained in the anesthetized animals (13.53 μg eq·h/mL). Thus, the higher aqueous humor levels in anesthetized rabbits may be primarily due to decreased tear turnover and/or instilled solution drainage. A reduction in tear turnover and solution drainage was shown in rabbits in the presence of topical or systemic anesthesia.[14,15] A similar effect was reported for pilocarpine. When pilocarpine nitrate was administered topically to anesthetized rabbits, significantly greater (2.5 times) aqueous humor levels of pilocar-

Table II—Systemic Bioavailability and Pharmacokinetics of [^{14}C]Ketorolac Tromethamine After Ocular and Intravenous Administration of [^{14}C]Ketorolac Tromethamine[a]

Parameter	Topical	Intracameral	Intravenous (1 mg)
T_{max}, min	15 (0)	36 (13)	5 (0)
C_{max}, ng eq/mL	185 (26)	116 (39)	2689 (523)
AUC, ng eq·h/mL	1074 (434)	1342 (586)	2009 (726)
AUC (KET/TR)[b]	0.91	0.78	0.80
Plasma half-life, h	6.89 (2.16)	7.40 (3.38)	1.06 (0.35)
F_p, %[c]	106	131	—
CL_p, mL/min[d]	—	—	6 (2)
Vd_p, L[e]	—	—	0.518 (0.064)

[a] Values represent mean (SD); n = 12 for topical study; n = 10 for intracameral study (due to insufficient sample collected from 1 rabbit); n = 6 for intravenous study; 50 μL of 0.5% ketorolac tromethamine ophthalmic solution was administered topically; 20 μL of 1.27% ketorolac tromethamine solution was injected intracamerally. [b] KET = ketorolac tromethamine; TR = total radioactivity. [c] F_p = AUC (ocular)/AUC (iv) × Dose (iv)/Dose (ocular) × 100. [d] CL_p = Dose (iv)/AUC (iv). [e] $Vd_p = CL_p/\beta$ where β is the terminal decay rate constant of the logarithm of plasma concentration–time curve.

Table III—Pharmacokinetic Parameters of [^{14}C]Ketorolac Radioequivalents in Eye Tissues and Plasma After Topical Application of 0.5% [^{14}C]Ketorolac Tromethamine (50 μL) in Rabbit Eyes

Tissue/Fluid	T_{max}, h	C_{max}, ng eq/mL or g	Total AUC, ng eq·h/mL or g	AUC Ratio[b]	Half-Life, h[a]
Cornea	1.0	6058	72809	103.9	8.2
Sclera	1.0	1728	18775	26.8	22.3
Iris-ciliary body	4.0	306	4049	5.8	10.4
Lens	0.5	13	417	0.6	35.4
Retina-choroid	1.0	418	3902	5.6	10.6
Aqueous humor	1.0	217	2330	3.3	7.1
Vitreous humor	0.5	36	373	0.5	14.9
Plasma	0.5	139	701	1.0	4.5

[a] Half-life determined by linear regression of the log tissue or plasma concentrations at 6, 8, and 24 h. [b] AUC (tissue or fluid)/AUC (plasma).

Table IV—Percent of Dose Recovered in Eye Tissues After a Single Topical Application (50 μL) of 0.5% [^{14}C]Ketorolac Tromethamine in Rabbit Eyes[a]

Time, h	Percent of Dose							
	Cornea	Sclera	Iris-Ciliary Body	Lens	Retina-Choroid	Aqueous Humor	Vitreous Humor	Total
0.5	0.2557 (29)	0.4826 (35)	0.0059 (28)	0.0025 (39)	0.0186 (32)	0.0130 (30)	0.0274 (39)	0.8058 (29)
1	0.2419 (34)	0.5817 (51)	0.0072 (34)	0.0023 (39)	0.0179 (35)	0.0255 (57)	0.0226 (43)	0.8991 (37)
2	0.1782 (28)	0.2694 (65)	0.0051 (46)	0.0012 (51)	0.0127 (46)	0.0234 (51)	0.0157 (65)	0.5056 (49)
4	0.1587 (18)	0.2034 (58)	0.0061 (34)	0.0016 (46)	0.0096 (54)	0.0241 (35)	0.0091 (45)	0.4121 (35)
6	0.1448 (27)	0.1344 (108)	0.0041 (34)	0.0013 (36)	0.0058 (84)	0.0133 (29)	0.0082 (63)	0.3119 (51)
8	0.1005 (13)	0.0859 (64)	0.0025 (20)	0.0012 (50)	0.0053 (63)	0.0092 (33)	0.0103 (73)	0.2150 (28)
24	0.0318 (87)	0.0645 (152)	0.0011 (54)	0.0010 (56)	0.0017 (93)	0.0026 (73)	0.0032 (75)	0.1063 (122)

