from the β-cyclodextrin system than from the caffeine system. Since the total drug available for release in these systems was comparable, the rate of release in the caffeine system must have declined faster than in the β-cyclodextrin system.

To confirm the present interpretation, the permeation study was repeated using the saturated solution of the drug in 1% β-cyclodextrin (Fig. 5). At this concentration, the solubility of the complex was not exceeded. As can be seen from Fig. 5, the total amount of the drug available for permeation was equal to that of the previously studied permeation from the saturated solution of drug in 2% caffeine (*i.e.*, 0.31 mmole). Therefore, the permeation profiles presented in Fig. 5 for the 2% caffeine system and the 1% β-cyclodextrin system should have a common plateau. If so, the faster initial rate of release in the 2% caffeine system must lead to a shorter release time than in the 1% β-cyclodextrin system. The following generalization may, therefore, be made. For a fixed amount of total (free and complexed) drug, the more stable the complex is, the greater is the tendency of the system to sustain the release of drug; *i.e.*, the release of drug extends over a longer period.

The permeation profile from 1% caffeine is also shown in Fig. 5. Since the amount of total drug in 1% β-cyclodextrin was greater than that in the 1% caffeine system (Fig. 4), more drug was expected to be released from the 1% β-cyclodextrin system than from the 1% caffeine system at infinite time, even though the release rates were identical during the first 5 hr.

The following conclusions may therefore be drawn for the release of drug from its saturated solution containing various amounts of complexing agents. If such systems are capable of forming soluble, membrane-impermeable complexes, the release rate of drug from such systems is greater than that from the plain saturated solution of the drug, although never exceeding that from the suspension in water. Control of the release profile of drug between these limits may be possible by means of a proper choice of complexing agents. It is evident that the more stable the complex is, the greater is the reservoir of the drug available for release.

It has also been shown that for a fixed amount of total drug (*i.e.*, fixed amount of complex since free drug is independent of the complexing agent), the more stable the complex is, the slower is the initial rate of release but the longer is the time required for complete release. Therefore, control of permeation of drug by means of complexation may find its practical value in obtaining slow sustained release from membrane-encapsulated dosage forms containing drug in solution.

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ACKNOWLEDGMENTS AND ADDRESSES

Received March 31, 1975, from the *Faculty of Pharmaceutical Sciences, Hokkaido University, Sapporo 060, Japan.*

Accepted for publication July 21, 1975.

Supported by a grant from the Ministry of Education, Japan.

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Tritiated Naltrexone Binding in Plasma from Several Species and Tissue Distribution in Mice

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Abstract □ The binding of 15,16-^{3}H-naltrexone in human, monkey, dog, guinea pig, rat, and mouse plasma was investigated over a range of concentrations, including predicted therapeutic levels. Studies using equilibrium dialysis at 37° indicate that the extent of binding is independent of naltrexone concentration over the concentration range of 1–500 ng/ml for dog plasma and of 0.1–500 ng/ml for human, monkey, guinea pig, rat, and mouse plasma. The extent of naltrexone binding in plasma is similar in the six species studied, the range being from 20% bound in rat plasma to 26% in plasma from beagle and mongrel dogs. This relatively low extent of naltrexone binding in plasma is consistent with previous findings of a large apparent volume of distribution for this drug in the dog. To investigate further the distribution of tritiated naltrexone, the tissue levels of radioactivity in mice at 1, 5, and 15 min after intravenous administration of 8-^{3}H-naltrexone were determined. Naltrexone was rapidly distributed from plasma to tissues, with less than 4% of the dose being present in plasma at 1 min after injection.

Keyphrases □ Naltrexone—plasma protein binding in humans, monkeys, dogs, guinea pigs, rats, and mice, tissue distribution in mice □ Protein binding, plasma—naltrexone, humans, monkeys, dogs, guinea pigs, rats, and mice □ Distribution, tissue—naltrexone, mice □ Pharmacokinetics—naltrexone, humans, monkeys, dogs, guinea pigs, rats, and mice □ Antagonists, narcotic—naltrexone, pharmacokinetics, humans, monkeys, dogs, guinea pigs, rats, and mice

Naltrexone, a narcotic antagonist, has been suggested for use in the treatment of heroin dependence (1). Since narcotic antagonists provide no stimulus to the patient to return for frequent dosing (2), the availability of a sustained-release drug delivery system is desirable. Such potential delivery systems for naltrexone have been prepared and tested (3, 4).

