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Kinetics of three structurally related bicyclic antidepressant compounds—a comparison in the dog

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Kinetic experiments have been made with three structurally related compounds selected from a series of bicyclic antidepressant compounds. Plasma concentrations were assayed after doses of 5 mg kg^{-1} given as an intravenous infusion over 2 h to three dogs. The data were evaluated according to a two compartment open model, and kinetic parameters reflecting distribution and elimination characteristics of the drugs were calculated. An attempt was made to evaluate possible relations between chemical structure and these parameters. The lowest drug levels and the most rapid elimination were obtained with a compound utilizing sulphoxidation as an additional pathway of metabolism. The compounds lacking this possibility had considerably slower elimination kinetics, the less lipophilic being the slowest one. Little individual variation was seen. A relation between increased lipophilicity and increased localization in the peripheral compartment was indicated. However, the importance of other factors, such as binding phenomena within tissues, was also indicated.

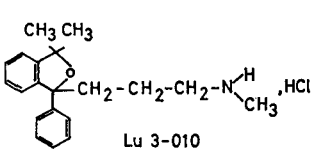
The present publication deals with a series of kinetic experiments in the dog with three structurally closely related compounds selected from a series of bicyclic antidepressant compounds (Petersen, Lassen & others, 1966, 1970). The evaluated parameters are discussed and compared.

METHODS

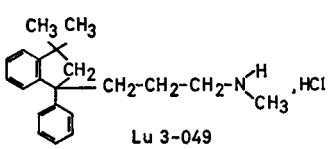
Animals, doses and samples

Three male, purebred beagle dogs of our own stock were given the three compounds Lu 3-010, Lu 3-049 and Lu 5-003 in doses of 5 mg kg^{-1} as an intravenous infusion over 2 h (dog B received only 1 mg kg^{-1} of Lu 5-003). The compounds were given in the following order: Lu 5-003, Lu 3-010, Lu 3-049 (dog B); Lu 5-003, Lu 3-049 and Lu 3-010 (dogs C and D). No drugs were given to the dogs for at least 2 weeks before each experiment. Blood samples were taken into glasses, rinsed with a few drops of heparin, at termination of the infusion (0) and after ½, 1, 2, 3, 4½, 22, 28 and 46 h.

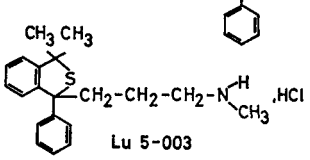
Plasma was obtained by centrifugation and then frozen.



Lu 3-010



Lu 3-049



Lu 5-003

Assay of plasma drug levels

Plasma concentrations of the three compounds were assayed by means of the [^{3}H]-acetic anhydride coupling technique described by Hammer & Brodie (1967), modified by the addition of 0·1 M salicylic aldehyde to the hexane phase (Fredricson Overø, 1972). The specificity of the method was controlled by submitting the remainder of the heptane phases of early (0, 1, 2 or 3 h) as well as late (22 or 28 h) samples to thin-layer chromatography (on silica gel plates in the solvent system benzene–acetone–diethylamine, 80 : 20 : 1), which revealed the presence of one spot only (corresponding to the labelled coupling product of the drug).

All samples were assayed in duplicate together with standards.

Evaluation of the data

Since the plasma concentration data indicated that a two compartment model might be applicable for kinetic evaluation, the best-fitting curves according to such a system were calculated by means of a computer using time/concentration data, the appropriate equation and graphical estimates of α, β and K_2 and an estimate on V_1 for a non-linear least squares curve fitting procedure.

The two compartment model and the equation describing the time-course of drug concentration in the central compartment, as represented by plasma, are shown in Fig. 1. The equation was derived according to Benet (1972).

