In Vivo Evaluation of Enteric-Coated Naproxen Tablets Using Gamma Scintigraphy

Ian R. Wilding,[1,5] John G. Hardy,[1] R. Andrew Sparrow,[2] Stanley S. Davis,[1,3] Paul B. Daly,[4] and John R. English[4]

Received January 20, 1992; accepted April 27, 1992

Seven healthy, male volunteers were entered into a randomized, open crossover study of the gastrointestinal transit of two enteric-coated 500-mg naproxen tablets. Two radiolabeled tablets were given to each volunteer on two occasions separated by 7 days, once in the fasted state and once after breakfast. Radiolabeling of tablets was achieved by the incorporation of samarium-152 oxide during manufacture, followed by neutron activation of the tablet to produce the gamma-emitting isotope samarium-153. No loss of tablet integrity was seen in the stomach and all tablets disintegrated in the small intestine. Onset of tablet disintegration was controlled predominantly by gastric emptying. Time in the small intestine prior to tablet disintegration was independent of food intake. Naproxen blood levels with time were consistent with the delayed release of naproxen from the tablets. Overall, transit, disintegration, and absorption were as expected from an enteric-coated tablet.

KEY WORDS: naproxen; enteric-coated tablets; samarium-153; neutron activation; gastrointestinal transit.

INTRODUCTION

The usefulness of nonsteroidal, anti-inflammatory drugs (NSAIDs) is limited by the relatively high incidence of gastrointestinal (GI) side effects they produce (1). In most patients these are minor, i.e., not life-threatening, but they can cause patients to stop treatment. Major side effects occur much less often and the relationship between minor and major events is unclear. NSAID damage to the gastric mucosa can be observed by endoscopy. Acute studies show that this can be limited by preventing the active NSAID from coming into contact with the gastric mucosa (2). Again, the relevance of these effects to other minor and major side effects is unclear. However, it does appear that NSAID gastric damage is caused by both local and systemic effects (3).

The development of enteric-coated (EC) NSAID tablets has been one way of preventing active drug from coming into contact with the gastric mucosa. For these dosage forms to be effective, they must not disintegrate in the stomach, but they must release all their active drug after leaving the stomach. While this can be studied *in vitro*, there is no real substitute for confirming reliable performance *in vivo* in man.

The technique of gamma scintigraphy has become the most popular method to investigate the GI performance of pharmaceutical dosage forms. Although used for many years by GI physiologists (4), only in 1976 was the fate of pharmaceuticals *in vivo* first investigated by this approach (5,6). In recent years the combination of gamma scintigraphy with conventional pharmacokinetic assessment (pharmacoscintigraphy) has been used to relate the biodistribution of the delivery system to drug absorption (7–13).

Conventional methods of labeling pharmaceutical dosage forms require that the marker be incorporated as late as possible in the manufacturing process to minimize the risk of radioactive contamination. In many cases, the manufacturing process must be scaled down to minimize the amount of radioactivity handled. In the case of complicated delivery systems, such as EC tablets, this may significantly alter the behavior of the product.

These problems can be overcome by the use of stable nuclides and neutron activation methodology to radiolabel dosage forms for scintigraphic evaluation (11,13–15). A stable isotope (e.g., samarium-152, as the isotopically enriched oxide) can be incorporated into the dosage form at a low level and the product irradiated in a neutron source to convert it into the gamma-emitting isotope (i.e., samarium-153). The radiation exposure to a subject following dosing with these radionuclides is comparable to that received using conventional radiolabeling with technetium-99m and indium-111. Using this approach, radiation doses to staff can be minimized, quality assurance maintained, and complicated delivery systems labeled easily and efficiently.

The purpose of this study was to evaluate *in vivo* the GI transit and disintegration characteristics of an EC naproxen formulation manufactured under normal production conditions.

METHODS

Dosage Form Preparation

Granules suitable for tablet compression were manufactured on a Fielder granulator (Model PMA 400) using the currently marketed Naprosyn (Syntex) formulation (batch size, 118 kg). The granules were then dried in a Glatt fluid bed dryer (Model WST-CD 90/120) before being dry granulated through a Stokes Tornado Mill (Model 440). An aliquot of the dry granulation was taken and the isotopically enriched samarium oxide was incorporated in a proportion of 2 mg per tablet. Tablets were compressed on a Rotary D3B Manesty tablet press to contain a nominal 500 mg naproxen per tablet using 15.875 × 7.144-mm capsule-shaped tooling with a total compression weight of 544 mg.