It is well known that binding to plasma proteins can influence significantly the distribution of a drug, its pharmacokinetic profile, and its duration of phar-

macological action (5, 6). Therefore, comparative data on naltrexone distribution and binding in several species and in humans must be available for rational development of model animal systems for: (*a*) pharmacokinetic analysis of naltrexone, (*b*) pharmacokinetic and pharmacological evaluation of sustained-release naltrexone delivery systems, and (*c*) toxicological analysis of naltrexone and its delivery systems.

The purposes of the present study were to determine the binding characteristics of naltrexone in plasma from humans, monkeys, dogs, guinea pigs, rats, and mice over a range of therapeutic plasma concentrations and to determine the extent of tissue distribution of radioactivity in the mouse at 1, 5, and 15 min after intravenous administration of tritiated naltrexone.

EXPERIMENTAL

Reagents—The analytical grade reagents used were chloroform, absolute methanol, 1-butanol, and benzene. Concentrated ammonium hydroxide, acetic acid, monobasic potassium phosphate, and dibasic sodium phosphate were reagent grade.

Purity and Exchangeability of 8-^{3}H- and 15,16-^{3}H-Naltrexone—To an ampul containing 4.64 mCi (2 mg) of 8-^{3}H-naltrexone[1] as a solid was added 4.0 ml of 10% absolute ethanol in benzene containing 2 mg of unlabeled naltrexone[2]. The final concentration was 1.16 mCi (1 mg)/ml. The sample of 15,16-^{3}H-naltrexone[3], 45.0 mCi (1 mg), was received dissolved in 10% ethanol in benzene. This solution was diluted to 100 ml with the same solvent, yielding a final concentration of 0.45 mCi (10 μg)/ml. These solutions were stored under nitrogen in a refrigerator. In the preparation of aqueous solutions from these stock solutions, the organic solvents were removed by evaporation in a stream of nitrogen prior to the addition of buffer.

The purity of the 8-^{3}H- and 15,16-^{3}H-naltrexone was checked periodically by TLC on 0.25-mm thick sheets of silica gel on aluminum[4]. Each radioactive naltrexone stock solution (1 μl) was spotted on the thin-layer sheets after applying 5 μg of unlabeled naltrexone in a volume of 5 μl of methanol. The solvent systems used were: A, chloroform–methanol–concentrated ammonium hydroxide (46 ml:4 ml:2 drops)[5]; B, methanol–1-butanol–benzene–water (60:15:10:15) (7); and C, 1-butanol–acetic acid–water (60:15:30) (7). The times required for development at room temperature in Solvent Systems A, B, and C were 50, 150, and 240 min, respectively.

To determine the distribution of radioactivity on the thin-layer sheets, the silica gel was removed in small, measured increments by scraping. These samples were then placed in liquid scintillation counting vials. Four milliliters of distilled water was added to each vial, and the mixture was suspended with 10 ml of an all-purpose scintillation fluid[6]. The samples were assayed in a liquid scintillation spectrometer[7] equipped with automatic external standardization. The degree of quenching did not vary significantly from vial to vial.

The exchangeability of 8-^{3}H-naltrexone was determined in the following manner. A solution containing 86 ng (0.1 μCi) in 10 ml of phosphate-buffered saline (0.01 *M* phosphate buffer made isotonic with saline, pH 7.4) was prepared. At various times after preparation of the solution, a 1.0-ml aliquot was removed and a 100-μl sample of this aliquot was placed in a counting vial along with 0.4 ml of phosphate-buffered saline and 10 ml of all-purpose scintillation fluid. This assay yielded the concentration of total radioactivity in the sample.

To the remaining 0.9 ml of the aliquot was added 0.9 ml of a suspension of dextran-coated charcoal (dextran, 62.5 mg; phosphate-buffered saline, 25 ml; and activated charcoal, 2.5 g). The mixture was shaken intermittently for 3–4 min and then gravity filtered through filter paper[8]. A 0.5-ml sample of the filtrate was assayed. This filtrate should have contained any tritiated water formed by exchange plus any nonadsorbed drug. Therefore, the radioactivity in the filtrate indicates the maximum extent to which exchange could have occurred.

