mg) on a single day during an extended medication period was 0.51 μg/ml, in line with earlier findings. The corresponding value during a period in which the dosage was only 25 mg/day was 0.22 μg/ml. Plasma levels corresponding to these two values were 0.31 and 0.10 μg/ml, respectively. These values are presented in Table I, along with plasma levels for three individuals (Subjects G, K, and L) at dosage levels of 50 and 100 mg. Blood levels for Subject W (50 mg) at 1.5, 4, 6, and 24 hr, determined prior to introduction of the internal standard approach, are also presented in the table.

This paper presents a method for the determination of hydrochlorothiazide in small volumes of blood and plasma from persons receiving therapeutic levels of this drug. Drug levels are reported for a limited number of patients, demonstrating the adequacy of the method for measuring the drug in blood and plasma at clinical doses. The method described in this paper should provide the means for carrying out pharmacokinetic and bioavailability studies.

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[x] To whom inquiries should be directed.

Ocular Evaluation of Polyvinyl Alcohol Vehicle in Rabbits

THOMAS F. PATTON and JOSEPH R. ROBINSON [x]

Abstract □ The flow properties and viscosity of the vehicle into which drugs are incorporated can be determining factors in the bioavailability of topically applied ophthalmic drugs. It is shown, in rabbits, that when polyvinyl alcohol and methylcellulose are compared on a viscosity basis, there is essentially no difference in the two vehicles with regard to their influence on ocular drug bioavailability. Moreover, the rate of drainage loss for polyvinyl alcohol solutions, as determined by the radioactive technetium technique, compares favorably to methylcellulose solutions of similar viscosity. The relationship between viscosity and contact time or drainage loss of a drug is not a direct one, but an optimum viscosity range exists for polyvinyl alcohol solutions. This optimum range of 12–15 cps in rabbits is identical to that found for methylcellulose and differs considerably from the commonly employed viscosity in commercial preparations. Based on the methylcellulose–polyvinyl alcohol comparison, it appears that vehicles exhibiting or approximating Newtonian flow properties show comparable effects as ophthalmic vehicles. Finally, a discussion of non-Newtonian vehicles and their expected behavior in the eye is presented.

Keyphrases □ Polyvinyl alcohol—effect of vehicle viscosity on ocular drug bioavailability in rabbits, compared to methylcellulose □ Ocular bioavailability—effect of vehicle viscosity, polyvinyl alcohol and methylcellulose solutions, rabbits □ Ophthalmic vehicles—evaluation of polyvinyl alcohol, compared to methylcellulose, optimum viscosity range, rabbits

Great time and effort have been expended on determining the optimum liquid vehicle in which to incorporate drugs for instillation into the eye. Most efforts have been based on the premise that a highly viscous solution would prolong contact time of the drug with eye tissues, thus enabling a greater amount of drug to be absorbed into the desired area.

Recently, Adler *et al.* (1) made the important observation that high viscosity solutions do not greatly increase corneal contact time in humans and have only a small effect on bioavailability. An extensive study on methylcellulose solutions (2), however, has shown that there is considerable prolongation of contact time in rabbits and a two- to threefold improvement in bioavailability. This inconsistency apparently is due to differences between humans and rabbits with respect to the effect of medium viscosity solutions in the eye.

The present study explores the influence on ocular drug bioavailability of various polyvinyl alcohol solutions. Methylcellulose and polyvinyl alcohol are both used extensively as vehicles for ophthalmic drugs and the literature is confusing as to which polymer is best for ocular use. The aim of this report is to resolve that confusion as well as to provide a rational basis for predicting ophthalmic vehicle viscosity effects in general.

