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Studies on the Absorption of practically Water-insoluble Drugs following Injection. IV.[1] An Approach for Predicting Relative Intramuscular Absorption Rates of a Drug in Oily Solution, Aqueous Suspension and Aqueous Surfactant Solution in Rats

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This investigation was undertaken to evaluate the differences of intramuscular absorption rates of practically water-insoluble drugs in rats among the three basic dosage forms: oily solutions (including oily suspensions), aqueous suspensions, and aqueous solutions containing nonionic surfactants as solubilizers. A review of previous studies indicated that the rank order of magnitude of the absorption rates among such dosage forms was not fixed, but varied remarkably depending on the drug, the oily solvent, the surfactant and its concentration, and colloidal properties such as particle size, *etc.*, in addition to the initial drug concentration and injection volume. Thus, we sought to establish a method for predicting the relative absorption rates from the three dosage forms empirically. The feasibility of the proposed approach was confirmed experimentally with testosterone for the three dosage forms injected intramuscularly into intact rats. In conclusion, this approach is expected to provide a novel and useful guide not only for predicting the relative absorption rates from various dosage forms but also for selecting optimal preparations for more detailed screening and preclinical testing in laboratory animals of new drugs under development.

Keywords——practically water-insoluble drug; intramuscular injection; intramuscular absorption kinetics; comparison of absorption rate among dosage forms; oily solution; aqueous suspension; surfactant solution; prediction of relative absorption rate; rat

Parenteral drug administrations are as commonly used as oral ones for testing of drugs in laboratory animals.[2] When a new compound under development is practically water-insoluble, selection of optimal dosage forms in the early stages of testing in animals is one of the most important considerations, though it is a very difficult problem.

Practically water-insoluble drugs are commonly administered to laboratory animals as parenteral preparations in the form of oily solutions, aqueous or oily suspensions, aqueous solutions with nonionic surfactants or other solubilizers, or emulsions.[3] The absorption of these drugs administered by any extravascular route depends on the dosage form or physicochemical state in the preparation, and is in most cases slow enough to be the rate-determining step in their disposition in the body. This means that even the potency and duration of pharmacological activities may often be primarily governed by the formulation itself.

Recently, we have clarified the absorption kinetics of such drugs in water-immiscible oil solutions and suspensions,[4] aqueous suspensions,[5] and aqueous solutions solubilized with nonionic surfactants[1] injected intramuscularly into intact rats. Although the resultant kinetic equations were derived empirically, they were expected to have great practical value and to provide some basis for comparing intramuscular drug absorption rates among such dosage forms.

The purposes of this study were to clarify the differences of the intramuscular absorption kinetics of practically water-insoluble drugs among the above dosage forms and to find a possible approach for predicting the relative absorption rates by reviewing previous results.[1,4,5] The feasibility of the proposed approach was checked experimentally with testosterone prepar-

ations. The present work provides a novel and useful guide for the selection of appropriate dosage forms and should lead to improvements in the screening and preclinical testing in laboratory animals of new drugs under development.

Experimental

Materials——Azo dyes (such as *p*-aminoazobenzene, *p*-hydroxyazobenzene and *o*-aminoazotoluene) and steroids [such as 2α,3α-epithio-5α-androstan-17β-ol (epitiostanol) and testosterone] were used as model compounds for practically water-insoluble drugs. These were the same as the materials used in a previous study.[4] Sesame oil, methylcellulose, polysorbate 80, and HCO-40 were also the same as those used in our previous studies.[1,4,5]