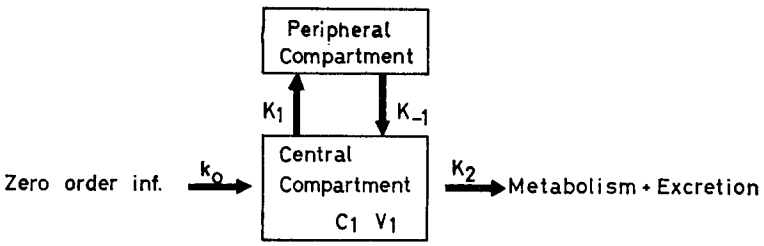
RESULTS AND DISCUSSION

Plasma concentration and parameters

The calculated best-fitting curves were in agreement with the experimental data (Fig. 2) justifying the assumption that a two compartment model may be used for kinetic evaluation of these drugs. The corresponding parameters and their mode of calculation are seen in Table 1. Standard errors on the computer calculated para-

Table 1. *Parameter data according to a two compartment model evaluated from plasma concentrations after intravenous infusion of the compounds Lu* 3-010, *Lu* 3-049, *and Lu* 5-003 *to three beagle dogs.*

Parameter		Mode of calculation	Dog B			Dog C			Dog D		
			3-010	3-049	5-003	3-010	3-049	5-003	3-010	3-049	5-003
α	(h^{-1})	Computer	1·0	2·5	2·9	1·8	1·5	0·79	1·1	2·3	2·1
β	(h^{-1})	,,	0·024	0·026	0·060	0·015	0·031	0·030	0·019	0·031	0·098
V_1	(litre)	,,	29	17	4·5	11	16	19	31	21	13
K_2	(h^{-1})	,,	0·033	0·079	0·95	0·034	0·056	0·071	0·028	0·072	0·30
$V_1 \cdot K_2$	(litre h^{-1})	,,	0·97	1·3	4·3	0·38	0·92	1·3	0·87	1·5	3·8
$t_{\frac{1}{2}}$	(h)	$\frac{0{\cdot}693}{\beta}$	29	26	11·5	45	23	23	36	22	7
K_{-1}	(h^{-1})	$\frac{\alpha \cdot \beta}{K_2}$	0·74	0·84	0·18	0·79	0·80	0·34	0·76	1·0	0·69
K_1	(h^{-1})	$\alpha+\beta-K_2-K_{-1}$	0·26	1·6	1·8	0·95	0·64	0·41	0·35	1·3	1·2
$(Vd)\beta$	(litre)	$V_1 \cdot \frac{K_2}{\beta}$	40	51	71	24	30	44	45	49	39
D_1	(h^{-1})	$\frac{1}{K_2}$	30	13	1·1	29	18	14	−36	−14	3·3
D_2	(h^{-1})	$\frac{K_1}{K_2 \cdot K_{-1}}$	11	24	11	35	14	17	16	18	5·7
D_T	(h^{-1})	$\frac{K_1+K_{-1}}{K_2 \cdot K_{-1}}$	41	37	12	64	32	31	52	32	9·1



$$C_1 = \frac{k_0}{V_1 \cdot K_2}\left[\frac{K_2-\beta}{\alpha-\beta}(1-e^{-\alpha \cdot b})e^{-\alpha(t-b)} - \frac{K_2-\alpha}{\alpha-\beta}(1-e^{-\beta b})e^{-\beta(t-b)}\right]$$

FIG. 1. Two compartment model with zero order infusion and equation describing the time-course of drug concentration in the central compartment, where C_1 = plasma concentration, t = time, b = time when infusion ends, V_1 = proportionality constant relating amount in central compartment to concentration, k_0 = infusion rate, K_2 = first order rate constant for loss of drug from central compartment, K_1 and K_{-1} = first order rate constants for transfer of drug between the compartments,

$$\left\{\begin{matrix}\alpha\\ \beta\end{matrix}\right\} = \tfrac{1}{2}[(K_1 + K_2 + K_{-1})\{\pm\}\sqrt{(K_1 + K_2 + K_{-1})^2 - 4.K_{-1}.K_2}]$$