EXPERIMENTAL

Materials—Water was double distilled from alkaline permanganate in an all-glass distillation apparatus. Technetium solutions

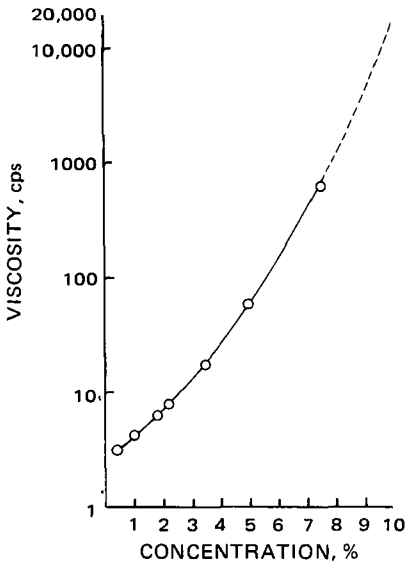

Figure 1—*Viscosity of polyvinyl alcohol solutions as a function of concentration at 30°. The dotted line represents extrapolated values for concentrations greater than 7.5%.*

were prepared using a package of solutions[1] and equipment used to prepare technetium, ^{99m}Tc, colloidal suspensions. The preparative procedure was described previously (3).

Polyvinyl alcohol[2], 20–90 grade, was used. Tritiated pilocarpine was obtained commercially[3] and was purified immediately before use. The reasons for the purification and the methods used were reported earlier (4). The specific activity of pilocarpine was 6.95 Ci/mmole. All other chemicals were either reagent or analytical grade.

Male, albino rabbits[4], 1.8–2.4 kg, were fed a regular diet with no restrictions on food or water intake.

Methods—*Determination of Instilled Solution Drainage Rate*—The general procedure for the determination of drainage rate by the nonsampling method was reported previously (3). In the present studies, twice the desired concentration of polyvinyl alcohol was diluted with an equal volume of technetium suspension to obtain the final concentration. The resulting solution was equilibrated in a water bath at 30° prior to instillation. Then 25 μl of polyvinyl alcohol–technetium solution was instilled with a microliter syringe[5], the probe was positioned, and the experiments were carried out as described previously (3).

Viscosity Studies—Viscosity profiles of the various polyvinyl alcohol solutions were obtained using a viscometer[6] and a No. 1 spindle. Solutions were maintained at 30 ± 0.1° throughout the viscosity measurements.

Aqueous Humor Drug Studies—The experimental details for aqueous humor drug concentration studies were reported previously (5). Unanesthetized rabbits were used in all experiments.

RESULTS

Viscosity Studies—Dilute polymer solutions generally exhibit or approximate Newtonian flow properties (6), and this was the case in the methylcellulose studies mentioned previously (2). It is well known that aqueous solutions of polyvinyl alcohol exhibit non-Newtonian viscosity (7). However, over the concentration range employed, the results approximated Newtonian behavior.

The relationship between viscosity and concentration of polyvinyl alcohol solutions at 30° is illustrated in Fig. 1. It is clear that dilute solutions of polyvinyl alcohol (<3%) show relatively small increases in viscosity with concentration. As the concentration of polyvinyl alcohol is further increased, however, viscosity changes may cover several orders of magnitude. It is known (8) that aqueous solutions of polyvinyl alcohol greater than approximately 1% contain entangled aggregates of hydrogen-bonded molecules. Such solutions may vary in their viscosities with age and with applied shear due to gelling.

[1] Collokit, Abbott Radio-Pharmaceuticals, North Chicago, Ill.
[2] Supplied by Allergan Pharmaceuticals, Irvine, Calif.
[3] New England Nuclear, Boston, Mass.
[4] Klubertanz, Edgerton, Wis.
[5] Hamilton Co., Reno, Nev.
[6] Brookfield model LVT.

Table I—Drainage Rate Constant of Polyvinyl Alcohol Solutions as a Function of Concentration and Viscosity

Concentration, %	Viscosity, cps	k, min^{-1}	Number of Determinations
0.0[a]	1.0	0.54	3
0.5	3.1	0.32 (0.05)[b]	5
1.1	4.2	0.25 (0.03)	5
1.9	6.1	0.25 (0.03)	10
2.25	7.7	0.31 (0.04)	5
3.5	17.5	0.08 (0.02)	7
5.0[c]	57.8	0.14 (0.02)	9

[a] Refers to data obtained using pure technetium suspension (2). [b] Values in parentheses refer to standard error of the mean. [c] Considerable difficulty was encountered in instilling this solution as well as in mixing it with the technetium suspension. These factors may limit the reliability of the drainage rate data.