Test Preparations——Test oily solutions were prepared by using sesame oil according to the method described in a previous paper.[4] Test aqueous suspensions were formulated in a disperse medium containing 0.5% (w/v) methylcellulose, 0.005% (w/v) polysorbate 80 and 0.9% (w/v) NaCl, according to the controlled preparation method described previously.[5] Hereafter in this paper, the suspensions prepared by this method will be referred to as 'controlled' suspensions. Test aqueous surfactant solutions were prepared with HCO-40 as a solubilizer according to the method employed in the previous study (each solution contained 0.9% (w/v) NaCl).[1] In this investigation, absorption data for two test preparations, the 'controlled' aqueous suspension and aqueous surfactant solution (10% HCO-40) of testosterone, were determined and those for others are cited from our previous studies.[1,4,5] The colloidal properties of the 'controlled' aqueous suspension of testosterone tested here were as follows: mean particle diameter (D_{ss}), 3.6 μm; distribution constant (n), 2.7; sedimentation volume (V_{sed}), 2.4 cm^3/g. These properties were similar to those of 'controlled' suspensions used previously.[5]

Absorption Experiment Procedure——Male Wistar albino rats weighing 240 to 280 g were used in all absorption experiments. All test preparations were injected into the center region of the *m. gastrocnemius* of the left hind leg and the drug absorption rates were determined by the local clearance method. Rats were kept intact during the absorption experiment. The details of this absorption experiment procedure were given previously.[4]

Determination of Physicochemical Properties——The solubility (at 37°) in 0.9% (w/v) NaCl aqueous solution (saline) and the crystal density of testosterone were measured as described elsewhere.[5] For other test compounds, previously determined values[5] were used. The distribution coefficient (at 37°) of testosterone between the micellar and water phases was measured according to a reported method.[1] Other data for distribution coefficients (at 37°) between sesame oil and saline or between the micellar and water phases are cited from previous papers.[1,4]

Analytical Method——Sample solutions of testosterone in absorption experiments were assayed by the GLC method described previously.[4] Testosterone in saline was analyzed by the spectrophotometric method reported elsewhere[4] in order to determine its solubility and distribution coefficient between the micellar and water phases.

Results and Discussion

Comparison of Kinetic Processes of Drug Absorption among Oily Solutions (Including Oily Suspensions), Aqueous Suspensions and Surfactant Solutions

Recently, we have studied in detail, using the local clearance method, the kinetic processes of intramuscular absorption of practically water-insoluble drugs from three conventional dosage forms, (i) water-immiscible oil solutions (including oily suspensions), (ii) aqueous suspensions and (iii) aqueous solutions containing nonionic surfactants as solubilizers, in intact rats. Table I summarizes the resultant equations describing the absorption kinetics, that is, representing the relationship between the residual fraction (W/W_0) of the drug in the injection site and time (t) for each dosage form. The details of these equations were given in our previous papers.[1,4,5] The kinetic process for dosage forms (i) (except for the oily suspensions) and (iii) was of first order, differing from that for dosage form (ii).

The main physicochemical factors influencing the drug absorption from dosage form (i) were the distribution coefficient (K_1, oil phase/saline or buffer phase) of the drug and the injection volume (V_{01}) [in addition, the ratio (p) of the initial drug concentration (C_{01}') to the solubility of the drug in the oil ($C_{s,oil}$) for oily suspensions]; those from dosage form (ii) were the solubility of the drug in saline (C_s), the density of the drug crystals (ρ), the initial drug

TABLE I. Kinetic Equations for Intramuscular Absorption of practically Water-insoluble Drugs from Three Dosage Forms[a]

