JPP Journal of Pharmacy and Pharmacology

JPP 2004, 56: 605–610

Received January 16, 2004
Accepted February 06, 2004
DOI 10.1211/0022357023367
ISSN 0022-3573

# Media to simulate the postprandial stomach I. Matching the physicochemical characteristics of standard breakfasts

Sandra Klein, James Butler, John M. Hempenstall, Christos Reppas and Jennifer B. Dressman

## Abstract

To better predict food effects on the bioavailability/bioequivalence of drugs and drug products from in-vitro data, a dissolution medium that simulates the initial composition of the postprandial stomach was developed. First, the physical parameters of two homogenized standard breakfasts often administered to assess food effects in pharmacokinetic studies were measured. These included pH, buffer capacity, osmolality, surface tension and viscosity. Subsequently, the match of the physical parameters of several commercially available liquid meals, including long-life milk, Ensure and Ensure Plus to those of the breakfasts was evaluated. Of the three liquid meals studied, Ensure Plus had the closest physicochemical behaviour to that of homogenized standard breakfasts. By increasing the viscosity of Ensure Plus with 0.45% pectin, it was possible to obtain a medium that closely resembles the FDA standard breakfast.

Department of Pharmaceutical Technology, Johann Wolfgang Goethe University, 9 Marie Curie Street, Frankfurt am Main 60439, Germany

Sandra Klein, Jennifer B. Dressman

GlaxoSmithKline, Research and Development, Park Road, Ware, Hertfordshire SG12 ODP, UK

James Butler

GlaxoSmithKline, Research and Development, New Frontiers Science Park, Third Avenue, Harlow, Essex CM19 5AW, UK

John M. Hempenstall

School of Pharmacy, University of Athens, Zografou 15771, Athens, Greece

Christos Reppas

**Correspondence:** J. B. Dressman, Institut für Pharmazeutische Technologie, Johann Wolfgang Goethe-Universität Frankfurt, Marie-Curie-Straße 9, 60439 Frankfurt am Main, Germany. E-mail: dressman@em.uni-frankfurt.de

**Acknowledgement and funding:** This work was supported by GlaxoSmithKline, UK and was partly presented in poster form at the 27th International Symposium on Controlled Release and Bioactive Materials, no. 8452, Controlled Release Society, Inc., Paris, France, 2000 and at the 4th World Meeting on Pharmaceutics, Biopharmaceutics and Pharmaceutical Technology, K12/145, APV, APGI, A.D.R.I.T.E.L.F., Florence, Italy, 2002.

## Introduction

The bioavailability of an oral dosage form depends only partly on the properties of the active substance and the excipients. The dosing conditions (i.e. the timing of administration and any co-administered fluids or food) are additional important criteria. In particular, the biopharmaceutical parameters of a formulation can show a broad range of behaviour depending on food intake (Welling 1996; Charman et al 1997).

To assess food effects on drug absorption, a crossover design pharmacokinetic study is usually performed in healthy subjects. In one arm of the study, the drug product is given in the fasted state, in the other it is administered with a standard meal. To reduce the size and number of human studies required to select a suitable drug product, it would be advantageous to be able to pre-screen formulations in-vitro. The choice of an appropriate medium for the in-vitro tests is crucial to the ability to correctly forecast the food effect in pharmacokinetic studies.

Media to simulate fasting conditions in the stomach and small intestine have already been proposed (Galia et al 1998, 1999). Likewise, a medium has been developed to simulate fed-state conditions in the small intestine (Galia et al 1998). Still lacking is a medium that adequately simulates gastric conditions in the fed state. From the following equation, based on Nernst-Brunner and Levich modifications of the Noyes-Whitney model (Hörter & Dressman 1997; Dressman et al 1998), the factors important to the rate of drug dissolution (DR) can be identified:

$$DR = dX_d/dt = A * D/\delta * (C_s - X_d/V) \quad (1)$$

where A is the effective surface area of the drug, D is the diffusion coefficient of the drug, $\delta$ is the effective diffusion boundary thickness adjacent to the dissolving surface, $C_s$ is the saturation solubility of the drug under lumenal conditions, $X_d$ is the amount of drug already in solution and V is the volume of the dissolution medium. Some of these parameters are mainly determined by the physicochemical properties of the drug itself, but several are strongly affected by the conditions in the gastrointestinal tract.