Less than 0.5% of the total radioactivity occurred in the filtrate from 0 to 48 hr and less than 1% from 48 to 96 hr. Thus, there appears to be no appreciable exchange of label with water over the time studied. Similarly, the label of 15,16-^{3}H-naltrexone was shown to be stable (less than 0.5% radioactivity in filtrate) for at least 6 hr in phosphate-buffered saline.

The lack of appreciable exchange of the tritium label of 8-^{3}H-naltrexone with water *in vivo* is indicated by the essentially complete recovery of label, 97.9 ± 7.9% (mean ± *SD*), obtained in the distribution study described later. Any tritiated water formed would have been lost when the samples were dried prior to preparation for counting. Likewise, the label of 15,16-^{3}H-naltrexone has been shown not to exchange with water *in vivo* when, after administration to guinea pigs, essentially total recovery of tritium, 97.6 ± 2.7%, in the dried excreta was obtained under conditions similar to those described for the distribution study.

Blood Collection—Blood from two mongrel dogs, two foxhounds, 14 beagles, and three human subjects was collected in heparinized syringes by venipuncture. Guinea pigs, rats, and mice were lightly anesthetized with ether, decapitated, and bled into heparinized beakers. After the collection of blood from these species, plasma was separated by centrifugation and used within 24 hr (storage at 10°). Fresh monkey (*Macca mulatta*) plasma samples from two males and one female were received frozen and then thawed just prior to use.

Equilibrium Dialysis—Dialysis (8) was performed using equilibrium dialysis cells[9], the half-cells being separated by a strip of cellophane membrane[10]. One milliliter of plasma was placed in one half-cell; 1 ml of 0.16 *M* phosphate buffer, pH 7.4 (prepared by mixing 0.16 *M* KH_2PO_4 and 0.16 *M* Na_2HPO_4 in a volume ratio of approximately 1:4), containing the desired concentration of 15,16-^{3}H-naltrexone and unlabeled naltrexone was placed in the other half-cell. (The 15,16-^{3}H-naltrexone was used in the binding studies, because its high specific activity permitted the measurement of binding at concentrations down to 0.1 ng/ml.) Each half-cell contained a small glass bead to aid in mixing. The dialysis cells were wrapped in plastic and placed in a shaking water bath at 37°.

In some cases, buffer containing drug, 2 ng/ml, was dialyzed against buffer alone to demonstrate that equilibrium across the membrane had been achieved by 24 hr. At the end of 24 hr, the ratio of radioactivity on the side initially without drug to that initially with drug was 0.994 ± 0.039, $n = 4$.

After 24 hr, duplicate 0.1-ml samples were removed from each half-cell, placed in a liquid scintillation counting vial with 10 ml of scintillation cocktail[11], and assayed by liquid scintillation counting. Count rates were corrected for counting efficiency, as determined by an external standard and reference to a quench correction curve; results are expressed as concentration of naltrexone.

The percentage of total naltrexone in the plasma that was bound was calculated from:

$$\text{percentage bound} = \frac{C_p - (C_b \times F)}{C_p} \times 100\% \qquad \text{(Eq. 1)}$$

where C_p = naltrexone concentration in plasma, C_b = naltrexone concentration in buffer, and F = fraction of plasma that is water.

The fraction of plasma that is water, F in Eq. 1, is used to correct for the volume occupied by plasma solutes, mainly proteins. The values of F for humans, dogs, and rats are 0.94 (9), 0.93 (9), and 0.935 (10), respectively. Values of F for guinea pig and mouse plasma were determined to be 0.938 ± 0.009 and 0.935 ± 0.001, re-