(i) Water-immiscible oil solution and suspension		(ii) Aqueous suspension		(iii) Aqueous surfactant solution	
Solution ($C_{01}<C_{s,oil}$)		$(W/W_0)^{1/3}=1-\alpha_2 t$	(Eq. 2a)	$\ln (W/W_0)=-\alpha_3 t$	(Eq. 3a)
$\ln (W/W_0)=-\alpha_1 t$	(Eq. 1a)	$\alpha_2=\beta_2\gamma_f C_s^q \rho^r C_{02}^g V_{02}^h$	(Eq. 2b)	$\alpha_3=\beta_3 K_3^{-1} C_{s0}^{-1} V_{03}^u (+\varepsilon_m)$	(Eq. 3b)
$\alpha_1=\beta_1 K_1^l V_{01}^m$	(Eq. 1b)	$\tau_2=0.206/\alpha_2$	(Eq. 2c)	$\tau_3=0.693/\alpha_3$	(Eq. 3c)
$\tau_1=0.693/\alpha_1$	(Eq. 1c)	q: 0.52, r: −0.45, g: −0.55, h: −0.32		(u: −0.33)	
(l: −0.85, m: −0.14(∼−0.32))					
Suspension ($C_{01}'>C_{s,oil}$, $V_{01}'=V_{01}$)					
$W/W_0=1-\alpha_1' t$ (t, small)	(Eq. 1d)[b]				
$\alpha_1'=f\alpha_1/p$	(Eq. 1e)				
$\tau_1'=0.5/\alpha_1'$	(Eq. 1f)				
(f: 0.5)[c]					

a) Symbols appearing in this table and the text are as follows: W_0, dose; W, drug amount remaining in the injection site at time t; α (α'), absorption rate constant: β, hybrid parameter containing *in vivo* drug diffusibility and dosage form factors; V_0 (V_0'), injection volume; C_0 (C_0'), initial drug concentration; τ (τ') absorption half-life; subscripts 1, 2 and 3 of α (α'), β, V_0 (V_0'), C_0 (C_0') and τ (τ') denote the dosage forms (i), (ii) and (iii), respectively; $C_{s,oil}$, solubility of drug in oil (37°); $p=C_{01}'/C_{s,oil}$; K_1, distribution coefficient of a drug between oil and saline or pH 7.25 isotonic buffer (37°); γ_f, unknown function depending on formulation conditions such as particle size, etc. (this equals unity for the 'controlled' suspension); ρ, density of drug crystals; K_3, distribution coefficient of a drug between the micellar and water phases (37°); C_{s0}, initial surfactant concentration; ε_m, correction term for the contribution of micelles containing the drug to absorption (this is negligible for low C_{s0}). l, m, g, h, q, r and u are constants estimated from experimental results obtained after administration into the *m. gastrocnemius* in intact rats. Detailed explanations of these kinetic equations were given in previous reports.[1,4,5]

b) Approximate equation describing the absorption process during a short period after injection.

c) Value determined experimentally under the following conditions: particle size, 37—149 μm; initial solid drug concentration in suspension, 16.7—74.7 mg/ml.

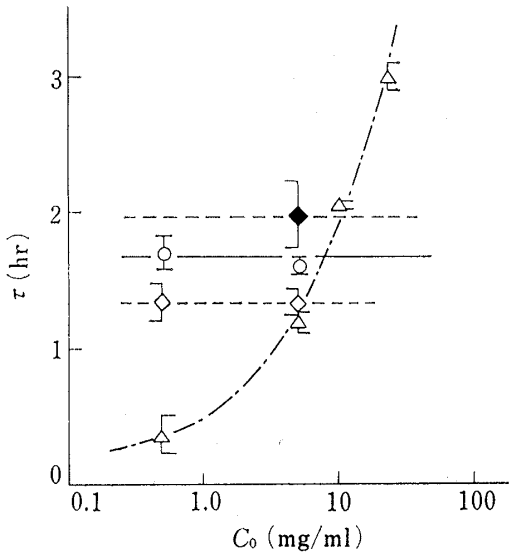

Fig. 1. Comparison of Absorption Half-lives (τ) of *p*-Hydroxyazobenzene among Three Dosage Forms at Various Initial Drug Concentrations (C_0) following Injection into the *M. Gastrocnemius* in Intact Rats

Key; —○—, sesame oil solution; -·-△-·-, 'controlled' aqueous suspension [D_{ss} (n), 4.1 μm (2.5)]; --◇--, 10% (w/v) HCO-40 aqueous solution; --◆--, 20% (w/v) HCO-40 aqueous solution. Data plotted here are cited from previous reports.[1,4,5] The vertical bar shows the standard error.

concentration (C_{02}), the injection volume (V_{02}) and preparation conditions (such as particle size, *etc.*); those from dosage form (iii) were the distribution coefficient (K_3, micellar phase/water phase) of the drug, the initial surfactant concentration (C_{s0}) and the injection volume (V_{03}). The equations representing the correlation between the absorption rate constant α_1 (α_1'), α_2 or α_3 and these main factors for each dosage form, which were obtained empirically, are presented in Table I together with the experimentally determined parameters (l, m, f, g, h, q, r and u).