Surfactants in gastric juice may influence the wetting of the drug, which in turn affects the effective surface area available for dissolution. Therefore, the surface tension of the gastric contents, and how this varies between the fed and the fasted state, is important to the prediction of food effects on dissolution. The solubility will obviously be affected by the composition (e.g. fat content of the gastric contents, micelle formation, etc.). In the case of ionizable drugs, buffer capacity and pH of the gastric milieu are also highly relevant to the dissolution rate (Ozturk et al 1988). Generally, an increase in the buffer concentration holds the pH of the dissolving drug surface closer to the bulk pH and therefore influences the solubility of the drug through its degree of ionization and, consequently, its dissolution rate. Since the buffer capacity can vary greatly between fasted and fed state, it is essential to account for this factor. The diffusivity of the drug is inversely proportional to the viscosity of the lumenal contents. Due to the increased viscosity in the postprandial stomach, a decreased diffusivity is expected in most cases. In cases where the release rate of a drug from the dosage form depends on osmotic pressure differences, osmolality is a further variable that can have an impact on the dissolution rate.

Thus, the physicochemical parameters studied (osmolality, pH, buffer capacity, surface tension, viscosity) clearly represent key factors for the in-vivo dissolution of drugs. To predict gastric dissolution more nearly in-vitro, it is particularly important to simulate these parameters adequately in both the fasted and fed state.

Attempts to simulate the initial composition of the gastric contents after food intake have been proposed by several researchers. For example, milk (Macheras et al 1986, 1987, 1988, 1989a, b, 1990; Buckton et al 1989; Krämer 1995; Galia et al 1998; Nicolaides et al 1999) and artificial liquid meals (Nutridrink and Ensure) (Ashby et al 1989; Buckton et al 1989; Junginger et al 1990), emulsions (Ashby et al 1989; Buckton et al 1989; Krämer 1995) and homogenized meals (Krämer 1995) have been used. However, the composition of these media may not optimally reflect the composition of the meals usually administered in relevant bioavailability/bioequivalence studies. For example, they do not address the secretion of acid and pepsin in response to the meal and the decrease in pH in the fed stomach over time. Some also disregard the contribution of solid meal components to the composition in the stomach. With the use of milk as a dissolution medium, further problems such as stability of the medium during the test (if using fresh milk), as well as variability in its composition with source and season, arise. A problem with the use of simple fat emulsions is that they cannot adequately simulate the carbohydrate and protein content of the meal.

It has been suggested (Buckton et al 1989) that a starting point for the design of a suitable dissolution medium to reflect the fed state in the stomach would be to homogenize the meal to be used in the clinical studies with the co-administered volume of water, and measure the pH, buffer capacity and osmolality. A volume of oil reflecting the fat content of the meal could then be added to an aqueous buffer simulating the aforementioned parameters, to produce an appropriate test medium. In an alternative approach to arrive at an in-vitro–in-vivo correlation, Krämer (1995) homogenized the breakfast given in the pharmacokinetic study as the dissolution medium. The medium was standardized by the addition of a pH 6.8 buffer solution, and the resulting homogeneous suspension used for the dissolution tests. Although the use of homogenized meals as a dissolution medium is directly relevant to the meals administered in pharmacokinetic studies, this approach is not ideal due to variability in its composition and the cumbersome and time-consuming preparation. Furthermore, the pH was high due to the addition of buffer.

In summary, improvements can be made in the design of media to simulate the fed state in the stomach. As a first step in designing a suitable medium to simulate the postprandial stomach, various physicochemical properties of long-life milk and two artificial (Ensure and Ensure Plus) liquid meals and two commonly administered standard breakfasts were determined and compared. To obtain a dissolution medium representative of initial gastric conditions in food-effect studies, the properties of the most suitable liquid meal were then adjusted to those of the standard FDA breakfast by the addition of a viscosity elevating agent.