The enteric coating was applied using a Manesty 24-in. Accelacota (Manesty Machines) fitted with Schlick spray guns. Approximately 200 tablets, each containing 2 mg of samarium oxide, were placed in the Accelacota together with approximately 6 kg of "bulking" tablets (each of which weighed approximately 60 mg more than the tablets containing samarium oxide). The tablets were coated using the stan-

[1] Pharmaceutical Profiles Ltd., 2 Faraday Building, Highfields Science Park, Nottingham, England.
[2] Department of Human Morphology, Queen's Medical Centre, Nottingham, England.
[3] Department of Pharmaceutical Sciences, University of Nottingham, England.
[4] Syntex Pharmaceuticals Ltd., Maidenhead, England.
[5] To whom correspondence should be addressed.

0724-8741/92/1100-1436$06.50/0

dard coating solution as per the marketed Naprosyn EC formulation.

Upon completion of tablet coating the tablets were check weighed on a Sade P2 weight sorter (CI Electronics) to recover the tablets containing samarium oxide. Samples of the tablets were disintegration/dissolution tested *in vitro* before (and after) irradiation to ensure integrity of the enteric coat. On each occasion the BP 1988 Disintegration Test for EC tablets was performed. For dissolution testing the tablets were placed in 0.1 *M* hydrochloric acid for 2 hr and then placed in pH 7.4 phosphate buffer for 45 min (Apparatus II BP 1988).

Three days before each leg of the study, tablets were irradiated for 4 min in a neutron flux of 10^{12} n/cm^2/sec. The delay in use allowed the decay of sodium-24 formed during the irradiation. At the time of administration each tablet contained approximately 0.5 MBq samarium-153.

Study Design

This was an open, single-dose, randomized crossover comparison of the GI transit of two 500-mg EC naproxen tablets dosed with and without a light breakfast. The study was carried out in seven male subjects (median age, 22 years) who were examined by a physician before the study and were judged to be in good health on the basis of medical history, physical examination, routine laboratory data, and standard electrocardiogram. Each subject gave written informed consent to participate in the investigation and the study was approved by the University of Nottingham Ethics Committee. Approval was obtained from the Department of Health, London, for the administration of radioactive substances. The study was conducted in accordance with the Declaration of Helsinki Guidelines for Ethics and Research and the Association of the British Pharmaceutical Industry guidelines for medical experiments in nonpatient healthy volunteers.

After an overnight fast volunteers took two tablets, either fasting or after a light breakfast of orange juice, two slices of toast, butter and preserve, and tea or coffee. Tablets were taken with 200 ml of water containing 1 MBq of technetium-99m-radiolabeled diethylenetriaminepentaacetic acid to enable an outline of the GI tract to be recorded. Anterior images of the abdomen, each of 1-min duration, were recorded using a gamma camera immediately after dosing, and at 10- to 15-min intervals throughout the morning. Imaging was at 15- to 30-min intervals in the afternoon and then hourly until 14 hr after dosing. A further image was taken at 24 hr. Tea or coffee was given 2 hr after the tablets, and lunch at 4 hr. Lunch consisted of two medium filled rolls, one packet of crisps, an apple or banana, and tea or coffee. Dinner was provided 10 hr after dosing. Water was allowed ad libitum between meals.

Blood samples were taken via an indwelling cannula, irrigated with heparin, and collected in normal biochemistry tubes. The tubes were gently mixed several times and allowed to stand for 30 min to allow clot formation. The blood was centrifuged and the serum transferred to polypropylene tubes prior to freezing at −80°C.

There were 7 days between each leg of the study. Naproxen was estimated in serum using high-performance liquid chromatography (HPLC). The internal standard and pH 4.5 phosphate buffer was added to a small volume of serum and then approximately 7 ml of extracting solvent. After separation, the solvent phase was evaporated to dryness and reconstituted in the HPLC mobile phase and injected onto the chromatograph. Detection was by UV monitoring of the effluent. The peak height ratios of drug to internal standard were related to a calibration graph to determine naproxen concentration.

RESULTS

The time of movement of the tablets from stomach to small intestine was taken as the midtime between recording the two images about the transition. *In vivo* disintegration was determined in an analogous manner. The GI transit and disintegration results for the EC tablets are presented in Tables I and II. A summary of the pharmacokinetic data is provided in Table III and a composite representation of the profiles is given in Figure I.