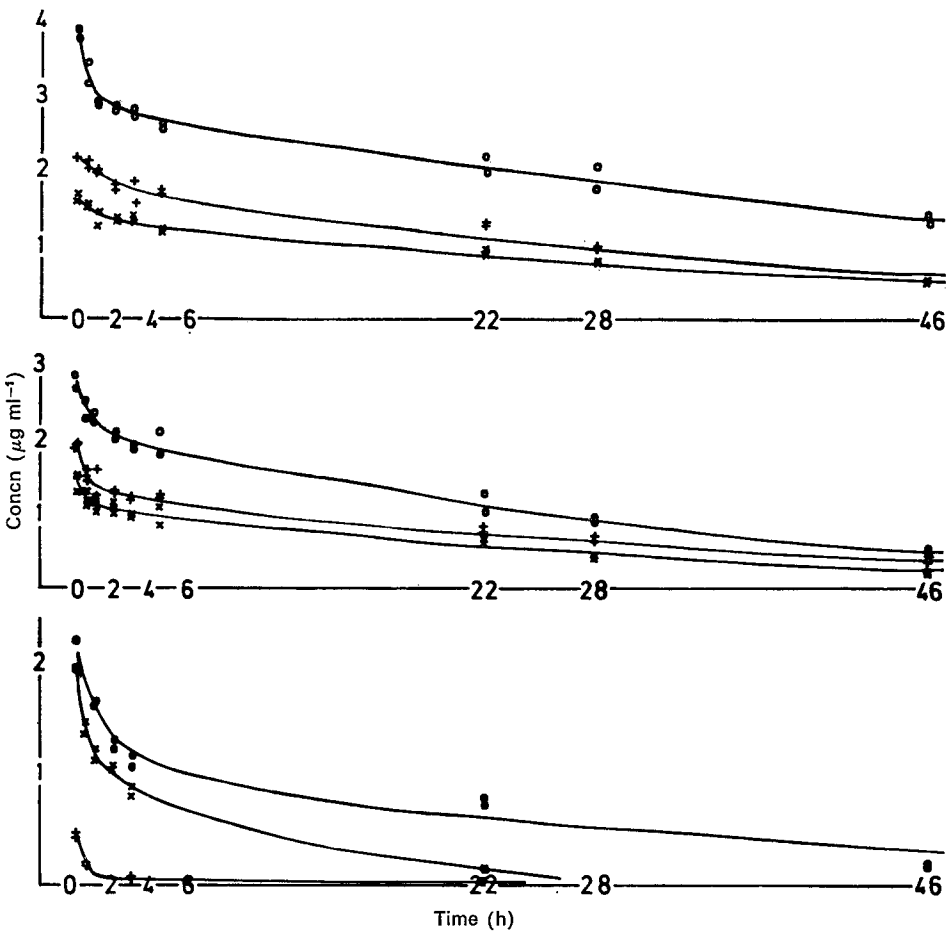


FIG. 2. Plasma concentrations of Lu 3-010 (top) Lu 3-049 (middle) and Lu 5-003 (bottom) after infusion of 5 mg kg^{-1} to dogs B (+), C (o) and D (x).
(For one of the dogs on Lu 5-003 the dose was 1 mg kg^{-1}).

meters for Lu 3-010, Lu 3-049 and Lu 5-003 were in the order of 23–82, 27–50, 14–46% (α); 7–10, 7–9, 17–47% (β); 15–22, 18–30, 18–30% (K_2) and 16–20, 16–29, 12–15% (V_1). There is therefore some error in the remaining parameters which are derived from these.

Elimination kinetics

For the comparison of elimination kinetics of the compounds, three different parameters may be taken into consideration; $t\frac{1}{2}$ (the biologic half-life), K_2 (the rate constant for metabolism and excretion) and $V_1 \cdot K_2$ (total clearance rate, metabolic plus renal). In all dogs Lu 5-003 is seen to have the shortest half-life, the greatest K_2-value and the most rapid clearance. The reason for this faster elimination is probably the utilization of sulphoxidation as an additional route of biotransformation (Fredricson Overø, Jørgensen & Hansen, 1970). Of the other two compounds Lu 3-010 has the longer half-life, the smaller rate constant for metabolism and excretion as well as a lower clearance rate. This might be explained by an additional (unknown) pathway of metabolism for Lu 3-049; Lu 3-010 is known to be metabolized by *N*-demethylation followed by deamination (Plym Forshell, Shauman & others, 1968), while the metabolism of Lu 3-049 has not yet been elucidated. Better access for the more lipophilic compound Lu 3-049 to drug metabolizing enzymes, or greater excretion via bile might be suggested as further explanations. The relative lipophility of Lu 3-010, Lu 3-049 and Lu 5-003 is 1 : 35 : 5 as determined by the partition of the unionized form between water and paraffin according to the method presented by Mercier & Dumont (1969).

