International Journal of Pharmaceutics, 40 (1987) 165–171
Elsevier

IJP 01388

Assessment of in vivo drug dissolution by means of numerical deconvolution considering gastrointestinal availability

Martin Nicklasson [1], Christina Graffner [1] and Maj-Inger Nilsson [2]

[1] *Pharmaceutics Solid Systems and* [2] *Clinical Pharmacology, Research and Development Laboratories, Astra Alab AB, Södertälje (Sweden)*

(Received 22 December 1986)
(Accepted 1 June 1987)

Key words: Numerical deconvolution; Dissolution in vivo; Drug absorption (gastrointestinal); Remoxipride; Microcapsule

Summary

Numerical deconvolution has been applied for calculating the in vivo dissolution process. Due to intraindividual variations in drug absorption on different occasions of administration, a misleading picture of the actual amount dissolved in vivo can be obtained. A modified method for calculating the amount of drug dissolved in vivo is presented which considers both the reference formulation (weighting function) and the test formulation (response function) as equally available to the gastrointestinal tract, i.e. the place where the dissolution process actually occurs. The potential antipsychotic compound, remoxipride, administered as controlled release microcapsules, is used as a model system. It is demonstrated that if the deconvolution data are compensated for the amount of remoxipride absorbed from each formulation (test and reference), very good quantitative in vitro/in vivo dissolution correlations are obtained for each subject. The impact of considering both the conventional deconvolution curve and the modified one will be discussed.

Introduction

Deconvolution has been found to be very useful for the assessment of drug input rate in vivo such as absorption and dissolution (Nogami and Hanano, 1967; Kiwada et al., 1977; Cutler, 1978; Cutler, 1981; Langenbucher, 1982a and b; Tucker, 1983). When calculating an input process in vivo, raw data to be used in the deconvolution calculation are generally obtained from plasma concentration vs. time relationships. Even if the applications of pharmacological responses for such calculations have been shown (Smolen, 1976b), plasma concentrations are most frequently used. In the case of in vivo dissolution, data are thus taken from a totally different in vivo compartment compared to where the dissolution process actually occurs. Furthermore, two different dosage forms of the drug (reference and test formulation) administered on two different occasions are necessary for the performance of a deconvolution calculation. Bearing these things in mind, one must realize that there is always a risk of obtaining a misleading picture of the actual amount of drug dissolved in vivo due to intraindividual variations regarding the oral absorption process. Thus, the concept of preabsorption, i.e. the gastrointestinal

Correspondence: M. Nicklasson, Research and Development Laboratories, Pharmaceutics Solid Systems, Astra Alab AB, S-151 85 Södertälje, Sweden

bioavailability, must be taken into account (Smolen, 1976a). The extent of drug released in the GI tract has also recently been described by Gillespie and Veng Pedersen (1985) as the gastrointestinal bioavailability.

It is the aim of this paper to present a modified method for calculating the cumulative amount of drug dissolved in vivo, generated from a numerical deconvolution method which assumes equal availability of the reference and test formulation in the GI tract. The method is regarded as applicable only when administering a physiologically stable drug with no time-variant disposition kinetics and with negligible first-pass metabolism. Calculations are based on data obtained from an absorption study on the potential antipsychotic compound remoxipride, administered as a controlled-release test formulation to healthy subjects.

Materials and Methods

Dosage forms

Microcapsule cores of remoxipride hydrochloride monohydrate (hereby denoted as remoxipride) were prepared by an extrusion and spheronization process. The cores were coated with an ethylcellulose membrane using an air-suspension technique. Microcapsules corresponding to a dosage strength of 100 mg remoxipride were filled into hard gelatin capsules. An oral aqueous solution containing 2 mg remoxipride per ml was used as a reference formulation and 50 ml of this solution was given.

In vitro dissolution

The in vitro dissolution rate of remoxipride was determined from the controlled-release microcapsules by means of both the USP XX Paddle method and the Flow-Through method (Langenbucher and Rettig, 1977; Möller, 1983).