In Vitro Testing

Preliminary *in vitro* studies on the EC tablets demonstrated that the neutron activation procedure did not alter the release behavior of the formulation or the stability of the drug. The integrity of the film coat, before and after irradi-

Table I. Transit and Disintegration Profile of EC Naproxen Tablets (min) After an Overnight Fast

Volunteer no.	Gastric residence		Disintegration time (total postdose)		Disintegration time (after gastric emptying)	
	Tablet 1	Tablet 2	Tablet 1	Tablet 2	Tablet 1	Tablet 2
1	28	28	142	162	114	134
2	6	6	89	89	83	83
3	17	17	133	133	116	116
4	162	162	209	263	47	101
5	67	78	78	89	11	11
6	16	16	89	101	73	85
7	26	26	143	154	117	128
Mean	47		134		87	
SE	14		14		11	
Median	24		133		93	

Table II. Transit and Disintegration Profile of EC Naproxen Tablets (min) After a Light Breakfast

Volunteer no.	Gastric residence		Disintegration time (total postdose)		Disintegration time (after gastric emptying)	
	Tablet 1	Tablet 2	Tablet 1	Tablet 2	Tablet 1	Tablet 2
1	100	110	240	240	140	130
2	128	187	199	247	71	60
3	114	151	141	208	27	57
4	68	100	186	240	118	140
5	110	121	196	196	86	75
6	100	110	240	240	140	130
7	67	67	130	151	63	84
Mean	110		204		94	
SE	9		11		10	
Median	110		204		85	

ation, was confirmed using the BP disintegration test for EC tablets. All the tablets remained intact for 120 min after testing in 0.1 *M* hydrochloric acid but disintegrated within 30 min, after the acid medium was exchanged for mixed phosphate buffer (pH 6.8). *In vitro* dissolution studies demonstrated that no drug was released from the irradiated tablets following testing for 2 hr in 0.1 *M* hydrochloric acid. The tablets were then placed in pH 7.4 buffer for 45 min, and in all cases greater than 80% of the drug was released within the testing period. The clinical supplies, before and after irradiation, complied fully with the product specification for Naprosyn EC.

Gastric Emptying

No loss of tablet integrity was observed in the stomach. In the fasting state the tablets remained in the stomach for a median of 24 min (range, 6–162 min), compared to 110 min (range, 67–187 mm) in the fed state.

In the fasted subjects the emptying time showed a larger range than for the fed subjects. Both tablets emptied at virtually the same time following administration in the fasted state, whereas in the fed condition they tended to empty at different times.

Disintegration

All tablets disintegrated in the small intestine. Disintegration time postdose was a median of 134 min (range, 78–263 min) in the fasted subjects and 204 min (range, 130–247 min) in the fed state. Subtracting gastric residence times from disintegration times produced a median postemptying disintegration time of 93 min (range, 11–134 min) fasted and 85 min (range, 27–140 min) fed, assuming that the first tablet to empty was the first to disintegrate.

Table III. Summary of Pharmacokinetic Results

	Fasted	Fed
Time to appearance of naproxen in blood (mean hr)	2.5 (2.0–4.5)	3.5 (2.5–4.5)
T_{max} (median hr)	5.0 (2.5–12)	6.0 (4.5–8)
C_{max} (median μg/ml)	106 (65–156)	103 (90–121)

Naproxen Serum Levels

The median time to the first detectable naproxen levels was 2.5 hr (range, 2.0–4.5 hr) fasted and 3.5 hr (range, 2.5–4.5 hr) fed. Corrected for gastric residence times these figures reduce to 2.0 and 1.7 hr, respectively. The time to maximum blood levels (t_{max}) was difficult to evaluate, as a number of serum profiles showed no real peak (e.g., Volunteer 7). The median value was 5.0 hr (range, 2.5–12.0 hr) for the fasted state and 6.0 hr (range, 4.5–8.0 hr) for the fed state. Corrected for gastric residence times, these figures reduce to 3.9 hr (range, 2.2–11.6 hr) and 4.3 hr (range, 2.7–6.9 hr), respectively. Maximum naproxen concentrations were similar fasted and fed. No adverse events were reported during the study.

DISCUSSION

The EC naproxen tablets behaved as anticipated, with no loss of integrity in the stomach and all tablets disintegrating in the small intestine. As each dose consisted of two tablets, determination of exact disintegration times was made difficult when both tablets were in close proximity.