## Materials and Methods

### Meal composition

Tables 1 and 2 show the composition of two typical meals administered in pharmacokinetic studies.

### Materials

Long-life milk (milk, 3.5% fat) was purchased from Meierei Trittau (Germany). Ensure (vanilla flavour) and Ensure Plus (drink, vanilla flavour) were purchased from Abbott Laboratories B.V. (Ross Product Manufacturer Zwolle, Netherlands). Pectin K from apples was obtained from Caesar & Loretz (Hilden, Germany). The standard meals were prepared with the ingredients shown in Table 3.

### Meal and media preparation

The standard breakfast meals (n = 3 per meal type) were prepared and then homogenized in a Wedo model BL-838

**Table 1** Composition of standard breakfast meal 1[a].

| |
|---|
| 2 Slices of toasted white bread with butter |
| 2 Eggs fried in butter |
| 2 Slices of bacon |
| 2 Ounces of hash browned (fried shredded) potatoes |
| 8 Ounces of whole milk |
| Carbohydrate 58 g, 232 kcal, 971 kJ, 24% of calories |
| Protein 33 g, 132 kcal, 552 kJ, 14% of calories |
| Fat 67 g, 603 kcal, 2523 kJ, 62% of calories |

[a]Composition courtesy of GlaxoSmithKline.

**Table 2** Composition of standard breakfast meal 2[a].

| |
|---|
| 1 English muffin with butter |
| 1 Fried egg |
| 1 Slice of cheese |
| 1 Slice Canadian bacon |
| 1 Serving of hash browned (fried shredded) potatoes |
| 6 Ounces of orange juice |
| 8 Ounces of whole milk |
| Carbohydrate 73 g, 292 kcal, 1222 kJ, 45% of calories |
| Protein 29 g, 116 kcal, 485 kJ, 18% of calories |
| Fat 27 g, 240 kcal, 1004 kJ, 37% of calories |

[a]FDA Office of Generic Drugs.

**Table 3** Ingredients of the standard breakfast meals.

| | |
|---|---|
| Bacon | Tulip bacon classic (Tulip International GmbH, Ratingen, Germany) |
| Butter | Kerrygold (Irish Dairy Board, Dublin, Ireland) |
| Canadian bacon | Gelderland bacon (Gelderland GmbH, Emmerich, Germany) |
| Cheese | Dairygold irish cheddar (Dairygold Food Products, Cork, Ireland) |
| Eggs | Quality class A, weight class M (Eiermann & Co. GmbH, Mönlingen, Germany) |
| English muffins | Weizen toastis (Golden Toast, Wendeln Brot, Garrel, Germany) |
| Hash browned potatoes | McCain 1·2·3 Rösti (McCain GmbH, Eschborn, Germany) |
| Orange juice | Granini orange juice without pulp (Eckes-Granini GmbH & Co. KG, Nieder-Olm, Germany) |
| White bread | American sandwich (Golden Toast, Wendeln Brot, Garrel, Germany ) |
| Whole milk | Milk, long life milk, homogenized, 3.5% fat (Meierei Trittau, Germany) |

mixer (Wedo, Germany). The measurement of the physicochemical properties was performed immediately after this procedure. The various Ensure Plus–pectin mixtures were prepared either with a colloid mill (ColloVelox Micro, type T80B2; Brogli & Co, Basel, Switzerland) or with a high-performance homogenizer constructed according to the rotor/stator principle (UltraTurrax, type 25; IKA Labortechnik Janke & Kunkel, Staufen, Germany).

## Characterization of the physicochemical properties

The pH value was measured with a pH meter (model 720A; Orion Research Inc., Beverly, MA). The buffer capacity was quantified by potentiometric titration using 0.1 M hydrochloric acid.