[1] Lot 1620-124, provided under National Institute on Drug Abuse Contract HSM 42-73-184 with Research Triangle Institute, Research Triangle Park, N.C.
[2] Endo Laboratories, Inc., Garden City, N.Y.
[3] Lot 1681-17-6, provided under National Institute on Drug Abuse Contract HSM 42-73-184 with Research Triangle Institute, Research Triangle Park, N.C.
[4] E. M. Reagents, E. M. Laboratories, Elmsford, N.Y.
[5] This solvent system was suggested by Dr. J. A. Kepler, Research Triangle Institute, Research Triangle Park, N.C.
[6] Instagel, Packard Instrument Co., Downers Grove, Ill.
[7] Packard Tri-Carb model 3375, Parkard Instrument Co., Downers Grove, Ill.
[8] Whatman No. 1, W & R Balston, Ltd., Maidstone, England.
[9] Chemical Rubber Co., Cleveland, Ohio.
[10] Will Scientific, Rochester, N.Y.
[11] Aquasol, New England Nuclear, Boston, Mass.

Table I—TLC Analysis of Purity of 8-^{3}H-Naltrexone and 15,16-^{3}H-Naltrexone

Solvent System	8-^{3}H-Naltrexone R_f[a]	8-^{3}H-Naltrexone Percentage Recovered[b]	15,16-^{3}H-Naltrexone R_f[a]	15,16-^{3}H-Naltrexone Percentage Recovered[b]
A	0.43[c]	95.1	0.54[c]	97.4
B	0.36	97.2	0.33	97.7
C	0.31	98.8	0.32	98.2

[a] R_f value of authentic naltrexone. [b] Percentage of radioactivity recovered from the plate, corrected for plate background, at the R_f value corresponding to authentic naltrexone. [c] The R_f value of naltrexone in Solvent System A varied up to 20% from day to day, as indicated by iodine visualization.

spectively. There was insufficient monkey plasma available to determine its water content; therefore, the approximate value of 0.935 was used in the calculations.

Determination of Plasma Water—Six samples each of guinea pig and pooled mouse plasma were weighed in tared vials and dried at 100–110° to constant weight. The loss in weight upon drying was taken as the water content and expressed as the fraction of total plasma weight.

Dialysis of Plasma *versus* Ultrafiltrate of Plasma—During dialysis of plasma against a buffer such as the one used in this study, it is likely that the plasma concentrations of various small, diffusible molecules and ions are altered. In some cases, this alteration might change the binding of the compound being studied. To investigate this possibility, dog (foxhound) plasma was dialyzed against an ultrafiltrate of the same plasma using the following procedure.

Plasma (5 ml) was added to each of two membrane filter cones[12]; these cones had been soaked overnight in distilled water. The cones were drained and blotted before the addition of plasma. The filter cones were then placed in polycarbonate centrifuge tubes and centrifuged (2000×*g*) for 5 min to remove water trapped in the walls of the filters. After transferring the cones to dry centrifuge tubes, centrifugation was continued for 1 hr.

At the end of this time, about 4 ml of ultrafiltrate was collected from each tube. Since these filter cones retain substances with molecular weights greater than 50,000 daltons, the ultrafiltrate should be free of most plasma proteins but still contain the smaller plasma solutes. The dialysis of plasma *versus* plasma ultrafiltrate containing naltrexone was carried out as previously described. Controls, plasma *versus* buffer containing naltrexone, were dialyzed simultaneously. The initial concentration of 15,16-^{3}H-naltrexone in buffer or ultrafiltrate was 2 ng/ml.

Distribution of Radioactivity in Mouse Tissues after Intravenous Administration of 8-^{3}H-Naltrexone—Male, albino, CFl mice[13], 22–32 g, were injected *via* the tail vein with 8-^{3}H-naltrexone[14] and nonradiolabeled compound, 0.925 mg (40 μCi)/kg. A 50-μl syringe with 1-μl graduations[15] was used for drug administration. The injection solution was prepared by dissolving 17.4 μg (20 μCi) of 8-^{3}H-naltrexone and 445 μg of nonradiolabeled compound in 0.5 ml of phosphate-buffered saline to give a final concentration of 925 μg (40 μCi)/ml. This solution was freshly prepared on the day of an experiment.