The effect of food on gastric residence time was as expected (7). Postprandial administration led to prolonged residence in the stomach and the time to onset of tablet disintegration was controlled predominantly by the gastric emptying time. The breakfast given was typical of that consumed by many people in the United Kingdom. All tablets had emptied before the volunteers ate lunch 4 hr after dosing. Residence time in the small intestine, prior to tablet disintegration, was independent of food intake but exhibited considerable inter- and intrasubject variation.

Under fasted conditions, the gastric emptying time of EC tablets is determined largely by the physiological process known as the migrating myoelectric complex (MMC), which occurs over a 2-hr cycle (16). The phases of the cycle range from a period of quiescence to strong contractions. It is the contractions in the third phase of the cycle that are important for gastric emptying, since they have the effect of "sweeping" indigestible material from the stomach through the open

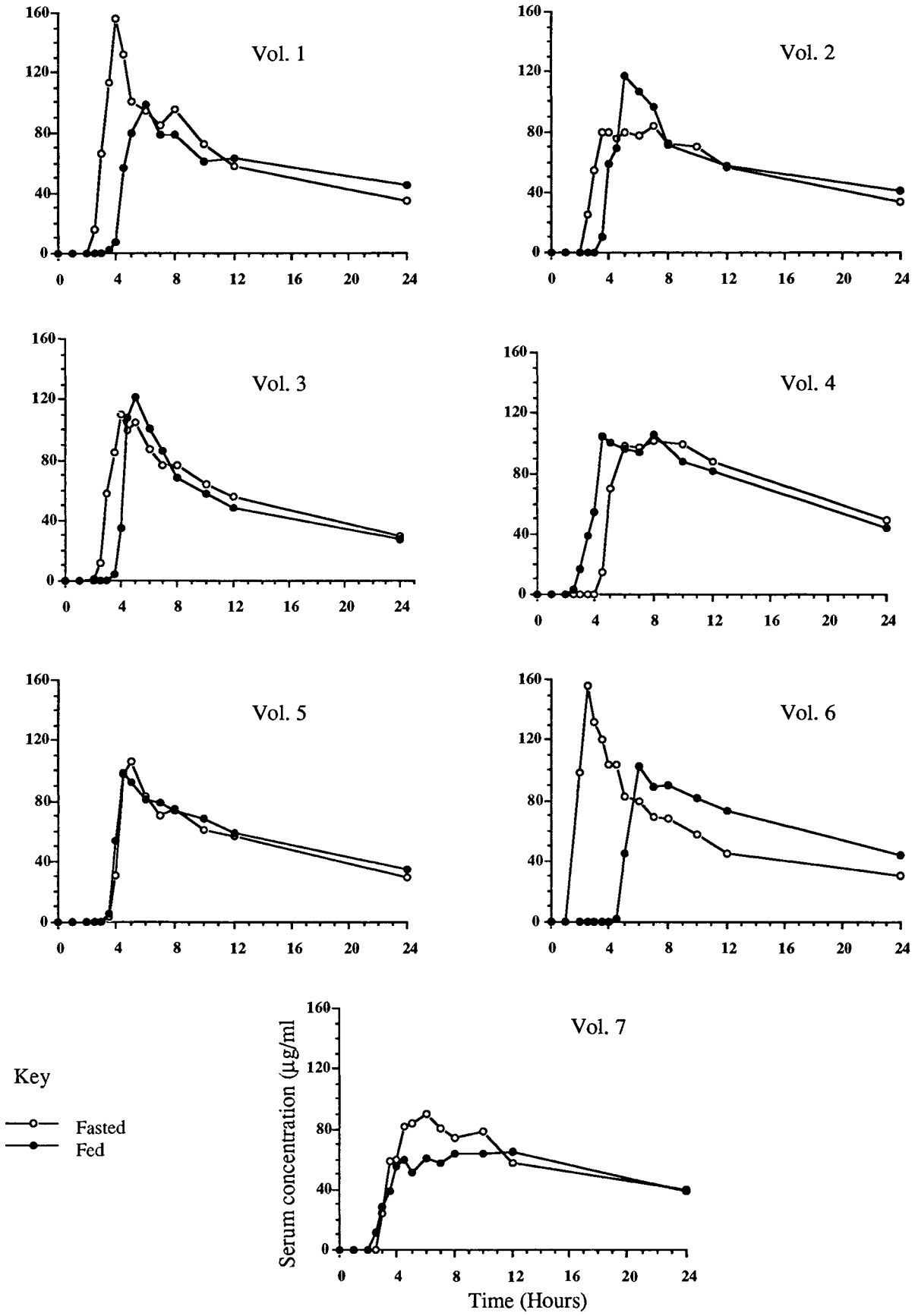

Fig. 1. Pharmacokinetic profiles—naproxen serum concentration vs time.

pylorus and into the small intestine (the so-called "housekeeper wave").