Drainage Rate of Polyvinyl Alcohol Solutions and Its Relationship to Viscosity and Concentration—Loss of an instilled drug solution through drainage can account for a tremendous decrease in the amount of drug available for absorption (3). Thus, a small drainage rate would favor increased drug absorption. The drainage rates of various concentrations of polyvinyl alcohol were determined as described under *Experimental*. Table I lists these data as well as the viscosities of the various polyvinyl alcohol solutions.

A very dilute (0.5%) solution of polyvinyl alcohol causes a significant decrease in the drainage rate constant over that of an aqueous solution. This decrease occurs over a threefold increase in solution viscosity. From 0.5 to 2.25% polyvinyl alcohol, another two- to threefold increase in viscosity occurs and the drainage rate constant remains essentially unchanged. Somewhere beyond 3% polyvinyl alcohol, the drainage rate undergoes another sharp decrease and then remains fairly constant.

Figure 2 illustrates the drainage rate *versus* viscosity data. Drawing a smooth curve through the points results in a major change in the slope of the line at about 12–15 cps, which is shown as the hatched area in Fig. 2. This area corresponds approximately to a 3% solution (see Fig. 1). The conclusion then is that approximately a 3% polyvinyl alcohol solution is the optimum because large increases in viscosity result in only minor decreases in drainage rate beyond this concentration. The concentration of polyvinyl alcohol commonly used commercially is 1.4%; presumably other factors, such as comfort, account for the difference.

Aqueous Humor Levels of Philcarpine Nitrate in Polyvinyl Alcohol Vehicle—It seemed reasonable to assume that a viscous

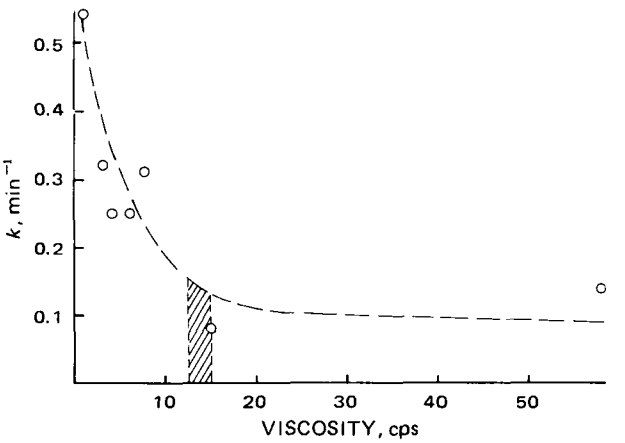

Figure 2—*Drainage rate of various polyvinyl alcohol solutions as a function of solution viscosity.*

Table II—Aqueous Humor Levels of Pilocarpine Nitrate from Various Viscosity Polyvinyl Alcohol Solutions

Polyvinyl Alcohol, %	Aqueous Humor[a], μg/ml		
	20 min	30 min	60 min
0	1.20 (0.10)[b]	0.66 (0.06)	0.67 (0.05)
0.5	1.16 (0.14)	1.38 (0.08)	
1.1	1.26 (0.20)	0.94 (0.06)	0.34 (0.03)
1.9	1.14 (0.09)	2.03 (0.31)	
2.25	1.11 (0.07)	1.28 (0.11)	0.27 (0.04)
3.5	1.65 (0.22)	1.48 (0.20)	
5.0	2.22 (0.20)	2.01 (0.28)	
10.0	5.41 (0.38)	3.18 (0.64)	2.05 (0.19)

[a] At least six determinations at each point. [b] Numbers in parentheses refer to standard error of the mean.

solution would prolong contact time of drug with the eye through decreased drainage loss and would provide the potential for increased drug absorption into and through the cornea. Since the decrease in drainage rate had already been shown for various polyvinyl alcohol solutions, the effect on aqueous humor drug concentration remained to be demonstrated.

