International Journal of Pharmaceutics, 27 (1985) 71–80
Elsevier

IJP 00902

Allopurinol prodrugs. IV. Improved rectal and parenteral delivery of allopurinol using the prodrug approach as evaluated in rabbits

Hans Bundgaard [1], Erik Falch [1], Søren B. Pedersen [2] and Grethe H. Nielsen [2]

[1] *Royal Danish School of Pharmacy, Departments of Pharmaceutical Chemistry AD and Chemistry BC, DK-2100 and* [2] *GEA Ltd., Copenhagen (Denmark)*

(Received April 24th, 1985)
(Accepted June 27th, 1985)

Key words: allopurinol – prodrugs – rectal absorption – parenteral delivery

Summary

Various N_1-acyl and N-acyloxymethyl prodrug derivatives of allopurinol were evaluated as parenteral and rectal delivery forms of the parent drug. The rectal and parenteral absorption characteristics of twelve derivatives and of allopurinol were assessed in rabbits. Whereas the rectal absorption of allopurinol was less than 3%, the various prodrug derivatives showed a bioavailability ranging from 11 to 94%. The most promising prodrug candidates were N_1-acyloxymethyl derivatives containing a weakly basic amino group in the acyl moiety, e.g. 1-(N,N-diethylglycyloxymethyl)allopurinol hydrochloride. Such compounds are highly soluble in water and possess an adequate lipophilicity at physiological pH. The varying extent of rectal absorption of the derivatives was discussed in relation to the water-solubility and lipophilicity, as expressed in terms of octanol–aqueous buffer partition coefficients.

Introduction

Allopurinol (1) is a widely used drug for the prevention and treatment of hyperuricemic states such as gout as well as for the prevention of the development of hyperuricosuria that often results from the rapid lysis of cells in patients with

Correspondence: H. Bundgaard, Royal Danish School of Pharmacy, Department of Pharmaceutical Chemistry AD, 2 Universitetsparken, DK-2100 Copenhagen, Denmark.

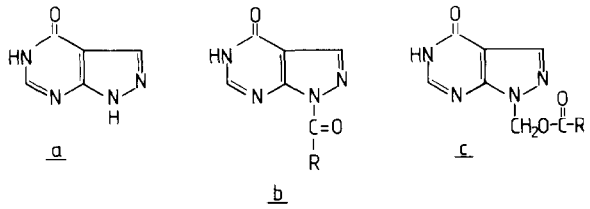


Fig. 1. Chemical structures of allopurinol (a), N_1-acyl (b) and N_1-acyloxymethyl (c) allopurinol derivatives.

malignancies who are undergoing treatment with cytotoxic drugs or radiation (e.g. Elion, 1978). Allopurinol is conventionally administered orally in the form of tablets or capsules. However, the development of nausea and vomiting among patients

TABLE 1

HYDROLYSIS RATE DATA, PARTITION COEFFICIENTS (P) AND WATER-SOLUBILITY (S) OF ALLOPURINOL AND VARIOUS ALLOPURINOL PRODRUGS [a]

Compound (no.)	$t_{1/2}$ (min) at 37°C		log P [b]	S (mg·ml^{-1})
	pH 7.40	80% human plasma		
Allopurinol (1)			−0.55	0.50
1-(Acetyl)allopurinol (2)	26	6	−0.35	0.75
1-(Butyryl)allopurinol (3)	36	2.5	0.85	0.11
1-(Butyryloxymethyl)-allopurinol (4)	193 h	9	0.60	0.35
2,5-Bis(butyryloxymethyl)-allopurinol (5)	35 h	35	1.60	0.094
1,5-Bis(butyryloxymethyl)-allopurinol (6)	24.5 h	25	1.82	0.050
1-(N,N-Dimethylglycyloxy-methyl)allopurinol, HCl (7)	72	7	−0.49 [c]	> 500
1-(N,N-Diethylglycyloxy-methyl)allopurinol, HCl (8)	49	10	0.20 [c]	> 500
–, free base	49	10	0.20 [c]	4.5
1-(N,N-Dipropylglycyloxy-methyl)allopurinol, HCl (9)	50	12	1.27 [c]	> 400
1-(DL-N,N-Diethylalanyloxy-methyl)allopurinol, HCl (10)	21	17	0.72 [c]	> 400
1-(DL-Phenylglycyloxy-methyl)allopurinol, HBr (11)	20	3	−0.15 [c]	> 200
1-(DL-Phenylalanyloxy-methyl)allopurinol, HBr (12)	40	9	0.40 [c]	> 200
1-(L-Leucyloxymethyl)-allopurinol, HBr (13)	17	6	0.19 [c]	> 500