In the fasted state, large EC tablets can therefore empty from the stomach in an unpredictable manner, depending on their time of arrival in the stomach in relation to the contractual activity of the MMC (17). Recent combined scintigraphy and telemetry studies (18) have confirmed that large single-unit devices, such as a radiotelemetry capsule (RTC), are emptied from the stomach only by the large phase 3 contractions of the MMC. However, gastric emptying under the control of a "phase 3 process" is not always efficient. It has been suggested that the RTC (or in this study, the EC tablets) can remain in the less muscular body of the stomach and is not propelled into the antrum of the stomach from which emptying is likely to take place. This may explain the prolonged gastric residence observed for Volunteer 4 following administration in the fasted state.

It is interesting to note that the two tablets often emptied from the fed stomach at different times, with clear implications for drug absorption. Variable gastric emptying of large tablets, while the stomach is still in the digestive state, is not without precedent (19–22).

Overall the blood level results correlated well with the GI transit data. For fed and fasted subjects, gastric residence times differed by just over an hour. Lag time and t_{max} showed a similar difference. Therefore the delay in the appearance of naproxen in the blood when the tablets were taken with food was determined by the extra time the tablets remained in the stomach. Correcting t_{max} for gastric residence time gives a value slightly larger than would be expected with instant-release naproxen tablets. The mean peak plasma concentrations are in close agreement with reported data for EC naproxen tablets (22).

While the median results show an "expected" result, individual results show considerable variation. In a small number of volunteers it appeared that absorption started before disintegration. In these cases naproxen blood levels were low prior to observed disintegration. It is possible for drug release to occur from small cracks in the dissolving enteric coat before tablet disintegration occurs. Of more interest are the flat serum naproxen profiles seen in some volunteers. The profiles are similar to those seen with some sustained-release preparations which therefore make estimation of t_{max} difficult. There are a number of possible explanations for these flat curves, including the following. (i) There was a large interval between the disintegration times of the two tablets. The effects of this were not consistent. (ii) Rapid GI transit of the products of disintegration into the colon may have led to colonic absorption playing a significant role in the pharmacokinetic performance. It is likely that absorption of naproxen from the colon will be slower than that from the small intestine.

In conclusion, these EC tablets of naproxen behaved as predicted by *in vitro* tests and as might be expected from previous studies with large EC tablets (7,17). The tablets remained intact in the stomach and all reliably disintegrated in the small intestine. Overall, feeding caused a predictable delay in transit but large individual variation was observed. This is unlikely to be of any clinical significance for the treatment of chronic conditions with naproxen. However, the effect of food on t_{max} could be minimized by giving the tablets at night between dinner and breakfast, when there would almost certainly be sufficient time for the tablets to leave the stomach. The method for labeling the tablets proved a highly useful procedure. Tablets were manufactured and tested under normal conditions, giving confidence that the results of this study can be extrapolated to the commercially available tablets.

ACKNOWLEDGMENTS

This study was supported by a grant from Syntex Pharmaceuticals Ltd., Maidenhead, Berkshire, England. The authors are particularly grateful to the expert technical assistance in tablet manufacture supplied by Ewen Kinnear of the Scottish Syntex Research facilities based in Edinburgh.