Comparison of these equations indicates that the drug absorption rate depends considerably on the preparation and administration conditions even for the same dosage form, and hence that the rank order of magnitude of the rates among the three dosage forms is not always fixed. The rate constant for aqueous suspensions (α_2) increases with decreasing initial drug concentration (C_{02}), as indicated by Eq. 2b in this table, while those (α_1 and α_3) for the other two solutions are independent of the initial drug concentration. A reversal in the rank order of magnitude of the absorption rates is, therefore, expected to take

place at some value of the initial drug concentration.

Figure 1 shows this phenomenon using experimental data for *p*-hydroxyazobenzene in sesame oil solution, 'controlled' aqueous suspension and HCO-40 aqueous solution. In this figure, absorption half-lives (τ), in place of absorption rate constants (α), are plotted against C_0 since the forms of the kinetic equations for these dosage forms are not all identical. As expected, a reversal in the rank order of the absorption rates occurred: the absorption half-life for the 'controlled' aqueous suspension was smaller than that for the other two preparations in the region below a certain C_0 value.

Figure 1 also indicates that the absorption rate from aqueous solution of high surfactant concentration (20% HCO-40) was smaller than that from sesame oil solution. This example illustrates that the absorption of practically water-insoluble drugs from aqueous surfactant solutions is not always faster than that from the other two dosage forms. It should be noted that the rank order of magnitude of the absorption rates among these dosage forms is remarkably variable, depending on the drug, the oily solvent, the surfactant and its concentration, and colloidal properties such as particle size, *etc.* in addition to the initial drug concentration and injection volume.

Parameters β_1, β_2 and β_3 and Their Ratios

When profiles similar to those in Fig. 1 are available, selection of the optimal dosage form for screening or testing of drugs in laboratory animals may be easier. However, the construction of these profiles for each compound through *in vivo* experiments requires too much time. The hybrid parameters β_1, β_2 and β_3 in Eqs. 1b. 2b and 3b shown in Table I include a factor related to the diffusibility of the drug into the inter- or intracellular space of the muscle, connective and vascular tissues in addition to a factor related to the geometry of the depot and other unknown factors by the dosage form itself. If the ratios of these parameters, β_2/β_1 and β_3/β_1, depend little on the compound, the rank order of magnitude of the intramuscular absorption rates among the three dosage forms (water-immiscible oil solutions, 'controlled' aqueous suspensions and nonionic surfactant solutions) can roughly be predicted from physicochemical parameters such as K_1, C_s, ρ and K_3 determined by simple *in vitro* experiments.

Table II lists the values of β_1, β_2 and β_3 together with their ratios β_2/β_1 and β_3/β_1 for four test compounds. These values were calculated from Eqs. 1b, 2b and 3b in Table I using the α values and physicochemical parameters determined in our previous studies.[1,4,5] Details of this calculation are explained in the footnotes to Table II. The results indicate that each ratio was relatively constant and that the values were in the same order for a variety of compounds: β_2/β_1 ranged from 0.0030 to 0.0099 and β_3/β_1 from 0.77 to 1.61 . The small variation of these ratios suggests the possibility of a rough prediction of relative absorption rates among

Table II. List of Experimentally Determined Parameters β_1, β_2 and β_3, and Their Ratios β_2/β_1 and β_3/β_1 for Four Test Compounds