With one exception (Lu 5-003 in dog C) individual variation seems to be limited. However, the comparatively low K_2-value for Lu 5-003 as well as the lower total clearance for all three compounds in dog C may indicate that this dog is a slow eliminator.

Distribution kinetics

The distribution kinetics of the compounds may also be illustrated by a series of expressions, the most obvious ones being K_1 and K_{-1} (the first order rate constants for transfer of drug between the compartments). Assuming lipophilicity to be one of the factors determining the rate of transfer of drug into another compartment, one would expect the least lipophilic compound, Lu 3 010, to have the lowest K_1-value and the more lipophilic Lu 3-049 to have a higher one. This is actually the case in dogs B and D. However a high K_1-value is also seen with Lu 5-003, which has intermediate lipophilicity. In addition, a different pattern is seen for dog C. Thus in addition to lipophilicity other factors seem to govern the rate of drug transfer. The K_{-1}-values show little variation as regards Lu 3-010 and Lu 3-049; lower and more varying values are seen for Lu 5-003, as if this compound were withheld in the peripheral compartment, e.g. by particular binding forces.

For the comparison of the distribution patterns of the compounds, however, the parameters K_1 and K_{-1} seem to be less informative than the relation between them. This relation is also expressed by the ratio between the integral coefficients D_1 and D_2 (as introduced by Jusko, Lewis & Dittert, 1972), which, when multiplied by the dose, provides the amount versus time area or integral for the central (D_1) and peripheral (D_2) compartments respectively. Thus the ratio D_1/D_2 (i.e. K_{-1}/K_1) is a direct measure of the ratio between amounts of drug in the two compartments.

In the present experiments this ratio decreases in the order Lu 3-010>Lu 3-049>Lu 5-003, the only exception being the value for Lu 3-010 in dog C. Thus the percentage of drug present in the central compartment (which may be expressed by the relationship between D_1 and D_T) is in two of the dogs highest for Lu 3-010, lower for Lu 3-049 and lowest for Lu 5-003, the percentages being 70–35–10 (dog B) and 70–45–35 (dog D). The corresponding figures for dog C are 45–55–45.

This pattern may be discussed in relation to the factors traditionally considered to be of importance in this context, viz. lipophilicity, pKa and protein binding (Schanker 1961; Rauflaub, 1970; Jørgensen, Hansen & Fredricson Overø, 1973). There is no reason to expect variations in pKa within this series of compounds, neither has the degree of binding to plasma proteins proven to vary much. At a total concentration of 1 μg ml^{-1} approximately 90% of Lu 3-010 and 94% of Lu 5-003 were found to be bound as determined by the ultracentrifugation technique described by Borgå, Azarnoff & others (1969); Lu 3-049 has not yet been investigated. Thus these compounds seem to differ most as regards lipophilicity.

The present results seem to indicate some relation between increased lipophilicity and increased localization in the peripheral compartment. However, since Lu 5-003 has a particularly strong localization to the peripheral compartment other factors are obviously equally important, as one of these binding phenomena related to the S-atom may be suggested. This suggestion is supported by the higher degree of protein binding found for Lu 5-003.

General aspects

The precision involved in the determination of the parameters belonging to the chosen model is of decisive importance for this kind of study. In the present experiments β, K_2 and V_1 were fairly well determined, while more accurate determination of α would have been most valuable, especially since it is used for the calculation of K_1 and K_{-1}.

The error involved is thus partly responsible for the variation seen in parameters. However some individual variation seemingly exists especially as regards Lu 5-003, which is particularly slowly eliminated in dog C and has different distribution kinetics in dog B (though this might be attributed to the lower dose).

Conclusions

We have shown that after equal doses of the three drugs the plasma levels of the parent drugs were higher after Lu 3-010 than after Lu 3-049 and lowest levels were obtained after Lu 5-003. This is illustrated by Fig. 3, in which the total amount of drug in the body has been calculated by multiplying plasma concentrations after attainment of pesudo-distribution equilibrium (i.e. in the mono-exponential β-phase) by the proportionality constant (Vd) β, as described by Gibaldi, Nagashima & Levy, 1969.