Table II shows the results of various polyvinyl alcohol concentrations on aqueous humor drug levels at selected time points. There appears to be little if any improvement in aqueous humor levels until something beyond 2.25% polyvinyl alcohol is used. A rather dramatic increase is noted with 10% polyvinyl alcohol, but the solution is so viscous at this concentration that it probably blocks the drainage duct and would also cause marked blurring of vision. These drawbacks probably would make it impractical for commerical use.

There is greater scatter in some of the data than was experienced in the past; based on experimental technique, an explanation for the deviation in the data cannot be offered. Nevertheless, trends can be discussed. For example, the 3.5, 5.0, and 10.0% polyvinyl alcohol solutions are all consistently higher than the lower concentrations.

DISCUSSION

To discuss viscosity effects, one must first have some understanding of the types of flow exhibited by various systems as well as an appreciation as to how these different systems behave in animal or human studies. For example, rabbits blink considerably less frequently than humans and therefore exert less stress on a polymer system.

The flow properties of any given system can have, in theory, a significant effect on the behavior of that system when instilled into the eye. By knowing the flow properties of a system, it should be possible to make some statements regarding its expected activity in the eye, as well as to account for possible differences in humans and animals. This information is summarized in Table III.

As mentioned, numerous studies have been conducted on methylcellulose and polyvinyl alcohol, both in rabbits and in humans, in an effort to determine the ideal ophthalmic vehicle. Almost all of these studies considered only vehicle contact time or bioavailability and ignored the very important aspects of patient acceptability and desirable properties for pharmaceutical handling, *e.g.*, sterile filtration. Many of these studies were confusing, contradictory, and, at best, incomplete. If, however, these studies are discussed in terms of the information in Table III and the findings reported here, the picture begins to become somewhat clearer.

In one early study, homatropine hydrobromide in 1% methylcellulose was administered to human subjects; this combination induced greater cycloplegia and mydriasis than a simple aqueous solution (9). The increased effectiveness was attributed to an increased contact time of drug with the eye. It was concluded that about double the quantity of drug could be expected to penetrate by increasing the viscosity of the instilled solution.

This effect was substantiated in a study in rabbits where a solution of 0.5% methylcellulose (4000 cps) increased contact time by 334% over an aqueous solution (10). Haas and Merrill (11), using human subjects, determined that a solution of pilocarpine in 0.5% methylcellulose produced greater miosis than a nonmethylcellulose solution of equal drug concentration.

Krishna and Brow (12), comparing vehicles, found that 1.4% polyvinyl alcohol in isotonic saline afforded greater surface contact time than 0.5% methylcellulose in isotonic saline alone. The results of this study, performed in rabbits, seem rather strange since 0.5% methylcellulose has a much higher viscosity than a 1.4% polyvinyl alcohol solution, assuming the standard grades of each were used.

It was found that in rabbits the increase in contact time of methylcellulose ophthalmic vehicles was about proportional to the

Table III—Various Viscosity Types and Their Expected Behavior in the Eye

Flow Type	Characteristics	Example	Potential Ocular Effects
Newtonian	Constant viscosity at constant temperature and pressure; viscosity independent of rate of shear	Simple liquids such as water and glycerin	Drainage loss of drug solution should be inversely proportional to viscosity; due to shear independence, blinking should have no effect on viscosity and such systems should behave the same in rabbits and humans
Non-Newtonian Pseudoplastic	Stress increases more rapidly at low rates of shear than at high rates; the apparent viscosity decreases as shear stress (shear thinning) increases	Concentrated dispersions of small particles and solution of long chain molecules	If the system undergoes shear in the eye, this system would be poor; in humans, if blinking causes shear, the system will thin and drain from the eye; in rabbits, such a system will probably resemble a Newtonian system
Plastic	Resembles pseudoplastic system, but the rate of shear does not acquire a finite value until the stress exceeds a certain yield value	Modeling clay	This system would be good as long as the yield value is not exceeded; in rabbits, this may be true, but in humans the system thins as with pseudoplastic once the yield value is exceeded
Dilatant	The opposite of pseudoplastic; the force increases faster than the rate of shear	Wet sea sand	If blinking in humans produces shear, the system will thicken; this system has good potential in humans, although the advantages of such a system would probably not be observed in rabbits
Thixotropic	A reversible and noninstantaneous decrease in apparent viscosity upon shear; the effect increases with the rate of shear; time-dependent shear thinning	Bentonite magma	Probably not a desirable situation in humans or in rabbits since the system will thin with time