The mice were sacrificed by decapitation at either 1, 5, or 15 min after the intravenous injection. Blood was collected in a heparinized beaker, and plasma and red cells were separated in a hematocrit tube by centrifugation. The brain, lungs, heart, spleen, liver, and kidneys were removed and placed in tared combustion cones[16] containing cellulose powder. Organs too large to be placed in one cone were divided and placed in as many cones as necessary. Samples of fat, muscle, skin, red cells, and plasma were also added to similarly prepared combustion cones, as was the portion of the tail containing the injection site.

The GI tract (stomach and small and large intestines) was homogenized[17] in 2 ml of water. Similarly, a homogenate of the carcass, predominantly muscle and bone, was prepared using 35 ml of water. Samples of GI and carcass homogenates were weighed into combustion cones. All organ, tissue, and homogenate samples were air dried a minimum of 16 hr before being oxidized[18]. During the combustion procedure, the tritiated water resulting from the oxidation was collected in liquid scintillation counting vials and 15 ml of scintillation liquid (naphthalene, 100 g; 2,5-diphenyloxazole, 5 g; 1,4-bis[2-(4-methyl-5-phenyloxazolyl)]benzene, 0.3 g; dioxane, 720 ml; toluene, 180 ml; and absolute methanol, 35 ml) was subsequently added. The samples were assayed by liquid scintillation counting as previously described.

The amount of radioactivity administered was determined by a similar assay of the tritiated water produced by oxidation of aliquots of a 1:1000 dilution of the injection solution. The percentage of administered radioactivity in each organ or tissue was then calculated. The total recovery of radioactivity in all tissue and organ samples plus the carcass was essentially complete, 97.9 ± 7.9% of the dose.

Table II—Binding of Naltrexone in Monkey, Guinea Pig, Rat, and Mouse Plasma over a Range of Relevant Concentrations[a]

Mean Assayed Total Concentration of Naltrexone in Plasma, ng/ml	Percentage Bound[b]	Mean Assayed Total Concentration of Naltrexone in Plasma, ng/ml	Percentage Bound[b]
Monkey		Guinea Pig	
0.108	18.9 ± 2.49	0.104	20.4 ± 2.78
7.73	21.3 ± 2.86	7.50	20.9 ± 2.43
527	20.7 ± 0.23	512	21.9 ± 4.46
Rat[c]		Mouse[d]	
0.0995	19.8 ± 2.3	0.104	23.1 ± 1.56
7.80	19.9 ± 1.35	7.80	21.9 ± 2.25
514	19.4 ± 1.82	517	23.3 ± 1.20

[a] Measurements made at 37° using equilibrium dialysis. [b] Mean ± *SD*; n = 6 for guinea pigs and rats, and n = 3 for monkeys and mice; n is the number of plasma samples, each from a different animal. [c] Each sample obtained by pooling plasma from two rats. [d] Each sample obtained by pooling plasma from 10 mice.

RESULTS

Purity of 8-^{3}H- and 15,16-^{3}H-Naltrexone—The R_f values of authentic naltrexone chromatographed in three solvent systems are presented in Table I together with the percentage of radioactivity corresponding to those R_f values. It is evident that the purities of 8-^{3}H-naltrexone and 15,16-^{3}H-naltrexone, as measured by this technique, are greater than 95 and 97%, respectively.

Dialysis of Plasma *versus* Plasma Ultrafiltrate—The percentages bound in dog plasma dialyzed against the ultrafiltrate and dog plasma dialyzed against 0.16 *M* phosphate buffer are 19.1 ± 2.4 and 21.3 ± 3.3, respectively, for seven determinations. There is no significant difference between these values (t test, $p > 0.05$), indicating that any diffusion of small molecules between plasma and phosphate buffer does not significantly alter the extent of naltrexone binding in plasma.

Concentration Studies—In Fig. 1, the relationship between percentage bound and plasma naltrexone concentration for three human subjects and two mongrel dogs is shown. The lines in Fig. 1 are the least-squares regression lines of best fit to the data points on a linear scale. (In Fig. 1, a log scale is used on the abscissa for ease of presentation.) For the human subjects, the slope of this line is 0.008 ± 0.003, which is significantly different from zero (t test, $p < 0.05$). However, when the data for each subject are fit individually, only the slope obtained with data from Subject TL is significantly different than zero.