[a] From Bundgaard and Falch (1985a, b and c).
[b] P is the partition coefficient between octanol and water at 22°C.
[c] Partition coefficient between octanol and a 0.05 M borate buffer of pH 8.0.

undergoing cancer chemotherapy frequently precludes the use of oral preparations in these patients as well as in other individuals who are unable to take or retain oral medications. Alternative means of administering allopurinol may be provided by the use of injectable and rectal preparations.

Parenteral dosage forms for a simple injection are, however, not available because of the low solubility of allopurinol in water (0.5 mg · ml^{-1} at 25°C) or other solvents suitable for parenteral administration, and when given rectally to man in the form of various suppository preparations only very minute amounts (< 5%) are absorbed (Chang et al., 1981; Appelbaum et al., 1980, 1982).

Since these delivery problems can primarily be attributed to the low water and lipid solubility of allopurinol (Bundgaard and Falch, 1985a), it appears likely that the delivery characteristics of the compound can be improved by using the prodrug approach, i.e. development of derivatives possessing both a high water-solubility and lipophilicity at physiological pH (pH 7–8) and being capable of reverting rapidly and quantitatively to the parent drug following absorption.

Studies along this direction were undertaken in our laboratories and two potentially useful prodrug types were developed, N_1-acyl derivatives and N-acyloxymethyl derivatives (Fig. 1). A detailed description of the synthesis of these prodrugs and of their stability and physicochemical properties has been given in previous papers (Bundgaard and Falch, 1985a, b, c). In the present work, the bioavailability of a number of these allopurinol prodrugs has been assessed in rabbits following rectal and parenteral administration. Some preliminary data on these studies have already been published elsewhere (Bundgaard and Falch, 1985d). The derivatives studied include two N_1-acyl derivatives (2 and 3) and ten N-acyloxymethyl derivatives (4–13). The physicochemical properties of these compounds are summarized in Table 1.

Materials and Methods

Materials

Allopurinol is a product of Gea A/S, Copenhagen. Oxipurinol was purchased from Sigma Chemicals, St. Louis. The preparation of the allopurinol prodrugs studied has been described previously (Bundgaard and Falch, 1985a, b and c). Polyethylene glycol (PEG) 6000 and adeps solidus (Ph. Nord. 63 grade) were obtained from Mecobenzon A/S, Copenhagen.

Preparation of suppositories and enema

Suppositories were prepared by mixing allopurinol or the prodrugs (particle size 25–50 μm) with the molten suppository bases and pouring the mass into brass moulds (1.15 ml). The suppositories contained 25 mg allopurinol or the equivalent amount (on a molar basis) of prodrug.

To prepare the micro-enema the water-soluble allopurinol prodrug was dissolved in water containing 0.5% methylcellulose, the pH being adjusted to 6 by the addition of sodium hydroxide.

Bioavailability studies in rabbits

Male albino rabbits weighing 2.9–3.1 kg were fasted for 24 h prior to rectal drug administration. The animal was secured in supine position and a suppository or enema was administered into the anus. The suppository was inserted just inside the internal sphincter and the enema (2 ml) administered about 5 cm within the rectum using a rectal polyethylene tube. During the experiments it was controlled that there was no leakage from the anus. After administration, blood samples were taken from the ear vein at appropriate times in heparinized test tubes. The plasma samples obtained after centrifugation were frozen until the time of analysis. An interval of at least 7 days was allowed prior to the next experiment in the same rabbit.

For intravenous administration 2.5 ml of an alkaline allopurinol sodium solution (10 mg · ml^{-1} of allopurinol) or 2.5 ml of an aqueous solution of 1-(N,N-diethylglycyloxymethyl)allopurinol hydrochloride (23 mg · ml^{-1}, equivalent to 10 mg · ml^{-1} of allopurinol) was given in the marginal ear vein.

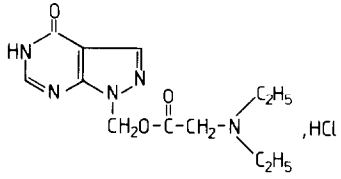
Analysis of plasma samples

The plasma samples were analyzed for allopurinol and oxipurinol by using a slight modification of the HPLC procedure described by Kramer and Feldman (1979). The reversed-phase column (Spherisorb ODS, 5 μm) was eluted with a phosphate buffer (0.05 M, pH 6.0) containing 1.5% v/v of acetonitrile, the eluate being monitored at 252 nm.