For the Paddle method, the release rate from each 100 mg dose was studied in water and at pH 1.2 and 7 using 900 ml of each medium. The rotating speed of the paddles was 50 rpm but, in water, the influence of agitation was also investigated at 100 rpm. The dissolution medium was continuously recirculated through flow cuvettes via a peristaltic pump, and the amount dissolved was determined spectrophotometrically at 286 nm.

The Flow-Through technique (SOTAX AG, Switzerland) was used in order to investigate if there was any influence on the dissolution rate due to hydrodynamic or sink conditions. For this experimental design, fresh water (37 °C) was continuously pumped through each flow cell containing the remoxipride microcapsules and samples were taken from the output flow for spectrophotometry. Two cell diameters were used at a flow rate of 8 ml/min, i.e. 12 and 22.6 mm, respectively. For the 12 mm diameter, a flow rate of 16 ml/min was also tested. This experimental technique and the mathematical applications for the calculation of in vitro dissolution rates have thoroughly been described by Langenbucher and Rettig (1977). The mean values of 6 determinations are reported.

Bioavailability study

The study was performed at the Department of Anesthesiology at St. Erik's Hospital, Stockholm, Sweden and was approved by the Ethics Committee at St. Erik's Hospital. The subjects were informed both orally and in writing about the aim of the study.

The study was conducted in 8 Caucasian males aged between 27 and 38 years with a body weight between 71 and 91 kg. All subjects were found to be healthy by medical history, physical examination, blood and urine analyses and ECG. Two experiments were performed in each subject. First, all subjects received the microcapsule formulation at the same time, and then, on a later occasion, the reference solution. At least one week elapsed between the experiments. On each occasion, the subjects were given a single dose of 100 mg remoxipride together with 150 ml of water. The subjects had been fasting overnight and they remained fasting for another 2 h after administration; then a standardized breakfast was served. Lunch was served 4 h after administration. Blood samples were collected into heparinized tubes (Venoject) at standardized time intervals up to 32 h (cf. Fig. 3). Plasma was separated by centrifugation and stored at −20 °C until analysis. Plasma concentrations of remoxipride were determined

after alkalinization of 0.50 ml plasma and extraction into diethyl ether/*n*-hexane (80 : 20 v/v). After centrifugation and evaporation of the organic phase, the residue was dissolved in phosphate buffer pH 2 and injected on an HPLC column (Rainin, MiChrosorb C18, 3 μm, 100 × 4.6 mm). The mobile phase was phosphate buffer pH 2 and acetonitrile (74 : 26 v/v) with the addition of DMNA (*N*,*N*-dimethyl-*N*-nonylamine, 5×10^{-4} M). UV-detection was performed at 214 nm with a detection limit of 0.02 μM.

Calculations

Extent of bioavailability

The area under the remoxipride plasma concentration vs time curve (AUC) between 0 and 32 h was calculated by using the trapezoidal method. The log-mean trapezoidal method was applied during the log-linear portion of the plasma concentration vs time curve. The remaining area (from 32 h to time infinity) was calculated from the ratio between the last determinable plasma concentration and the calculated elimination rate constant. The latter was obtained by linear regression analysis of log plasma concentration vs time between 12 and 32 h ($n = 4$).

Numerical deconvolution

The unit input impulse to the gastrointestinal tract is considered to be the response to a unit dose of an oral solution of the drug. The plasma concentration time course thus obtained is defined as the weighting function of the system (W(t)). The plasma concentration–time function obtained following administration of a solid oral dosage form of the same drug is defined as the response function of the system (R(t)). The in vivo dissolution time function (I(t)), can then be calculated by combining the weighting and response functions by means of deconvolution. The following numerical algorithm can be applied,

$$I_n = (R_n/\Delta t - I_1 W_n - I_2 W_{n-1} - \ldots I_{n-1} W_2)/W_1 \quad (1)$$