For the determination of the surface tension and osmolality, appropriate dilutions of the standard breakfast meals were prepared with water; they contained 66.7%, 50%, 33.3%, 25% and 20% (m/m) of breakfast, respectively. The dilutions were mixed for 1 min using a Vortex mixer (Vortex-Genie 2; Scientific industries, INC., Bohemia NY) and then centrifuged for 30 min at 3000 rev $min^{-1}$. Following centrifugation, the surface tension of the aqueous phase was measured by a bubble pressure tensiometer (no. 100517A; Sita Messtechnik GmbH, Dresden, Germany). Dilutions containing 25% breakfast exhibited a horizontal, linear relationship in surface activity ($R^2$ 0.990), indicating that the concentrations were above the critical micelle concentration (CMC). It was therefore possible to extrapolate to the surface tension of the undiluted breakfast.

After centrifugation, the aqueous phase of the diluted meals was used to measure the osmolality by semi-micro osmometry (osmometer type ML, no. A0299; Knauer, Berlin, Germany). Similar to surface activity, a linear relationship between meal concentration and osmolality was observed ($R^2$ 0.995). In this case it was possible to use the complete set of dilutions (20–66.7%) for the extrapolation. Unlike the standard meals, milk and Ensure required no dilution for these measurements.

Due to the very different consistencies of the various samples, it was necessary to use two methods for the investigation of viscosity. The viscosity of milk, Ensure and Ensure Plus was determined with an Ubbelohde viscometer (DIN 24501; Schott Glass, Mainz, Germany). However, it was obvious that, due to the complex nature of the meals, the homogenized preparations would not exhibit Newtonian flow characteristics. Therefore, the rheological profiles of the homogenized, standard breakfast preparations were measured ($n = 3$ or 6, as needed) using a rotational viscometer (Physica Rheolab MC1 portable, standard cylinder system Z3/DIN; Physica Messtechnik GmbH, Stuttgart, Germany), which operates according to the Searle principle (cup and bob) at room temperature (~25 °C) at shear stresses in the range 0–500 Pa. The same procedure was followed to establish the rheological profiles of Ensure Plus with the addition of various concentrations of pectin.

## Statistical analysis

Data for the physicochemical properties (in each case at least $n = 6$ per meal composition) was statistically analysed using SigmaStat for Windows Version 3.00 (SPSS Inc. Headquarters, 233 S. Wacker Drive, Chicago, IL) software. Data sets were tested for normality and then differences between the physicochemical properties of the various meals/formulations were examined using either analysis of variance or the Kruskal–Wallis analysis of variance test, as appropriate. Individual differences were determined using post-hoc analysis. In all experiments, $P < 0.05$ was accepted to denote significance. In all rheograms, standard deviations have been omitted for clarity. Coefficients of variation of the data were less than 1% for all combinations of Ensure Plus with pectin, while for the standard breakfasts they were 5–15%.

## Results and Discussion

To aid in the design of a representative, yet easy to handle, medium for dissolution testing, various physical parameters of homogenized breakfasts were measured. The ability of commercially available liquid meals (milk, Ensure and Ensure Plus) to match these parameters was then evaluated. Results are given in Table 4.

In general, the properties of Ensure were much closer to those of the homogenized meals than was the case for milk. Nevertheless, there still were some discrepancies, especially in osmolality and viscosity. The complete balanced nutrition drink Ensure Plus seemed to be the more suitable medium of the two nutrition drinks. Although the two are of similar composition (Table 5), the higher osmolality of Ensure Plus corresponds better to the osmolality of the homogenized, standard meals. Like the other liquid meals, however, the viscosity of Ensure Plus was far lower than that of the homogenized breakfasts. Since it is possible that Ensure Plus drinks slightly vary in composition and flavour between different countries, it needs to be specified here that the European vanilla-flavoured Ensure Plus drink was used in these studies.

**Table 5** Composition and nutritive values[a] of Ensure Plus.

| Nutritive value | Per 200 mL | Per Litre |
|---|---|---|
| Energy (kJ) | 1263 | 6320 |
| Energy (kcal) | 300 | 1500 |
| Carbohydrate (g) | 40.4 | 202 |
| Protein (g) | 12.5 | 62.5 |
| Total fat (g) | 8.4 | 49.2 |
| Saturated fatty acids (g) | 0.98 | 4.9 |
| Essential fatty acids (g) | 2.9 | 14.5 |
| Dietary fibre (g) | 0 | 0 |
| Water (g) | 155 | 773 |
| Minerals, Vitamins | | |

[a]Information from manufacturer.