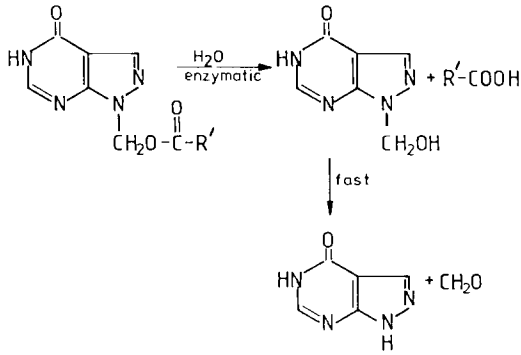
Results and Discussion

Intravenous administration

N_1-Acyloxymethyl derivatives of allopurinol containing an amino function in the acyl moiety have previously been identified as potentially useful parenteral delivery forms of the parent drug (Bundgaard and Falch, 1985c). Such derivatives are in salt form (e.g. hydrochlorides) highly soluble (> 30% w/v) in water and they are rapidly hydrolyzed in human plasma, resulting in the regeneration of allopurinol in quantitative amounts (cf. Table 1). The hydrolytic removal of the 1-acyloxymethyl groups takes place via a two-step reaction as depicted in Scheme 1 (Bundgaard and Falch, 1985b and c). One of the derivatives, 1-(N,N-diethylglycyloxymethyl)allopurinol hydrochloride (8), was administered intravenously to four rabbits in a dosage corresponding to 25 mg of allopurinol. An alkaline solution of allopurinol sodium was also given intravenously to the same animals in an equivalent amount. As seen from Fig. 2 essentially the same plasma levels of allopurinol and its major metabolite



8



Scheme 1.

oxipurinol were obtained by administration of the prodrug and the parent drug. The hydrolysis of compound 8 to produce allopurinol proceeds very rapidly. Thus, whereas the half-life of hydrolysis in human plasma is 10 min (Table 1), it was found to be less than 10 s in 80% rabbit plasma at 37°C in vitro.

The pharmacokinetics of allopurinol in rabbits has apparently not been reported previously. It can be seen from Fig. 2 that allopurinol is rapidly metabolized to oxipurinol. The area under the plasma concentration–time curves (AUC) for allopurinol is less than 3% of the AUC for oxipurinol. This parallels the behaviour of allopurinol in humans where the drug is metabolized to oxipurinol to an extent of about 80–90% (Elion, 1978; Hande et al., 1978; Breithaupt and Tittel, 1982). The elimination of oxipurinol from plasma proceeds according to first-order kinetics (Fig. 2), the half-life being 1.7 h.

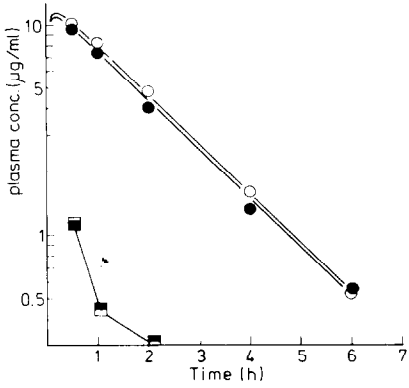


Fig. 2. Mean plasma concentrations of oxipurinol (circles) and allopurinol (squares) following intravenous administration to rabbits (n = 4) of allopurinol sodium (○, □) and 1-(N,N-diethylglycyloxymethyl)allopurinol hydrochloride (8) (●, ■) in amounts corresponding to 25 mg of allopurinol.

TABLE 2

ABSORPTION CHARACTERISTICS AND BIOAVAILABILITY OF ALLOPURINOL AND VARIOUS PRODRUGS FOLLOWING RECTAL ADMINISTRATION TO RABBITS [a]