where I_n is the amount of drug dissolved in the nth time interval using the midpoint method described by Langenbucher (1982a) and Δt is the selected time interval. A time interval of 1 h was used with linear interpolations for the functions involved (Langenbucher, 1982a; Langenbucher, 1982b; Nicklasson et al., 1984; Nicklasson et al., 1985). The cumulative amount dissolved at time t, Q_n (%), is then obtained by

$$Q_n = 100 \times (I_1 + I_2 + I_3 + \ldots I_n) \quad (2)$$

However, Eqns. 1 and 2 do not just consider the true dissolution process as absorption is highly involved simultaneously and erroneous results may be obtained. Also, the drug may dissolve in the gastrointestinal tract but not be absorbed. In rare cases, a smaller fraction of the dose may be absorbed from the reference formulation compared to the test formulation. In order to calculate the true in vivo dissolution, one has to assume the test and reference formulations as equally available to the gastrointestinal tract on both occasions of administration in the same subject. By incorporating the actually found amount of drug absorbed from each test formulation into Eqn. 2, a more adequate in vivo dissolution process is assumed to be obtained, i.e.,

$$Q_n = 100 \times \left(I_1 + I_2 + I_3 + \ldots I_n \times \frac{\mathrm{AUC}_{\mathrm{solution}}}{\mathrm{AUC}_{\mathrm{solid}}} \right) \quad (3)$$

where AUC is the amount of drug absorbed from each dosage form. Eqn. 3 describes a way of estimating the in vivo dissolution per se without any influence from the absorption process.

Results and Discussion

Remoxipride is a chemically stable, freely soluble compound with no known time-variant disposition kinetics or first-pass metabolism. The pK_a value is 8.9, and its intrinsic dissolution characteristics have been reported (Nicklasson et al., 1983; Nicklasson and Magnusson, 1985). Linear system and moment analysis have been applied to remoxipride, and the results showed that these two

non-compartmental methods produced valuable information which can be used in a dosage form design program on the compound (Nicklasson et al., 1985).

The in vitro dissolution results in Figs. 1 and 2 demonstrate a very consistent behavior of the microcapsule formulation at various experimental conditions. Both the Paddle method and the Flow-Through method produce similar results independent of agitation or pH conditions. The total in vitro dissolution time is about 10–12 h and the average mean dissolution time, obtained by means of moment analysis (Yamaoka et al., 1978), was found to be about 3.5 h.

Fig. 3 shows the mean plasma concentration vs time curves for the two remoxipride formulations. Table 1 shows some pharmacokinetic data. As can be seen, relatively large intraindividual variations of the AUCs are obtained following administration of the two different dosage forms. As mentioned, the raw data to be used in Eqn. 1 are taken from the plasma, a totally different place in the body compared to where the dissolution actually occurs. Thus, problems with intraindividual variations regarding drug absorption might therefore interfere with the outcome of the in vivo dissolution calculation. Values like the hypothetical curve shown in Fig. 4 may be generated which makes

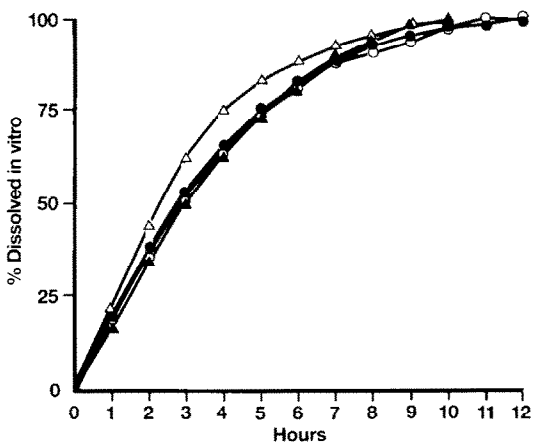

Fig. 1. In vitro dissolution profiles for remoxipride microcapsules using the USP XX Paddle method. (○), 50 rpm water; (●), 100 rpm water (△), 50 rpm pH 1.2; (▲), 50 rpm pH 7. Mean values, $n = 6$.