### Adjustment of the viscosity

By adjusting the viscosity of Ensure Plus, we were able to match both the composition and physical behaviour of the resulting medium to that of the two standard breakfasts. Pectin appeared to be a suitable substance for this purpose for the following reasons: polysaccharides, including pectin, are the basic building materials of plant tissues and are found in many vegetables and fruits; pectin is partially digested to mono- and disaccharides, which are then absorbed and utilized in various metabolic processes; as such, pectin is considered to be a generally-regarded-as-safe (GRAS) material. Further, since pectin is water soluble at room temperature and has excellent dispersion properties, it can be readily incorporated into aqueous solutions.

To ascertain the amount of pectin required to achieve viscosities similar to those of the homogenized, standard meals, mixtures of Ensure Plus with different pectin concentrations (0.25–0.75%) were prepared. Their rheological profiles (Figure 1) were compared with those of the standard breakfast meals (Figure 2).

From the curves in Figures 1 and 2 it can be seen that the rheological profiles of mixtures of Ensure Plus with 0.45% and 0.46% pectin approximately correspond to those of the standard breakfast of the FDA Office of Generic Drugs. To approximate the rheological profile of the GSK breakfast, substantially more pectin would need to be added.

As a second step, several Ensure Plus–pectin mixtures with a pectin concentration of 0.45% were prepared by two different methods: colloid mill and UltraTurrax. Manual preparation proved to be unsuitable for this purpose, since it was very difficult to obtain a homogenous mixture. An advantage of using a colloid mill was that homogeneous

**Table 4** Physicochemical parameters of standard breakfast meals, milk, Ensure and Ensure Plus.

| | | Standard breakfast 1 (62% fat[a]) | Standard breakfast 2 (37% fat[a]) | Milk (48.1% fat[a]) | Ensure (30.1% fat[a]) | Ensure Plus (29.5% fat[a]) |
|---|---|---|---|---|---|---|
| Mass (g) | | 516 (6.0) | 540 (5.5) | | | |
| Volume (mL) | 25 °C | 474 (7.7) | 513 (7.3) | | | |
| Density ($g\,mL^{-1}$) | 25 °C | 1.09 (0.03) | 1.05 (0.03) | 1.03 (0.005) | 1.04 (0.016) | 1.08 (0.003) |
| pH-value | 25 °C | 6.51 (0.01) | 5.28 (0.03) | 6.72 (0.02) | 6.68 (0.01) | 6.62 (0.03) |
| | 37 °C | 6.61 (0.03) | 5.12 (0.04) | 6.63 (0.01) | 6.58 (0.01) | 6.45 (0.02) |
| Buffer capacity | 25 °C | 29.3 (0.9) | 49.6 (1.7) | 14.4 (0.2) | 15.4 (0.1) | 20.0 (0.7) |
| ($mEq\,pH^{-1}\,L^{-1}$) | 37 °C | 30.1 (1.8) | 47.2 (1.5) | 13.9 (0.2) | 16.4 (0.0) | 21.0 (0.3) |
| Osmolality ($mOsmol\,kg^{-1}$) | | 771 (10) | 713 (10) | 285 (2.7) | 375 (3.5) | 730 (10) |
| Surface tension ($mN\,m^{-1}$) | 25 °C | 52 (1) | 49 (1) | 54.2 (0.4) | 50.5 (0.2) | 53.2 (0.2) |
| | 37 °C | 44 (1) | 45 (1) | 49.8 (0.6) | 47.8 (0.1) | 48.4 (0.1) |
| Viscosity (mPa s) | 25 °C | | | 1.9[b] (0.04) | 6.3[b] (0.09) | 19.1[b] (0.10) |
| | 37 °C | | | 1.5[b] (0.04) | 4.4[b] (0.07) | 12.3[b] (0.10) |

Data are mean values (± s.d.), n = 6 per measurement. [a]% calories derived from fat content. [b]Measured with the Ubbelohde viscometer.