Compound (cf. Table 1)	No. of rabbits	C^{b}_{1h} ($\mu g \cdot ml^{-1}$)	$AUC_{0\text{-}6h}$ ($\mu g \cdot ml^{-1}$)×h	Bioavailability (%)
1	3	0.2±0.1	0.5±0.4	~3
2	2	1.5±0.4	5.0±2.8	23
3	2	2.0±0.1	6.3±1.3	29
4	2	3.7±1.6	10.1±4.8	46
5	2	1.1±0.8	3.5±2.8	16
6	3	0.9±1.1	2.4±2.1	11
7	2	2.3±0	7.4±1.6	34
8	12	3.8±1.2	12.6±3.9	57
9	4	5.9±2.3	16.3±9.1	74
10	4	6.5±1.7	20.7±5.3	94
11	4	2.1±0.6	6.6±3.5	30
12	4	1.7±0.8	4.7±2.2	21
13	4	1.4±0.4	4.3±1.4	20

[a] All compounds were administered in the form of suppositories made with adeps solidus, the amount of prodrug being equivalent to 25 mg of allopurinol.
[b] Plasma concentration of oxipurinol after 1 h.

Rectal delivery

The major objective of the present work was to identify an allopurinol prodrug showing good bioavailability of the parent drug following rectal administration. Therefore, to sort out the most promising derivatives, the absorption characteristics of several prodrugs was assessed in rabbits. All compounds were given in the form of fatty acid suppositories, the amount of prodrug being equivalent to 25 mg of allopurinol. Blood samples were taken at 1, 2.5 and 6 h and the absorption was characterized by the oxipurinol plasma concentration–time curves. The area under these curves ($AUC_{0\text{-}6h}$) were determined by the trapezoidal rule and the systemic or absolute bioavailability (F%) of the rectal preparations was determined according to:

$$F\% = \frac{AUC_{(rectal)}}{AUC_{(allopurinol\,i.v.)}} \times 100 \qquad (1)$$

The doses given rectally and of allopurinol intravenously were identical on a molar basis. The AUC value for allopurinol i.v. was determined in 4 rabbits.

The results of the rectal absorption studies are shown in Table 2 and Fig. 3. It is readily seen that it is feasible to select prodrug derivatives affording a greatly enhanced rectal delivery of allopurinol. The observed bioavailability of allopurinol is only about 3%, corresponding to previous findings in man (Chang et al., 1981; Appelbaum et al., 1980, 1982). The maximal plasma concentrations of oxipurinol occurred for all compounds at the first sampling time (1 h), thus indicating a rapid absorption. The peak plasma concentration time (T_{max}) may certainly occur even

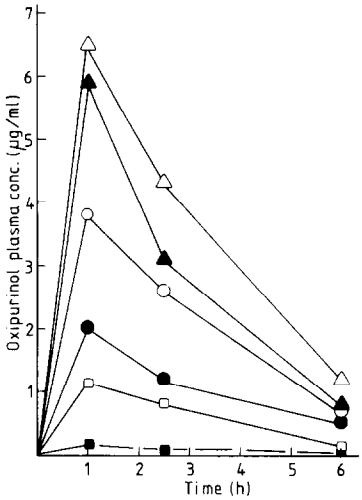


Fig. 3. Mean plasma concentrations of oxipurinol following rectal administration to rabbits of fatty acid suppositories containing allopurinol or allopurinol prodrugs in equivalent amounts (~ 25 mg allopurinol). Key (see Table 1): ■, compound 1; □, compound 5; ●, compound 3; ○, compound 8; ▲, compound 9; △, compound 10.

earlier since an experiment with compound 8 in two rabbits, involving more frequent blood sampling times, showed a T_{max} of 15–20 min. Therefore, the bioavailability calculated from the AUC values given in Table 2 is most likely slightly underestimated. No intact prodrug was detected in any plasma samples in accordance with the rapid prodrug–drug conversion.

Drug absorption from the rectum is not essentially different from that in other parts of the gastrointestinal tract and passive diffusion is regarded as the main governing absorption mechanism (de Blaey and Polderman, 1980; de Boer et al., 1982). Therefore, the solubility and partitioning properties of the drug or prodrug substances are considered to be of paramount importance. However, only sparse information is available on the water and lipid solubilities required to ensure optimal rectal absorption of drug substances, even in qualitative terms, which contrasts with the much greater knowledge concerning the oral absorption (e.g. Ho et al., 1977; Yalkowsky and Morozowich, 1980). Since only a little fluid is present in the rectum, a greater water-solubility is certainly required for a compound to be absorbed rectally as compared with oral absorption. For rabbits, Nishihata et al. (1984) have recently shown that the effective rectal fluid volume available to dissolve drugs is only about 0.1 ml. On the other hand, a certain lipophilicity, e.g. as expressed in terms of octanol–water partition coefficients, is required to allow a permeation through the rectal membrane. Thus, it has been shown that the extent of rectal absorption of some water-soluble penicillins in rabbits increases with increasing partition coefficients (Murakami et al., 1981). Since water-solubility and lipophilicity generally show a reverse relationship it is apparent that an optimal balance between these properties is required to achieve optimal absorption.