Compound	Parameter			Ratio	
	β_1[a]	β_2[b]	β_3[c]	β_2/β_1[d]	β_3/β_1[e]
p-Aminoazobenzene	89.6	0.89	69.1	0.0099	0.77
p-Hydroxyazobenzene	125.7	1.07	99.9	0.0085	0.80
o-Aminoazotoluene	164.1	1.25	263.5	0.0076	1.61
Epitiostanol	138.3	0.42	150.0	0.0030	1.09

a) $[hr^{-1}\cdot ml^{0.14}]$. Calculated from Eq. 1b using previous data for α_1 (at $V_{01}=0.05$ ml) and K_{11} for each compound in sesame oil solution.[4]
b) $[hr^{-1}\cdot mg^{0.48}\cdot ml^{-0.16}]$. Calculated from Eq. 2b setting $\gamma_f=1$ and using previous data for α_2 (at $C_{02}=5$ mg/ml and $V_{02}=0.05$ ml), ρ and C_s for each compound in 'controlled' suspension.[5]
c) $[hr^{-1}\cdot ml^{0.33}]$. Calculated from Eq. 3b setting $\varepsilon_m=0$ and using previous data for α_3 (at $V_{03}=0.05$ ml) and K_3 for each compound in 10% (w/v) HCO-40.[1]
d) $[mg^{0.48}\cdot ml^{-0.30}]$.
e) $[ml^{0.19}]$.

such dosage forms.

Prediction of Relative Absorption Half-lives for Three Dosage Forms

The foregoing results suggest that rough prediction may be possible as follows. When the absorption half-life of a drug for one dosage form *e.g.* oily solution, τ_1, is known, that for the other two dosage forms, τ_2 and τ_3, can be approximately predicted on the basis of physicochemical parameters determined by *in vitro* experiments and the reciprocals of the ratios β_2/β_1 and β_3/β_1 by using the following equations. For 'controlled' aqueous suspension,

$$\tau_2 = \frac{0.206}{0.693} \times \frac{\beta_1}{\beta_2} \times \frac{K_1{}^l V_{01}{}^m}{C_s{}^q \rho^r C_{02}{}^g V_{02}{}^h} \times \tau_1 \qquad \text{(Eq. 4)}$$

(β_1/β_2: 101—333)

and for aqueous surfactant solution,

$$\tau_3 = \frac{\beta_1}{\beta_3} \times \frac{K_1{}^l V_{01}{}^m}{K_3{}^{-1} C_{s0}{}^{-1} V_{03}{}^u} \times \tau_1 \qquad \text{(Eq. 5)}$$

(β_1/β_3: 0.62—1.30)

Even if τ_1 is not known, the relative magnitude of the absorption half-life can be easily estimated from these tentative equations.

To check the feasibility of this approach, the absorption half-lives of another compound, testosterone, injected intramuscularly into the *m. gastrocnemius* in intact rats were compared among three dosage forms, sesame oil solution, 'controlled' aqueous suspension and aqueous nonionic surfactant solution (10% HCO-40). The absorption half-life for testosterone in sesame oil solution had been measured in a previous study.[4] On the basis of this value, the absorption half-lives for the other two preparations were estimated from Eqs. 4 and 5. These estimated values are shown in Table III and the parameters used for this prediction are given in the footnotes. The prediction was that the absorption half-life would increase in the order of the aqueous surfactant solution, the oil solution and the 'controlled' aqueous suspension under the conditions used (C_0, 5 mg/ml; V_0, 0.05 ml). Figure 2 shows the experimental absorption time curves for such preparations under the same conditions. The observed absorption half-lives obtained from this figure are also listed in Table III for comparison with the predicted values. The observed half-lives for the 'controlled' aqueous suspension and the aqueous surfactant solution were within the ranges predicted. These satisfactory results suggest that the ratios β_2/β_1 and β_3/β_1 have practical value for finding the rank order of magnitude of the absorption rates of a drug injected intramuscularly among the dosage forms presented here. In conclusion, the proposed approach also seems to be applicable to other practically water-insoluble drugs in such dosage forms, provided that the conditions discussed in connection with the kinetic equations shown in Table I hold.