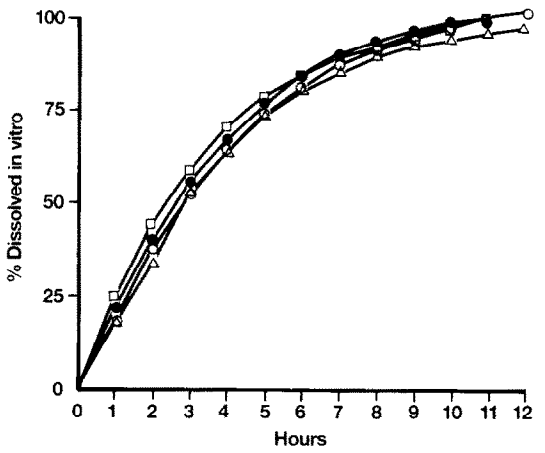

Fig. 2. In vitro dissolution profiles for remoxipride microcapsules in water, 37°C. (○), Paddle method 50 rpm; (●), Flow-Through 8 ml/min 12 mm cell diameter; (△), Flow-Through 8 ml/min 22.6 mm cell diameter; (□), Flow Through 16 ml/min 12 mm cell diameter. Mean values, $n = 6$.

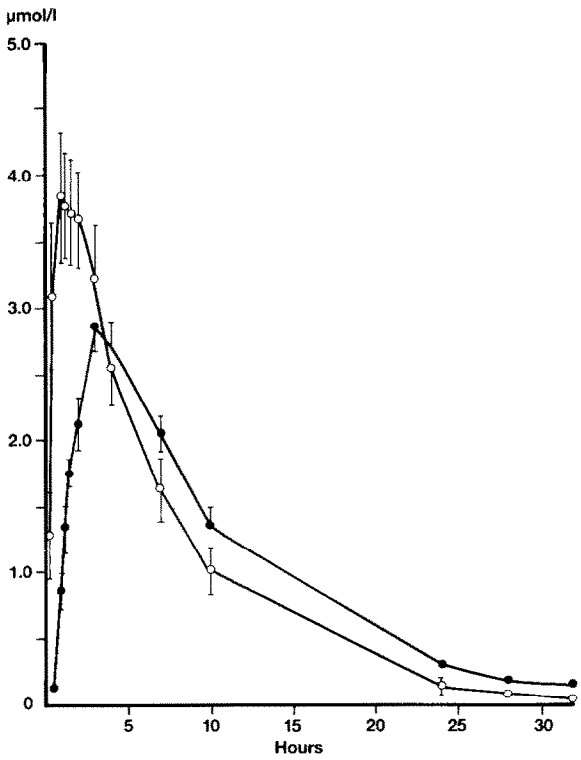

Fig. 3. Mean plasma concentrations ± S.E.M. after administration of a single oral dose of 100 mg remoxipride to 8 healthy volunteers. (○), Aqueous solution; (●), microcapsules.

TABLE 1

Experimental values

Subject	Microcapsules				Solution				F
	C_{max}	t_{max}	AUC	MRT	C_{max}	t_{max}	AUC	MRT	
A	3.71	3	44.86	11.7	5.71	1.5	48.06	8.0	0.93
B	2.09	3	25.06	13.0	2.85	1	23.71	8.2	1.06
C	3.17	3	31.29	8.6	4.30	3	24.42	5.5	1.28
D	2.59	3	22.10	7.7	2.59	1.5	12.47	4.6	1.77
E	3.17	4	46.14	13.5	5.33	1	49.08	9.4	0.94
F	3.01	3	28.12	9.0	2.70	2	15.27	5.7	1.84
G	2.16	3	20.74	10.2	4.03	2	31.83	7.3	0.65
H	3.33	3	33.64	10.1	5.08	0.5	38.04	8.2	0.88
Mean	2.90	3.1	31.49	10.5	4.07	1.5	30.36	7.1	1.17
S.D.	0.573	0.35	9.669	2.09	1.248	0.78	13.898	1.66	0.431

C_{max} (μM), t_{max} (h), AUC (0- time infinity, $\mu M \cdot h$), mean residence time MTR, (h) and relative extent of bioavailability (F), in each subject after administration of 100 mg remoxipride as a controlled-release microcapsule formulation and an oral aqueous solution.

any correlation to in vitro dissolution inadequate since a total amount dissolved above 100% cannot be obtained in the latter case.