The data obtained using plasma from two mongrel dogs give a line of best fit having a slope of 0.002 ± 0.005. This value, as well

[12] Type CF50A, Amicon Corp., Lexington, Mass.
[13] Carworth Farms.
[14] This form of tritiated naltrexone, having a lower specific activity, was the only radiolabeled material available at the time of the experiment.
[15] Hamilton Co., Reno, Nev.
[16] Packard Instrument Co., Downers Grove, Ill.
[17] Sorvall Omni-Mixer, Ivan Sorvall Inc., Norwalk, Conn.
[18] Tritium oxidizer model 300, Packard Instrument Co., Downers Grove, Ill.

Table III—Binding of Naltrexone in Plasma Obtained from Humans, Monkeys, Dogs, Guinea Pigs, Rats, and Mice

Species	n	Mean Assayed Total Concentration of Naltrexone in Plasma, ng/ml	Percentage Bound[a]
Human[b]	3	13.5	20.7 ± 0.47
Monkey[b]	3	7.73	21.3 ± 2.86
Dog[b]			
Mongrels	2	13.8	26.2
Beagles	14	13.3	26.8 ± 2.12
Foxhounds	2	12.7	20.2
Guinea pig[b]	6	7.50	20.9 ± 2.43
Rat[c]	6	7.80	19.9 ± 1.35
Mouse[d]	3	7.80	21.9 ± 2.25

[a] Mean ± SD. [b] n is the number of plasma samples, each from a different subject or animal. [c] n is the number of samples, each obtained by pooling plasma from two rats. [d] n is the number of samples, each obtained by pooling plasma from 10 mice.

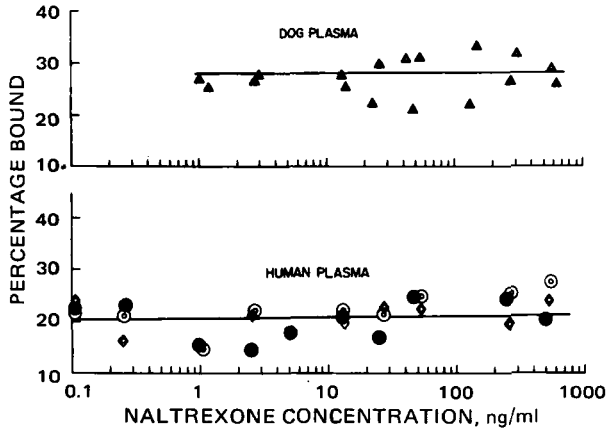

Figure 1—*Effect of concentration of naltrexone on the percentage of naltrexone bound in dog and human plasma at 37°. Key: ▲, Dog GS; △, Dog PS; ●, Human TM; ⊙, Human TL; and ◆, Human RR.*

as the values obtained when the data from each dog are fit individually, does not differ significantly from zero ($p > 0.05$). In any case, the very low values of these slopes indicate that the concentration of naltrexone has negligible influence on the percentage bound in human and dog plasma.

Similar results were obtained for the monkey, guinea pig, rat, and mouse. In Table II, the percentage bound in monkey, guinea pig, rat, and mouse plasma at low, intermediate, and high plasma concentrations is shown. These data clearly demonstrate that the plasma binding of naltrexone in these species is independent of naltrexone concentration over the large concentration range studied.

Species Comparisons—The percentage of naltrexone in plasma that is bound at comparable drug concentrations is given in Table III for the six species studied. None of the percentage bound values differs appreciably from one another.

Distribution of Radioactivity in the Mouse—The results of the distribution study in mice are presented in Table IV and Fig. 2. At 1 min after injection (injection time was 5–10 sec), less than 4% of the administered radioactivity remains in plasma. At 1–5 min postinjection, the radioactivity is distributed in rough proportion to tissue wet weight (except skin, kidneys, and lungs). However, at 15 min, 46% of the dose is localized in the organs of elimination, kidneys, liver, and GI tract, which represent only 20.3% of the body weight.