[a] Values are mean (% CV) recovery in tissues/fluids of 3 rabbits (n = 6) at each sampling time.

pine nitrate were obtained while the absorption and elimination rate constants were unchanged.[15]

Relative to plasma AUC values, the AUC values were higher for the cornea (104 fold), sclera (27 fold), iris-ciliary body (5.8 fold), retina-choroid (5.6 fold), and aqueous humor (3.3 fold), and lower for vitreous humor (0.5) and lens (0.6; Table III). Concentrations of total radioactivity in the cornea, sclera, iris-ciliary body, retina-choroid, and aqueous humor were generally higher than plasma levels during the 24-h period following instillation (Figure 2). Concentrations of radioactivity in the vitreous humor and lens were lower than the plasma concentrations during the first 8 h, but exceeded the plasma levels at 24 h. This was because plasma levels declined more rapidly, with a half-life of 4.5 h as compared with the half-lives observed in the vitreous humor or lens (14.9 and 35.4 h, respectively, Table III).

The amount of radioactivity retained in the eye was maximal at 1 h post-dose, and the recovery was 0.9% of the topical dose (Table IV). Much of this radioactivity was recovered in the sclera (0.58%) and cornea (0.24%), and lower amounts in the aqueous humor (0.026%), vitreous humor (0.023%), retina-choroid (0.018%), iris-ciliary body (0.007%), and lens (0.002%).

Although [^{14}C]ketorolac tromethamine was administered topically at a dose equivalent to one-half of that administered intravenously, the total radioactivity levels were considerably higher in the cornea (200 to 645 fold) and aqueous humor (79 to 263 fold) after topical application compared with levels obtained after intravenous dosing (Table V). Differences in concentrations of total radioactivity were less remarkable in the iris-ciliary body (2.6 to 12.5 fold) and retina-choroid (5.6 to 9.5 fold); furthermore, concentrations of total radioactivity tend to be highest in these vascular tissues after intravenous dosing. The concentrations of radioactivity in vitreous humor were 11- to 16-fold higher after topical dosing; small amounts of radioactivity reached the lens tissue after topical administration, while no detectable levels were observed after intra-

Table V—Influence of Route of Administration[a] of [^{14}C]Ketorolac Tromethamine on the Concentrations of Total Radioactivity (ng eq per mL or g) in Ocular Tissues of Rabbits

Ocular Tissue/ Fluid	Time, h	Route Administered		
		Ophthalmic	Intravenous	Oph/iv
Cornea	0.5	5417	8.4	645
	1	6058	30.3	200
	5	4006[b]	11.8	339
Iris-Ciliary Body	0.5	301	144.3	2.6
	1	301	37.7	8.0
	5	253[b]	20.2	12.5
Lens	0.5	13.3	BDL[c]	—
	1	12.4	BDL	—
	5	8.5[b]	BDL	—
Retina-Choroid	0.5	359	64.1	5.6
	1	418	64.7	6.5
	5	169[b]	17.7	9.5
Aqueous Humor	0.5	111	1.4	79
	1	217	1.3	167
	5	158[b]	0.6	263
Vitreous Humor	0.5	36	2.2	16
	1	27	2.5	11
	5	11[b]	0.7	16

[a] The total ophthalmic dose (0.493 mg) instilled on both eyes of each rabbit was approximately one-half of the intravenous dose (1 mg). [b] Interpolated from 4- and 6-h values. [c] BDL = below detectable limit.

venous dosing. Much higher concentrations were also achieved in ocular tissues when timolol was administered topically as compared with intravenous administration.[8] The results of the ocular distribution study indicate little probability of back diffusion into the eye from the systemic circulation after topical administration, and support the use of **1** by the topical route for its anti-inflammatory activity.

References and Notes

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