Fig. 5 shows in vivo dissolution curves obtained for two subjects. In subject C, a total amount dissolved in vivo of 130% is obtained if one uses Eqns. 1 and 2. The value of 130% is consistent with the calculated relative extent of bioavailability obtained in this subject, i.e. 128%; cf. Table 1. Thus, it seems as if remoxipride is better absorbed on this specific occasion and in this specific subject when administered as a controlled release microcapsule formulation compared to an oral aqueous solution. If, however, the deconvolution data are compensated according to Eqn. 3, a final amount dissolved very close to 100% is obtained as shown by the solid symbols in Fig. 5. Furthermore, if this latter curve is correlated to the in vitro dissolution process, e.g. water, Flow-Through,

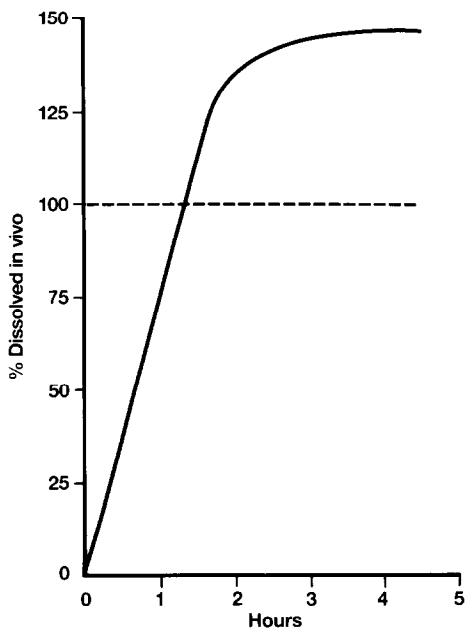

Fig. 4. Schematic presentation of an in vivo dissolution curve obtained by deconvolution.

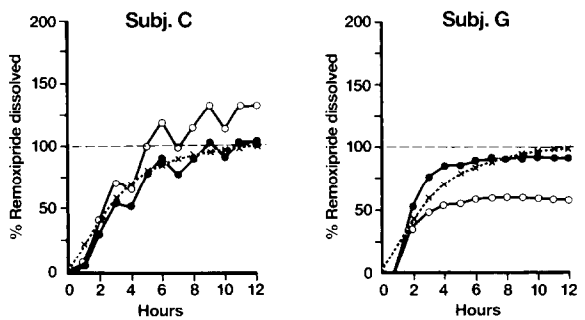

Fig. 5. Cumulative amounts of remoxipride dissolved as a function of time in two healthy subjects, (○), Deconvolution; (●), deconvolution considering the AUC-ratio according to Eqn. 3. The dotted lines denote the in vitro dissolution curve using the Flow-Through method, water 16 ml/min, 12 mm cell diameter.

16 ml/min, 12 mm cell diameter, a very close agreement is obtained. As shown in Figs. 1 and 2, the in vitro dissolution of remoxipride did not show any significant changes due to pH, agitation, or even dissolution method. Due to the high aqueous solubility of remoxipride between pH 1 and 7 (Nicklasson et al., 1983; Nicklasson and Magnusson, 1985), precipitation is not expected following administration of the reference solution. A possible complex formation in the GI-tract can also be excluded. The good in vitro/in vivo correlation demonstrated in Fig. 5 for subject C gives support to the suggested approach of applying AUC ratios when calculating the true in vivo dissolution of a drug by means of numerical deconvolution. It must be emphasized that incomplete in vivo release of a drug can, in some cases, be the dominant reason for poor absorption. Eqn. 3 should, therefore, be used with caution. It is the authors' suggestion that Eqn. 3 always should be used in combination with Eqn. 2. If this is done, the benefit will be twofold. First, the formulator is able to quantitatively correlate the true in vivo dissolution process with the corresponding process in vitro by applying Eqn. 3, and secondly, by comparing the two curves obtained from Eqns. 2 and 3, the formulator can make valuable biopharmaceutical interpretations in order to better understand the behavior of the dosage form in the gastrointestinal tract.