DISCUSSION

The percentage of naltrexone bound did not vary appreciably over the large concentration ranges of about 1–500 ng/ml (2.9×10^{-9} to 1.5×10^{-6} *M*) for dog plasma and of 0.1–500 ng/ml (2.9×10^{-10} to 1.5×10^{-6} *M*) for monkey, guinea pig, rat, mouse, and human plasma. Plasma binding that is independent of drug concentration also was reported for the basic drugs amphetamine (8), codeine (12), morphine (12, 13), pentazocine (14), and desipramine (15). The binding of methadone (16), quinidine (17), nicotine (18), propranolol (19), and the benzodiazepines (20) to plasma or albumin has been characterized as concentration dependent.

The binding of the adrenergic β-receptor antagonist alprenolol to human serum is concentration dependent while the binding to human serum albumin is not (21). This difference underlines the importance of using serum or plasma instead of purified protein fractions if the results of binding studies are to simulate conditions *in vivo*. There does not appear to be any relationship between the extent of binding of these basic drugs and whether or not the binding is concentration dependent. Thus, it is important to obtain binding data for basic drugs in several species so that this factor can be better understood. No significant species differences were noted in the binding of naltrexone in human, monkey, dog, guinea pig, rat, and mouse plasma. However, species differences have been found for other basic drugs such as amphetamine (9), codeine (12), morphine (12), propranolol (19), and desipramine (22).

Results of preliminary studies concerning the pharmacokinetics of naltrexone in dogs (foxhounds) indicate that this compound has a relatively large apparent volume of distribution (3.2–3.6 liters/kg) (23). This finding is consistent with the low extent of plasma binding observed in the present study; *i.e.*, most of the drug in plasma is free to cross capillary membranes readily and to accumulate in tissues.

The results of the present study concerning the pharmacokinetics of naltrexone in mouse tissues at 1, 5, and 15 min after intrave-

Table IV—Distribution of Radioactivity in the Mouse after Intravenous Administration of 8-^{3}H-Naltrexone, 1 mg/kg

Tissue	Percentage of Body Weight[a]	Concentration, μg Naltrexone Equivalent per g Wet Weight[b]			Percentage of Dose[b]		
		1 min	5 min	15 min	1 min	5 min	15 min
Brain	1.49 (0.25)	0.568 (0.069)	0.803 (0.063)	0.508 (0.233)	0.67 (0.12)	1.43 (0.18)	0.89 (0.47)
Lung	0.70 (0.14)	3.31 (0.87)	1.25 (0.19)	0.980 (0.175)	2.08 (0.85)	0.99 (0.12)	0.80 (0.13)
Heart	0.49 (0.09)	1.66 (0.37)	0.683 (0.060)	0.516 (0.051)	0.91 (0.20)	0.36 (0.04)	0.29 (0.07)
Spleen	0.35 (0.09)	1.19 (0.36)	1.25 (0.20)	0.883 (0.077)	0.34 (0.16)	0.44 (0.08)	0.38 (0.18)
Liver	5.50 (0.91)	1.40 (0.09)	3.26 (0.20)	3.81 (0.65)	6.27 (1.00)	20.6 (2.9)	23.1 (4.4)
Fat	—	0.222 (0.125)	0.349 (0.153)	0.359 (0.145)	—	—	—
Kidney	1.52 (0.11)	5.27 (0.98)	3.47 (0.46)	2.87 (0.34)	7.97 (1.62)	5.66 (1.11)	4.98 (0.66)
GI tract	13.3 (1.9)	0.98 (0.080)	1.06 (0.18)	1.27 (0.20)	11.8 (0.9)	16.9 (2.1)	17.8 (1.2)
Skin	16.6 (0.9)[c]	0.361 (0.074)	0.547 (0.192)	0.739 (0.238)	6.46 (1.30)	10.3 (3.4)	12.8 (4.3)
Muscle	39.9[d]	0.781 (0.280)	0.794 (0.098)	0.459 (0.054)	33.7 (12.4)	39.1 (1.2)	24.7 (2.4)
Plasma	5.1[d]	0.646 (0.179)	0.506 (0.095)	0.931 (0.060)	3.57 (1.02)	2.70 (0.41)	5.69 (0.69)
Red cells	4.98 (1.22)[e]	0.675 (0.062)	0.411 (0.024)	0.246 (0.029)	5.23 (1.97)	1.94 (0.29)	1.44 (0.19)
Injection site	—	—	—	—	7.30 (2.25)	3.79 (2.08)	2.93 (3.18)
Total					86.3	104.2	95.8

[a] Mean of 12 mice (except where noted), standard deviation in parenthesis. [b] n = 3. [c] n = 8. [d] From Ref. 11. [e] Calculated from the hematocrit (Hct) as follows: red cells, percent of body weight = 5.1% [Hct/(1 − Hct)].