REFERENCES

1. I. Haslock. Review of NSAID-induced upper gastrointestinal morbidity and mortality. In R. Cheli (ed.), *Royal Society of Medicine International Congress and Symposium Series 147*, Royal Society of Medicine, London, 1989, pp. 3–10.
2. F. L. Lanza. Endoscopic studies of gastric and duodenal injury after the use of ibuprofen, aspirin and other nonsteroidal anti-inflammatory agents. *Medicine* 77:19–24 (1984).
3. F. Halter. Mechanism of gastrointestinal toxicity of NSAIDs. *Scand. Rheumatol.* 73:16–21 (1988).
4. G. H. Griffiths, G. M. Owen, S. Kirkman, and R. Shields. Measurement of the rate of gastric emptying using chromium-51. *Lancet* 1:1244–1245 (1966).
5. M. Alpsten, G. Ekenved, and L. Solvell. A profile scanning method of studying the release properties of different types of tablets in man. *Acta Pharm. Suec.* 13:107–122 (1976).
6. D. L. Casey, R. M. Beihn, G. A. Digenis, and M. B. Shabhu. Method for monitoring hard gelatin capsule disintegration times in humans using external scintigraphy. *J. Pharm. Sci.* 65:1412–1413 (1976).
7. J. G. Hardy, J. N. C. Healey, S. W. Lee, and J. R. Reynolds. Gastrointestinal transit of an enteric-coated delayed-release 5-aminosalicylic acid tablet. *Aliment. Pharmacol. Ther.* 1:209–216 (1987).
8. S. S. Davis, N. Washington, G. D. Parr, *et al.* Relationship between the rate of appearance of oxprenolol in the systemic circulation and the location of an oxprenolol Oros 16/260 drug delivery system within the gastrointestinal tract as determined by scintigraphy. *Br. J. Clin. Pharmacol.* 26:435–443 (1988).
9. I. R. Wilding, S. S. Davis, C. D. Melia, *et al.* Gastrointestinal transit of Sinemet CR in healthy volunteers. *Neurology* 39 (*II Suppl. 2*):53–58 (1989).
10. C. G. Wilson, N. Washington, J. L. Greaves, *et al.* Bimodal release of ibuprofen in a sustained-release formulation: A scintigraphic and pharmacokinetic open study in healthy volunteers under different conditions of food intake. *Int. J. Pharm.* 50:155–161 (1989).
11. G. A. Digenis, E. P. Sandefer, A. F. Parr, *et al.* Gastrointestinal behaviour of orally administered radiolabeled erythromycin pellets in man as determined by gamma scintigraphy. *J. Clin. Pharmacol.* 30:621–631 (1990).
12. S. S. Davis, R. Khosla, C. G. Wilson, N. Washington, S. T. Leslie, and S. Malkowska. The gastrointestinal transit of a controlled release formulation of indomethacin. *Int. J. Pharm.* 60:191–196 (1990).
13. J. G. Hardy, G. L. Lamont, D. F. Evans, A. K. Haga, and O. N. Gamst. Evaluation of an enteric-coated naproxen pellet formulation. *Aliment. Pharmacol. Ther.* 5:69–75 (1991).
14. A. F. Parr, R. M. Beihn, R. M. Franz, G. J. Szpunar, and M. Jay. Correlation of ibuprofen bioavailability with gastrointestinal transit by scintigraphic monitoring of 171 Er-labeled sustained-release tablets. *Pharm. Res.* 4:486–489 (1987).
15. J. G. Devane, M. Kavanagh, S. S. Davis, R. A. Sparrow, and

I. R. Wilding. Correlation of GI transit parameters and pharmacokinetic characteristics of a controlled release nifedipine formulation. American Society of Clinical Pharmacology and Therapeutics, San Antonio, TX, 1991.

16. S. F. Phillips. Small bowel. In D. Kumar and S. Gustavsson (eds.), *An Illustrated Guide to Gastrointestinal Motility,* John Wiley and Sons, Chichester, 1988, pp. 187–206.
17. H. M. Park, J. M. Chernish, B. D. Rosenbek, R. L. Brunelle, B. Hargrove, and H. N. Wellman. Gastric emptying of enteric-coated tablets. *Digest. Dis. Sci.* **29**:207–212 (1984).
18. A. J. Coupe, S. S. Davis, D. F. Evans, and I. R. Wilding. Correlation of the gastric emptying of non-disintegrating tablets with gastrointestinal motility. *Pharm. Res.* 8:1281–1285 (1991).
19. R. Khosla, L. C. Feely, and S. S. Davis. Gastrointestinal transit of non-disintegrating tablets in fed subjects. *Int. J. Pharm.* **53**:107–117 (1989).
20. R. C. Khosla and S. S. Davis. The effect of tablet size on the gastric emptying of non-disintegrating tablets. *Int. J. Pharm.* **62**:R9–R11 (1990).
21. A. J. Coupe, S. S. Davis, and I. R. Wilding. Variation in gastrointestinal transit of pharmaceutical dosage forms in healthy subjects. *Pharm. Res.* 8:360–364 (1991).
22. J. G. Hardy, D. F. Evans, I. Zaki, *et al.* Evaluation of an enteric-coated naproxen tablet using gamma scintigraphy. *Int. J. Pharm.* **37**:245–250 (1987).