Incomplete absorption may still occur even if the drug actually dissolves due to the position of the formulation in the gut. This is an important aspect since one limitation of the deconvolution approach arises from the assumption that drug released from a solid formulation is absorbed in a manner identical to drug administered as an aqueous solution (Gillespie and Veng Pedersen, 1985). As can be seen for subject G in Fig. 5, a good correlation between in vitro and in vivo is obtained by applying Eqn. 3. However, when using Eqn. 2 the final dissolved amount of 62% is obtained which is close to the value of the relative extent of bioavailability, i.e. 65% (see Table 1). The value obtained from the numerical deconvolution calculation (Eqns. 1 and 2) thus seems to reflect the total amount absorbed in relationship to an oral aqueous solution rather than the true amount dissolved in the GI tract. The difference between the two input functions for subject G in Fig. 5 can be interpreted as the amount that actually has been dissolved but has not been absorbed. As can be seen for subject G in Fig. 5, the total amount dissolved using Eqn. 3 is 90%, which correlates much better to the in vitro dissolution process. However, this also demonstrates that even if one compensates for the actual amount absorbed on each occasion of administration in terms of AUC ratios, the suggested approach seems to be capable of revealing an incomplete in vivo dissolution as well. Fig. 6 finally shows the in vivo dissolution profiles in each subject. The in vivo dissolution profiles have been calculated by means of Eqns. 1 and 3, respectively, and as can be seen, a quantitatively good correlation to in vitro dissolution is found in each subject.

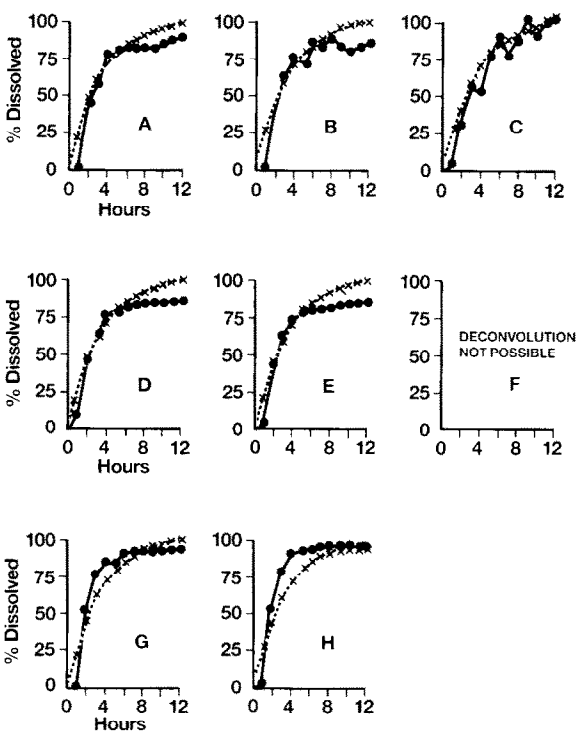

Fig. 6. Comparison between in vitro and in vivo dissolution of remoxipride for each healthy subject. (●), In vivo by means of deconvolution according to Eqn. 3; (×), in vitro by means of the Flow-Through method, water, 16 ml/min, 12 mm cell diameter.