Table IV—Comparison of Methylcellulose and Polyvinyl Alcohol Vehicles in Rabbit Eyes

Methylcellulose (1)		Polyvinyl Alcohol	
From 1 to 15 cps, drainage rate constant decreases by a factor of 3; from 15 to 100 cps, drainage rate constant further decreases by a factor of 3		From 1 to 7.7 cps, drainage rate constant decreases by a factor of 2; another two- to threefold decrease in k from 7.7 to 57.8 cps	
A rapid decrease in drainage rate with a small increase in viscosity, followed by a smaller decrease in drainage with a large increase in viscosity		True with polyvinyl alcohol also	
cps	k, min^{-1}	cps	k, min^{-1}
4.2	0.36	4.2	0.25
12.5	0.19	17.5	0.08
40.0	0.14	57.8	0.14
Approximately a 1.6-fold increase in aqueous humor concentration at 20 min from 1 to 12.5 cps, followed by a 1.2-fold increase in concentration from 12.5 to 100 cps		Approximately a 1.4-fold increase in aqueous concentration at 20 min from 1 to 17.5 cps, followed by a 1.4-fold increase from 17.5 to 57.8 cps	

viscosity of the solution in the lower viscosity region (5–25 cps) and leveled off at 55 cps or above (13). Even though methylcellulose is strictly a pseudoplastic system, Table III predicts Newtonian behavior in rabbits, making the 5–25-cps findings consistent. When a methylcellulose solution at 55 cps was used, mydriasis from a 0.02% solution of scopolamine hydrobromide was doubled.

Linn and Jones (14) contradicted the results of Krishna and Brow (12) by finding that the length of time needed for 0.5% hydroxypropyl methylcellulose to pass through the drainage apparatus was about twice that of 1.4% polyvinyl alcohol. This work, however, was carried out in humans whereas the earlier work was carried out in rabbits.

Bach *et al.* (15), using rabbits, and Waltman and Patrowicz (16), using human subjects, confirmed the superiority of 0.5% methylcellulose over 1.4% polyvinyl alcohol, the former in preventing ocular infections with neomycin sulfate and the latter in producing corneal and aqueous humor fluorescein concentrations. Again, these findings appear reasonable due to the higher viscosity imparted by a 0.5% methylcellulose solution over a 1.4% polyvinyl alcohol solution. Other studies (17, 18) also confirmed this finding.

A comprehensive evaluation of methylcellulose solutions in improving ocular drug bioavailability was conducted (2). Methylcellulose delayed solution drainage from the eye, and the major change occurred in the lower viscosity range. Moreover, it was found that pilocarpine aqueous humor levels could be approximately doubled through the use of methylcellulose solutions.

Despite some of the seeming contradictions in these studies, it does seem reasonable to assume that a viscous solution would prolong the contact time of drug with the eye and provide the potential for greater drug absorption into the desired area. Recently, however, this belief was challenged by Adler *et al.* (1) who concluded that increasing vehicle viscosity does not prolong ocular contact time nor markedly improve ocular drug bioavailability in humans. If the vehicles under consideration are behaving as Newtonian systems, these findings appear intuitively incorrect (Table III); however, if the vehicles are, in fact, pseudoplastic, the shear created by blinking would cause them to thin so that a large increase in instilled solution viscosity might provide only slight increases in contact time and aqueous humor levels. This is exactly what was found.