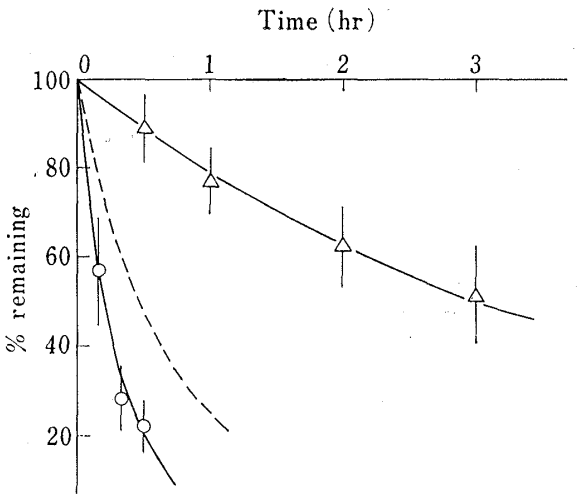

Fig. 2. Absorption Time Curves of Testosterone from the Three Dosage Forms following Injection into the *M. Gastrocnemius* in Intact Rats

Key; —△—, 'controlled' aqueous suspension; —○—, 10% (w/v) HCO-40 aqueous solution; broken line, sesame oil solution (cited from a previous paper[4]). V_0, 0.05 ml; C_0, 5 mg/ml (2.5 mg/ml for 10% HCO-40 aqueous solution). Each data point represents the mean of 4 experiments and the vertical bar indicates the standard deviation.

In this investigation, we mainly treated simple systems. However, these basic systems are related to other more complex ones. Accordingly, we believe that this fundamen-

Table III. Predicted and Observed Absorption Half-lives (τ) for Three Testosterone Preparations following Intramuscular Injection into Intact Rats[a]

Dosage Form	τ (hr)		
	Known (S.E.)[b]	Predicted[c]	Observed (S.E.)
(i) Sesame oil solution[d]	0.502(0.047)		
(ii) 'Controlled' aqueous suspension[e]		2.10—6.93	2.91(0.18)
(iii) 10% (w/v) HCO-40[f]		0.13—0.26	0.23(0.04)

a) Injection volume, 0.05 ml.
b) Data reported previously.[4]
c) Predicted from Eqs. 4 and 5 using the following parameters: K_1, 130; C_s, 0.035 mg/ml; ρ, 1.22 g/cm^3; K_3, 0.45 ml/mg; V_{01}, V_{02} and V_{03}, 0.05 ml; C_{02}, 5 mg/ml; C_{s0}, 100 mg/ml; τ_1, 0.502 hr; β_1/β_2, 101—333; β_1/β_3, 0.62—1.30.
d) C_{01}, 5 mg/ml.
e) C_{02}, 5 mg/ml (see 'Experimental' section for colloidal properties).
f) C_{03}, 2.5 mg/ml.

tal approach will provide a novel and useful guide for estimating intramuscular absorption rates from various dosage forms and thus for selecting optimal preparations for detailed screening and testing in laboratory animals of new compounds under development.

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References and Notes

1) Part III: K. Hirano, T. Ichihashi, and H. Yamada, *Chem. Pharm. Bull.*, **29**, 834 (1981).
2) W.A. Ritschel, *Pharm. Ind.*, **35**, 273 (1973).
3) A. Osol (ed.), "Remington's Pharmaceutical Sciences," 15th ed., Mack Publishing Co., Pennsylvania, 1975, pp. 1461—1487, 1631—1643.
4) K. Hirano, T. Ichihashi, and H. Yamada, *Chem. Pharm. Bull.*, **29**, 519 (1981).
5) K. Hirano, T. Ichihashi, and H. Yamada, *Chem. Pharm. Bull.*, **29**, 817 (1981).