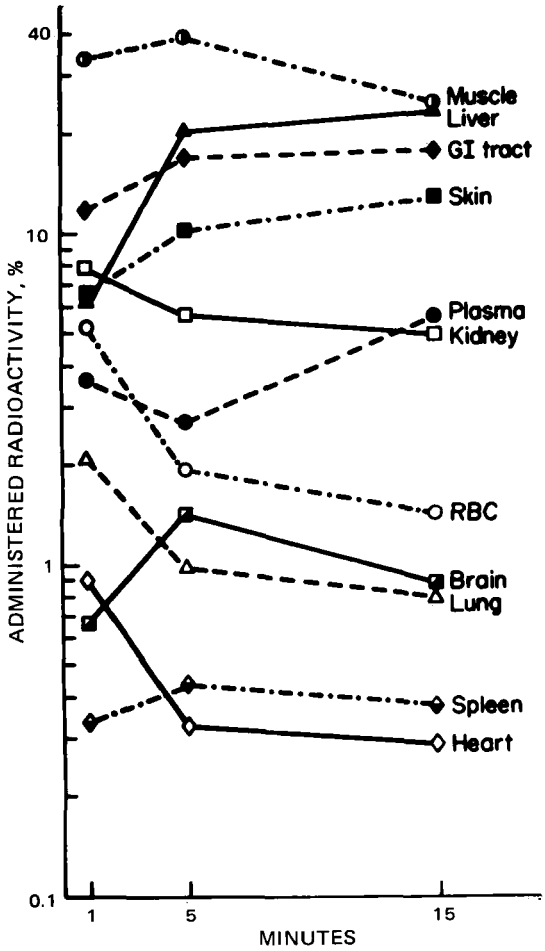

Figure 2—*Distribution of radioactivity in the mouse as a function of time after intravenous injection of 8-^{3}H-naltrexone. Each point represents the mean of three values.*

nous administration further illustrate the rapid and extensive distribution from plasma to body tissues; less than 4% of the administered radioactivity remains in plasma after 1 min. The amount of radioactivity in the brain reaches a maximum at 5 min (Fig. 2). A similar early peak in brain levels was reported for pentazocine (24).

The ratio of radioactivity concentration (expressed as micrograms of naltrexone equivalents per gram wet weight of tissue or fluid) in plasma to that in the red cell fraction increases with time (0.9 at 1 min, 1.2 at 5 min, and 3.8 at 15 min). This fact, plus the absolute increase in plasma radioactivity at 15 min, suggests that significant amounts of radiolabeled metabolites may be accumulating in plasma and that these are too polar to penetrate red cells readily. Thus, the distribution pattern, particularly at 15 min, may not be that of only unaltered naltrexone.

The low extent of binding suggests that potential interactions involving displacement from binding sites will not be a therapeutic problem with naltrexone (25). In addition, the concentration independence of binding over a wide concentration range indicates that this factor can be considered a constant in the interpretation of naltrexone pharmacokinetics.

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ACKNOWLEDGMENTS AND ADDRESSES

Received March 28, 1975, from the *College of Pharmacy and the ‡College of Veterinary Medicine, Ohio State University, Columbus, OH 43210

Accepted for publication July 28, 1975.

Supported by Contract HSM 42-73-182 and Grant DA-00473 from the National Institute on Drug Abuse (NIDA), U.S. Department of Health, Education and Welfare, Rockville, Md.

The authors are indebted to Dr. J. A. Kepler for providing the tritiated naltrexone under NIDA Contract HSM 42-73-184 with Research Triangle Institute, Research Triangle Park, N.C., and to Dr. D. A. McCarthy, Parke-Davis & Co., Ann Arbor, Mich., for providing samples of monkey plasma.

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