In almost all studies comparing polyvinyl alcohol and methylcellulose, the conclusion is reached that one vehicle is superior to the other based on contact time or bioavailability. But in many cases, the authors are basing their conclusions on polymer concentrations rather than viscosity. It is intuitively clear that as long as two vehicles exhibit the same flow properties, solutions with the same viscosity should exhibit the same drainage behavior in the eye. The important factors to consider then are the flow properties of the vehicle in question and its viscosity—*not* the concentration. There is no reason to suspect that methylcellulose should behave any differently from polyvinyl alcohol in the eye when a comparison is made on an equal viscosity basis.

A comparison between data obtained in this laboratory for methylcellulose (2) and the present report on polyvinyl alcohol seems to bear out this conclusion. Table IV summarizes the more important points of the polyvinyl alcohol–methylcellulose comparison. Of course, the basis for distinguishing the superiority of one vehicle over another must include factors other than drainage and bioavailability such as patient acceptability, influence on wound healing, and pharmaceutical handling.

It is clear from most of the work that has been conducted in humans (9, 18) or animals (13, 15), as well as from the methylcellulose and polyvinyl alcohol studies in this laboratory, that contact time is *not* proportional to drug bioavailability. Even with as much as a 100-fold increase in viscosity and a 10-fold decrease in drainage rate, the maximum improvement in drug activity, be it miosis, inhibition of infection, or aqueous humor levels, is about twice that of an aqueous solution.

The explanation for this is not clear. However, it is possible that a shear is created in the eye, even in rabbits, which causes these systems to thin. Thus, the responses obtained are not proportional to what would be expected from a consideration of the viscosity of the instilled solutions. Of course, more than a twofold increase in aqueous humor levels was obtained with pilocarpine in 10% polyvinyl alcohol. However, as pointed out earlier, the 10% polyvinyl alcohol is probably not practical because of the blurring of vision which would accompany its use. The relatively large increase in drug levels is probably due to a blockage of the drainage duct.

Large decreases in the drainage rate of instilled solutions can be obtained with relatively minor alterations in solution viscosity, and these large decreases do not lead to significant improvement in aqueous humor levels. With both methylcellulose and polyvinyl alcohol, it appears that the optimum viscosity solution to use would be in the range of 12–15 cps. Further increases in viscosity above this level do not appear to give proportional increases in drug levels. In addition, viscosities greater than 15 cps may present problems with respect to accuracy of instillation and blurring of vision. For these reasons, using polyvinyl alcohol, methylcellulose, or any system which approximates Newtonian behavior, a viscosity of 12–15 cps seems to be the optimum value in rabbits. Further work needs to be done to clarify why the substantial lowering of drainage rate and the corresponding increase in contact time with low to medium viscosity solutions is not reflected in a proportional increase in drug bioavailability.

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[x] To whom inquiries should be directed.

Absorption, Distribution, and Metabolic Fate of 7-Chloro-3,3a-dihydro-2-methyl-2*H*,9*H*-isoxazolo-(3,2-*b*)(1,3)-benzoxazin-9-one in Rats, Dogs, and Humans

JEROME EDELSON [x], J. F. DOUGLAS, B. J. LUDWIG, E. B. SCHUSTER, and S. SHAHINIAN

Abstract □ The absorption and metabolic fate of 7-chloro-3,3a-dihydro-2-methyl-2*H*,9*H*-isoxazolo-(3,2-*b*)(1,3)-benzoxazin-9-one (I) was studied in rats, dogs, and humans. Orally administered I was readily absorbed by all species. In the rat, orally administered I was converted to its metabolite, 5-chlorosalicylic acid, by the intestinal wall. The half-lives of blood radioactivity, after the oral administration of I-9-^{14}C, were about 18 and 12 hr in the rat and beagle hound, respectively. In human subjects, no intact I was detected in the bloodstream; however, the clearance of the metabolite, 5-chlorosalicylic acid, had a half-life of about 33 hr. Cleavage of the oxazine ring of I generated 5-chlorosalicylic acid, which was excreted both in the free form and conjugated with glycine and glucuronic acid. The isoxazole moiety was converted to β-hydroxybutyric acid and its metabolites carbon dioxide and fumaric, citric, α-ketoglutaric, succinic, and malic acids. Binding of I to plasma proteins was extensive but was less than that of 5-chlorosalicylic acid.