Conclusions

It is important to consider the pharmaceutical and pharmacokinetic properties of a compound before linear system analysis is applied. The suggested method for obtaining the true in vivo dissolution process has been applied to remoxipride which is a chemically stable compound with no known time-variant disposition kinetics or first-pass metabolism. All assumptions made apply only to these premises. If applicable, it is suggested that the presented modified method for the calculation of the in vivo dissolution process is used together with the conventional numerical deconvolution since both approaches can give the formulator valuable information regarding the behavior of the dosage form in the gastrointestinal tract together with a quantitative in vitro/in vivo correlation of the dissolution process itself. Phenomena such as incomplete in vivo dissolution or complete in vivo dissolution but incomplete absorption can be indicated. The method also eliminates absurd dissolution data (more than 100% dissolved) which are due to intraindividual variations in drug absorption on different occasions of administration.

Acknowledgements

The authors wish to thank Dr. Lars Nilsson, Astra Alab AB for skillful accomplishment of the plasma analyses and Erik Vinnars, M.D., St. Erik's Hospital, Stockholm, Sweden for medical supervision of the absorption study.

References

Cutler, D., Linear systems analysis in pharmacokinetics. *J. Pharmacokin. Biopharm.*, 6 (1978) 265–282.

Cutler, D., Assessment of rate and extent of drug absorption. *Pharmacol. Ther.*, 14 (1981) 123–160.

Gillespie, W.R. and Veng Pedersen, P., Gastrointestinal bioavailability: determination of in vivo release profiles of solid oral dosage forms by deconvolution. *Biopharm. Drug Disp.*, 6 (1985) 351–355.

Kiwada, H., Morita, K., Hayashki, M., Awazu, S. and Hanano, M., A new numerical calculation method for deconvolution in linear compartment analysis of pharmacokinetics. *Chem. Pharm. Bull.*, 25 (1977) 1312–1318.

Langenbucher, F. and Rettig, H., Dissolution rate testing with the column method. Methodology and results. *Drug Dev. Ind. Pharm.*, 3 (1977) 241–263.

Langenbucher, F., Numerical convolution/deconvolution as a tool for correlating in vitro with in vivo drug availability. *Pharm. Ind.*, 44 (1982a) 1166–1172.

Langenbucher, F., Improved understanding of convolution algorithms correlating body response with drug input. *Pharm. Ind.*, 44 (1982b) 1275–1278.

Möller, H., Dissolution testing of different dosage forms using the flow-through method. *Pharm. Ind.*, 45 (1983) 617–622.

Nicklasson, M., Brodin, A. and Sundelöf, L.-O., On the determination of true dissolution rate parameters from rotating disc experiments. *Int. J. Pharm.*, 15 (1983) 87–95.

Nicklasson, M., Ellström, K., Sjöqvist, R. and Sjövall, J., Linear systems analysis and moment analysis in the evaluation of bacampicillin bioavailability from microcapsule suspensions. *J. Pharmacokin. Biopharm.*, 12 (1984) 467–478.

Nicklasson, M., Graffner, C., Nilsson, L., Nilsson, M.-I. and Wahlen, A., Absorption properties of the new potential antipsychotic drug remoxipride after oral administration to healthy volunteers. *Pharm. Ind.*, 47 (1985) 986–990.

Nicklasson, M. and Magnusson, A.-B., Program for evaluating drug dissolution in preformulation. *Pharm. Res.*, 6 (1985) 262–266.

Nogami, H. and Hanano, M., Studies on absorption and excretion of drug VIII. The release rate in vivo of antipyretic and analgesic drugs from commercial sugar coated tablets. *Chem. Pharm. Bull.*, 15 (1967) 1002–1009.

Smolen, V.F., Theoretical and computational basis for drug bioavailability determinations using pharmacological data. I. General considerations and procedures. *J. Pharmacokin. Biopharm.*, 4 (1976a) 337–353.

Smolen, V.F., Theoretical and computational basis for drug bioavailability determinations using pharmacological data. II. Drug input-response relationships. *J. Pharmacokin. Biopharm.*, (1976b) 355–375.

Tucker, G.T., The determination of in vivo drug absorption rate. *Acta Pharm. Tech.*, 29 (1983) 159–164.

Yamaoka, K., Nakagawa, T. and Uno, T., Statistical moments in pharmacokinetics. *J. Pharmacokin. Biopharm.*, 6 (1978) 547–558.