The average steady state plasma concentrations in long term toxicity tests on the three drugs may be predicted from the present data by means of the formula of Wagner, Northam & others (1965):

$$\bar{C}_\infty = \frac{F \cdot D}{(Vd)\ \beta \cdot \beta \cdot \tau}$$

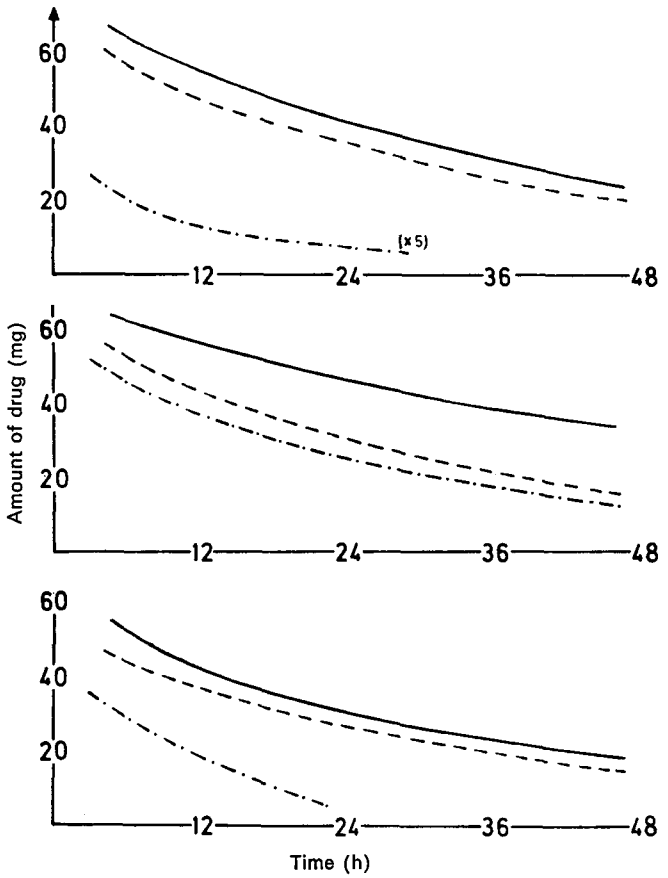


FIG. 3. Amount of drug in the body after attainment of pseudo-distribution equilibrium. Drugs: Lu 3-010 (————), Lu 3-049 (– – –), and Lu 5-003 (–.–.–.–), given as intravenous infusion of 5 mg kg^{-1} during 2 h to dogs B, C and D. (The Lu 5-003 curve for dog B has been multiplied by 5 to compensate for the lower dose.)

where $\bar{C}_{\infty}$ is the average steady state plasma concentration, F is the fraction of the dose (D) absorbed and τ the dosage interval. Assuming complete absorption (F = 1) and using the values on (Vd)β and β obtained from the present experiments (choosing dog D as a typical dog) and a dose of 5 mg kg^{-1} day^{-1}, the following average levels may be estimated: (μg ml^{-1}) Lu 3-010 2·4, Lu 3-049, 4, Lu 5-003 0·5. These figures illustrate the difference in the amount of drug available for uptake by individual tissues. The estimated value for Lu 5-003 corresponds well with practical data (unpublished results), a fact which confirms the utility of the model. (Data for Lu 3-010 and Lu 3-049 are not yet available.)

Certain guides for future drug design can be obtained from the present study. It was indicated that with increasing lipophilicity an increasing proportion of the drug will be in the peripheral compartment, which may or may not be an advantage depending on where the effector tissues are situated. *S*-Substitution also appeared to cause increased localization to the peripheral compartment. In addition it was demonstrated that the incorporation of structures susceptible to biotransformation (such as a sulphur atom) markedly reduces the biological half-life.

While one would expect the distribution characteristics revealed by this dog study to be relevant in man as well, differences regarding metabolism and excretion are not unlikely and require further studies.

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