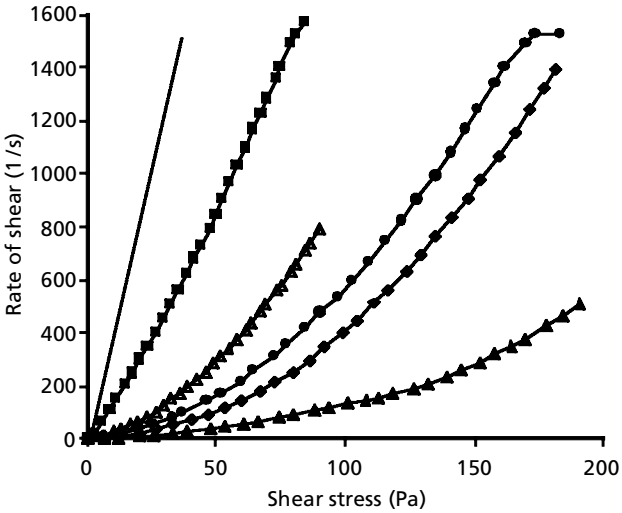


**Figure 1** Rheological profiles of Ensure Plus (—) and Ensure Plus mixtures containing different amounts of pectin (■, 0.25%; △, 0.45%; ●, 0.46%; ◇, 0.50%; ▲, 0.75% pectin); n = 3, mean values are shown.

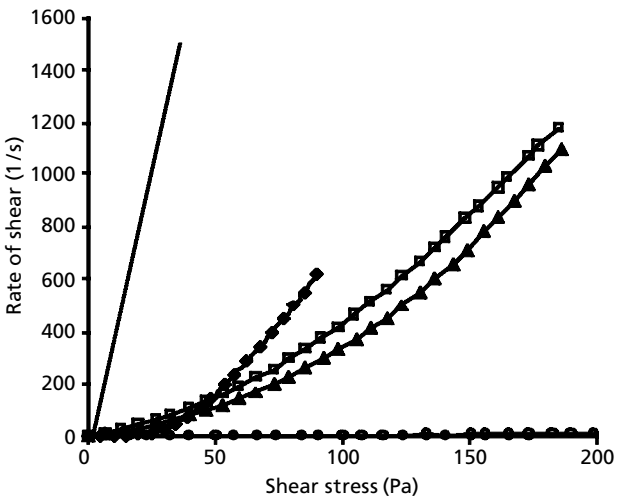


**Figure 2** Rheological profiles observed for Ensure Plus (—), the standard breakfast meals (◆, standard breakfast 1; ◇, standard breakfast 2) and the Ensure Plus mixture containing 0.45% pectin (□, prepared with an UltraTurrax; ▲, prepared with a colloid mill); n = 6, mean values are shown.

preparations were readily obtained, although a high volume (>300–400 mL) of the preparation was needed. In the case of the UltraTurrax, lower volumes could be processed, but homogeneity was compromised at higher volumes. Also, the high shear stresses employed with the UltraTurrax could possibly affect the internal structure of the pectin. Despite these concerns, both manufacturing methods were generally easy to use and delivered reproducible results at the volumes and pectin percentages of interest. The media obtained were homogenous and showed stability against mechanical stress. None of the parameters (pH, buffer capacity, osmolality, surface tension and viscosity) showed significant ($P < 0.05$) changes after stirring at 75 rev min$^{-1}$ and a temperature of 37 °C for 24 h.

Ensure Plus with 0.45% pectin prepared with a colloid mill or UltraTurrax appears to be a suitable medium for the simulation of initial gastric conditions after administration of the FDA standard meal. Further work is needed to determine whether this medium offers advantages over the alternatives in predicting food effects and for establishing in-vitro–in-vivo correlations. Moreover, the secretion of acid and pepsin during gastric residence and its importance for dissolution remain to be evaluated.