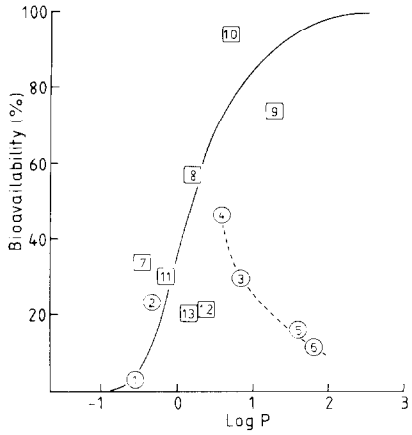


Fig. 4. The rectal bioavailability of allopurinol and various allopurinol prodrugs plotted against log P for the compounds. The numbers refer to the compounds listed in Table 1

Although several of the allopurinol prodrugs were studied in only a limited number of animals the results obtained may provide some generally useful information on the physicochemical properties required for optimal rectal absorption. The prodrugs show a great variation in their aqueous solubilities and lipophilicities (Table 1) as do the observed extents of absorption (Table 2). In Fig. 4 the bioavailability of the compounds has been plotted against log P. The pH value of the rectal fluid is in the range of 7.5–8 (de Blaey and Polderman, 1980; Bitterman et al., 1967) and therefore, the partition coefficients used for the ionizable derivatives were determined using an aqueous phase with a physiological pH (cf. Table 1). At first glance the plot in Fig. 4 reveals no apparent relationship between bioavailability and lipophilicity. If, however, the different water-solubilities of the compounds are taken into account the data indicate that the extent of absorption increases in the expected manner with increasing log P. For example, compounds 1, 2 and 4 have almost the same water-solubilities and for these compounds the bioavailability is increased with increased log P values. Compounds 3, 5 and 6 are very poorly soluble in water (< 0.1 mg $\cdot$ ml^{-1}) and their diminished bioavailability can certainly be ascribed to this. It should be noticed that degradation of the prodrugs to allopurinol at pre-absorption sites in the rectum cannot be excluded; this may possibly explain the unexpectedly low bioavailability observed for compounds 12 and 13.

The absorption study shows that the most promising prodrug candidates are N_1-acyloxymethyl derivatives containing a slightly basic amino function (pK_a 6.5–7.1) in the ester moiety (compounds 8–10).

These derivatives combine good aqueous solubility properties with an adequate lipophilicity at pH values corresponding to those in the rectum and show a facile enzymatic hydrolysis under physiological conditions (cf. Table 1).

It is recognized that formulation factors can be of crucial importance for rectal absorption (de Blaey and Polderman, 1980). The influence of such variables was assessed with compound 8. As seen from Table 3 suppository formulations of the

TABLE 3

BIOAVAILABILITY OF ALLOPURINOL FOLLOWING RECTAL ADMINISTRATION OF 1-(N,N-DIETHYLGLYCYLOXYMETHYL)ALLOPURINOL (8) (AS HYDROCHLORIDE OR FREE BASE) IN VARIOUS FORMULATIONS TO RABBITS

Formulation	No. of rabbits	Bioavailability (%)
Suppository based on adeps solidus (8, HCl)	12	57 ± 7
Suppository based on PEG 6000 (8, HCl)	6	59 ± 18
Suppository based on PEG 6000 (8, free base)	6	85 ± 22
Micro-enema of 8, HCl (pH ~ 6)	6	53 ± 19

hydrochloride salt of the compound, prepared with adeps solidus or a water-soluble suppository base (polyethylene glycol 6000), showed the same bioavailability. An aqueous solution of the compound administered as an enema preparation showed a bioavailability similar to the suppository formulations, thus indicating that the release of the prodrug from the suppositories is not the rate-determining step in the overall absorption. Suppositories prepared by dissolution of compound 8 in its free base form in polyethylene glycol 6000 showed the greatest bioavailability (85%) but the differences are not statistically significant ($P > 0.05$).