Keyphrases □ 7-Chloro-3,3a-dihydro-2-methyl-2*H*,9*H*-isoxazolo-(3,2-*b*)(1,3)-benzoxazin-9-one—absorption, distribution, and metabolic fate, rats, dogs, and humans □ 5-Chlorosalicylic acid—identified as major metabolite of 7-chloro-3,3a-dihydro-2-methyl-2*H*,9*H*-isoxazolo-(3,2-*b*)(1,3)-benzoxazin-9-one in rats, dogs, and humans □ GLC—analysis, 7-chloro-3,3a-dihydro-2-methyl-2*H*,9*H*-isoxazolo-(3,2-*b*)(1,3)-benzoxazin-9-one in plasma

7-Chloro-3,3a-dihydro-2-methyl-2*H*,9*H*-isoxazolo-(3,2-*b*)(1,3)-benzoxazin-9-one (I) is a new orally active anti-inflammatory agent, which is chemically dissimilar to the steroids. Unlike most other clinically effective nonsteroidal anti-inflammatory agents, I is not ulcerogenic in the rat and does not induce GI blood loss as measured with ^{51}Cr-labeled rat blood cells (1). Studies on the absorption, distribution, metabolism, and excretion of I in rats, dogs, and human subjects are presented in this report.

EXPERIMENTAL

Materials—I-9-^{14}C and I-3-^{14}C were synthesized using a recently patented procedure (2). Since I has asymmetric carbon atoms at positions 2 and 3a and is obtained as a mixture of diastereoisomers, the chromatographic systems employed (Table I) were chosen carefully in order not to resolve the diastereoisomeric pairs.

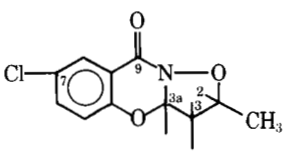
Absorption in Rats—The small intestine of male Sprague–Dawley rats, 230–250 g, was rapidly filled with a solution of 90 μg/ml of I-9-^{14}C (2.13 μCi/ml), using a previously described technique (3). Constant blood volume was maintained by transfusion with blood from a donor animal. At appropriate intervals, samples were taken of hepatic portal and systemic blood, and portions of the contents of the intestinal lumen were removed. These materials were assayed for radioactivity and subjected to TLC. The developed plates were scanned for radioactivity, and the area under the trace was determined by integration to measure the ^{14}C in each zone.

Rat Intestine Preparation—Mature male Sprague–Dawley rats were exsanguinated, and the entire small intestine was excised. A fine mince was prepared by cutting the tissue with scissors and scalpels, and the preparation was centrifuged at 48,000×*g* at 4°. Three milliliters of the supernatant fluid was incubated at 37° with 2.0 ml of I-9-^{14}C (57 μg/ml, 1.35 μCi/ml) for 1.5 hr with shaking. Then an equal volume of acetone was added, and the mixture was again centrifuged. The supernatant fluid, containing over 95% of the radioactivity, was spotted on thin-layer plates and treated as already described.

Concentration in Circulating Blood—A solution containing 5 mg of I-9-^{14}C (129 μCi) dissolved in 0.5 ml of polyethylene glycol 400 was administered by stomach tube to each of six male Sprague–Dawley rats, weighing an average of 293 g. Six other rats received 0.5 mg of I-3-^{14}C (7.43 μCi) dissolved in 0.5 ml of the same vehicle. Blood samples were taken from the tail vein at appropriate intervals, and radioactivity was determined by liquid scintillation counting (4).

Beagle hounds, 7.6–10.2 kg, received single oral doses of I-9-^{14}C of 30.8 mg/kg (three animals, 42.9 μCi each), 66 mg/kg (one animal, 495 μCi), and 490 mg/kg (one animal, 1.98 μCi). At suitable intervals, blood was taken from the jugular vein and the plasma radioactivity of each sample was determined.

Human Studies—Six human volunteers each received 1500 mg of nonradioactive I. Heparinized blood samples were taken at various times postadministration and immediately centrifuged. The plasma was frozen until assayed by GLC.



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