## Conclusion

A dissolution medium which closely resembles initial conditions in the stomach after administering the FDA standard breakfast was developed. This consisted of Ensure Plus with the addition of 0.45% pectin.

## References

Ashby, L. J., Beezer, A. E., Buckton, G. (1989) *In vitro* dissolution testing of oral controlled release preparations in the presence of artificial foodstuffs. I. Exploration of alternative methodology: microcalorimetry. *Int. J. Pharm.* **51**: 245–251

Buckton, G., Beezer, A. E., Chatham, S. M., Patel, K. K. (1989) *In vitro* dissolution testing of oral controlled release preparations in the presence of artificial foodstuffs. Part 2. Probing drug/food interactions using microcalorimetry. *Int. J. Pharm.* **56**: 151–157

Charman, W. N., Porter, C. J., Mithani, S., Dressman, J. B. (1997) Physicochemical and physiological mechanisms for the effects of food on drug absorption: the role of lipids and pH. *J Pharm Sci.* **86**: 269–282

Dressman, J. B., Amidon, G. L., Reppas, C., Shah, V. P. (1998) Dissolution testing as a prognostic tool for oral drug absorption: immediate release dosage forms (Review). *Pharm Res.* **15**: 11–22

Galia, E., Nicolaides, E., Horter, D., Lobenberg, R., Reppas, C., Dressman, J. B. (1998) Evaluation of various dissolution media for predicting in vivo performance of class I and II drugs. *Pharm Res.* **15**: 698–705

Galia, E., Horton, J., Dressman, J. B. (1999) Albendazole generics – a comparative in vitro study. *Pharm Res.* **16**: 1871–1875

Hörter, D., Dressman, J. B. (1997) Influence of physicochemical properties on dissolution of drugs in the gastrointestinal tract (Review). *Adv. Drug Del. Rev.* **25**: 3–14

Junginger, H. E., Verhoeven, J., Peschier, L. J. C. (1990) A new in vitro model to detect interactions between controlled release dosage forms and food. *Acta Pharm. Technol.* **36**: 155–160

Krämer, J. (1995) Korrelation biopharmazeutischer *in-vivo-* und *in-vitro*-Daten von Theophyllin und Verapamil Retardpräparaten. Ph.D. thesis, University of Heidelberg

Macheras, P., Koupparis, M., Tsaprounis, C. (1986) Drug dissolution studies in milk using the automated flow injection serial dynamic dialysis technique. *Int. J. Pharm.* **33Z** 125–136

Macheras, P., Koupparis, M., Apostolelli, E. (1987) Dissolution of 4 controlled release theophylline formulations in milk. *Int. J. Pharm.* **36**: 73–79

Macheras, P. E., Koupparis, M. A., Antimisiaris, S. G. (1988) Effect of temperature and fat content on the binding of hydrochlorothiazide and chlorothiazide to milk. *J. Pharm. Sci.* **77**: 334–336

Macheras, P., Koupparis, M., Antimisiaris, S. (1989a) *In vitro* model for exploring CR theophylline–milk fat interactions. *Int. J. Pharm.* **54**: 123–130

Macheras, P. E., Koupparis, M. A., Antimisiaris, S. G. (1989b) Effect of temperature and fat content on the solubility of hydrochlorothiazide and chlorothiazide in milk. *J. Pharm. Sci.* **78**: 933–936

Macheras, P. E., Koupparis, M. A., Antimisiaris, S. G. (1990) Drug binding and solubility in milk *Pharm. Res.* **7**: 537–541

Nicolaides, E., Galia, E., Efthymiopoulos, C., Dressman, J. B., Reppas, C. (1999) Forecasting the in vivo performance of four low solubility drugs from their in vitro dissolution data. *Pharm. Res.* **16**: 1876–1882

Ozturk, S. S., Palsson, B. O., Dressman, J. B. (1988) Dissolution of ionizable drugs in buffered and unbuffered solutions. *Pharm. Res.* **5**: 272–282

Welling, P. G. (1996) Effects of food on drug absorption. *Annu. Rev. Nutr.* **16**: 383–415