From the data obtained, the derivatives 8, 9 and 10 appear to be the most promising prodrug candidates. Taking several other criteria into account, e.g. in vitro stability, toxicity of the prodrug as well as of the pro-moiety released from the derivative, and ease of synthesis and purification, compound 8 was chosen as the potentially most suitable prodrug of allopurinol for both parenteral and rectal administration. In experiments in humans (to be reported later) this compound has been found to be well absorbed with an absolute bioavailability of about 40% following rectal administration.

References

Appelbaum, S.J., Mayersohn, M., Perrier, D. and Dorr, R.T., Allopurinol absorption from rectal suppositories. Drug Intell. Clin. Pharm., 13 (1980) 789.

Appelbaum, S.J., Mayersohn, M., Dorr, R.T. and Perrier, D., Allopurinol kinetics and bioavailability. Intravenous, oral and rectal administration. Cancer Chemother. Pharmacol., 8 (1982) 93–98.

Bitterman, W., Spencer, R.J., Huizenga, K.A. and Shorter, R.G., Measurement of contact pH in the human rectum in health and disease. Gastroenterology, 53 (1967) 288–290.

Breithaupt, H. and Tittel, M., Kinetics of allopurinol after single intravenous and oral doses. Noninteraction with benzbromarone and hydrochlorothiazide. Eur. J. Clin. Pharmacol., 22 (1982) 77–84.

Bundgaard, H. and Falch, E., Allopurinol prodrugs. I. Synthesis, stability and physicochemical properties of various N_1-acyl allopurinol derivatives. Int. J. Pharm., 23 (1985a) 223–237.

Bundgaard, H. and Falch, E., Allopurinol prodrugs. II. Synthesis, hydrolysis kinetics and physicochemical properties of various N-acyloxymethyl allopurinol derivatives. Int. J. Pharm., 24 (1985b) 307–325.

Bundgaard, H. and Falch, E., Allopurinol prodrugs. III. Water-soluble N-acyloxymethyl allopurinol derivatives for rectal or parenteral use. Int. J. Pharm., 25 (1985c) 27–39.

Bundgaard, H. and Falch, E., Improved rectal and parenteral delivery of allopurinol using the prodrug approach. Arch. Pharm. Chem., Sci. Edn., 13 (1985d) 39–48.

Chang, S.-L., Kramer, W.G., Feldman, S., Ballentine, R. and Frankel, L.S., Bioavailability of allopurinol from oral and rectal dosage forms. Am. J. Hosp. Pharm., 39 (1981) 365–368.

de Blaey, C.J. and Polderman, J., Rationales in the design of rectal and vaginal delivery forms of drugs. In E.J. Ariëns (Ed.), Drug Design, Vol. IX, Academic Press, London, 1980, pp. 237–266.

de Boer, A.G., Moolenaar, F., de Leede, L.G.J. and Breimer, D.D., Rectal drug administration: clinical pharmacokinetic considerations. Clin. Pharmacokin., 7 (1982) 285–311.

Elion, G.B., Allopurinol and other inhibitors of urate synthesis. Handbook Exp. Pharmacol., 51 (1978) 485–514.

Hande, K., Reed, E. and Chabner, B., Allopurinol kinetics. Clin. Pharmacol. Ther., 23 (1978) 598–605.

Ho, N.F.H., Park, J.Y., Morozowich, W. and Higuchi, W.I., Physical model approach to the design of drugs with improved intestinal absorption. In Roche, E.B. (Ed.), Design of biopharmaceutical properties through prodrugs and analogs, American Pharmaceutical Association, Washington, DC, 1977, pp. 136–227.

Kramer, W.G. and Feldman, S., High-performance liquid chromatographic assay for allopurinol and oxipurinol in human plasma. J. Chromatogr., 162 (1979) 94–97.

Murakami, T., Tamauchi, H., Yamazaki, M., Kubo, K., Kamada, A. and Yata, N., Biopharmaceutical study on the oral and rectal administrations of enamine prodrugs of amino acid-like β-lactam antibiotics in rabbits. Chem. Pharm. Bull., 29 (1981) 1986–1997.

Nishihata, T., Sakakura, T., Hitomi, M., Yamazaki, M. and Kamada, A., Enhancement of rectal absorption of ampicillin by sodium salicylate in rabbits. Chem. Pharm. Bull., 32 (1984) 2433–2438.

Yalkowsky, S.H. and Morozowich, W., A physical chemical basis for the design of orally active prodrugs. In Ariëns, E.J. (Ed.), Drug Design, Vol. IX, Academic Press, London, 1980, pp. 121–185.