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Indirect gastrointestinal transit monitoring and absorption of theophylline

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Abstract

The gastrointestinal transit and absorption of a multiparticulate controlled-release theophylline preparation were investigated under fed and fasted conditions. Transit of the theophylline pellets in the gastrointestinal tract was monitored using two marker drugs, namely, paracetamol and sulfasalazine. Food was found to delay the gastric emptying and caecal arrival of the pellets, but had no significant effect on the small intestinal transit time. The delay in gastric emptying was associated with a delay in drug absorption. Under the fasted condition, approximately 13% of the drug was absorbed while the pellets were in the stomach, 57% while in the small intestine and the remaining amount while in the colon. As for the fed mode, the percent absorbed while the pellets were in these regions were 9%, 72%, and 19%, respectively.

Keywords: Theophylline pellets; Gastrointestinal transit monitoring; Paracetamol; Sulfasalazine/sulfapyridine

1. Introduction

Oral controlled-release preparations are designed to release their drug contents over an extended period of time. Hence, monitoring the drug absorption together with the gastrointestinal transit of such preparations can provide invaluable information in their design and development.

In recent years, gamma scintigraphy has emerged as a useful tool in gastrointestinal transit monitoring studies, but this technique requires an expensive gamma camera as well as the availability of suitable radionuclides. On the other hand, there are also indirect methods for monitoring gastric emptying and orocaecal transit time.

Absorption of paracetamol has been shown to be related to the rate of gastric emptying (Heading et al., 1973; Clements et al., 1978). This may be attributed to paracetamol being preferentially absorbed in the small intestine. Hence, the rate of

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appearance of the drug in the blood may be used as a measure of the gastric emptying rate. On the other hand, sulfasalazine when administered orally, is hydrolyzed by the flora of the large bowel to produce sulfapyridine and 5-aminosalicylic acid. Measurement of the absorbed sulfapyridine in the blood can then be used to determine the orocaecal transit time (Kellow et al., 1986; Staniforth et al., 1987).

Therefore, incorporating these two compounds into a dosage form may provide a cheaper alternative to gamma scintigraphy in monitoring its gastrointestinal transit behaviour. In the present study, these two drugs were used to monitor the gastrointestinal transit behaviour of a multiparticulate matrix controlled-release theophylline preparation (Peh and Yuen, 1995) in relation to the drug absorption. Both marker drugs were prepared as spherical pellets similar in size range and density to the preparation being investigated, and all three types of pellets were mixed and administered together in hard gelatin capsules.

2. Materials and methods

2.1. Materials

Microcrystalline cellulose (Avicel PH-101) was purchased from FMC Corporation. Paracetamol, Theophylline, Sulfasalazine, Sulfapyridine, β-hydroxyethyltheophylline were all purchased from Sigma. All other chemicals and reagents used are either analytical reagent (AR) or high performance liquid chromatography (HPLC) grade.

2.2. Pellet preparation

Both paracetamol and sulfasalazine pellets were produced using the same extrusion–spheronisation technique used in the preparation of the theophylline pellets (Peh and Yuen, 1995). The pellets were then sieved to give a size fraction of 1.18–1.70 mm diameter, being similar to that of the theophylline pellets. The density of all three types of pellets were determined using a gas multipycnometer (Quantachrome). The mean density values obtained ($n = 4$) were 1.354 ± 0.002 g/cm^3 for paracetamol, 1.283 ± 0.003 g/cm^3 for theophylline and 1.523 ± 0.007 g/cm^3 for sulfasalazine pellets.

2.3. In vivo study design

The study protocol was approved by an Ethics Committee. Twelve healthy non-smoking adult male volunteers between 29 and 43 years old (Mean = 36 years, SD = 6 years) and weighing from 49 to 78 kg (Mean = 66 kg, SD = 9 kg), participated in the study after providing written informed consent. All were judged to be healthy, were not receiving any medication during the study period, and had no history of any gastrointestinal disorder. The volunteers were randomly divided into two groups of six each, and administered the theophylline, paracetamol, and sulfasalazine pellets, which were mixed and filled in three size 0 hard gelatin capsules. The capsules contained an equivalent of 250 mg theophylline, 1 g paracetamol and 1 g sulfasalazine. During the first trial period, the first group was dosed after a 12-h overnight fast, while the second group was dosed immediately after a standard breakfast comprising two slices of toasted white bread with butter, two fried eggs, two strips of bacon, approximately 60 g of hash-browned potatoes, and 200 ml of whole milk (Skelly, 1984). After a 1-week washout period, the volunteers were then dosed under the alternate food condition. Food was withheld for at least 4 h after drug administration. Standard lunch and dinner comprising chicken with rice were served 5 and 10 h after dosing and water was given ad libitum. The volunteers were requested to abstain from alcohol and xanthine-containing food or beverages 24 h before and 36 h after the commencement of each study phase.

Venous blood samples of 5 ml volume were drawn via an in-dwelling cannula from the forearm into plain vacutainers at 0 (predose), 0.5, 1, 2, 3, 4, 6, 8, 10, 14, 18 and 24 h after drug dosing. Two additional blood samples were taken at 30 and 36 h by direct venipuncture. After standing for 2 h, the samples were centrifuged at 3500 rpm for 15 min, the serum transferred to separate glass containers and kept frozen until analysis.

2.4. Simultaneous measurement of theophylline, paracetamol and sulfapyridine

The serum concentrations of the three drugs were analyzed simultaneously using a reversed-phase HPLC method described by Yuen et al. (1996).

2.5. Analysis of serum data

The percentage absorbed versus time profiles of theophylline and paracetamol administered under fed and fasted conditions were calculated using the Wagner–Nelson method (Wagner and Nelson, 1964). The elimination rate constant (k_e) was estimated from the terminal slope of individual serum concentration curves by logarithmic transformation of the data and application of linear regression. In addition, the area under the serum concentration–time curve ($AUC_{0-\infty}$) of paracetamol was calculated by adding the area from time 0 to time t (AUC_{0-t}) and area from time t to infinity ($AUC_{t-\infty}$). The former was calculated using the trapezoidal rule, and the latter by dividing the last detectable serum concentration with the elimination rate constant.

2.6. Estimation of gastric emptying

Several parameters were estimated from the paracetamol absorption profile to describe the gastric emptying of the theophylline pellets. This was based on the assumption that the percent of paracetamol absorbed was directly related to the percent of pellets emptied from the stomach. The first parameter was lag time in emptying of the pellets (T_L), which was estimated using the lag time in paracetamol absorption. The second parameter was the time for 50% of pellets emptied (T_{50E}), estimated using the time for 50% of paracetamol absorbed and the third was the time for complete emptying of pellets (T_C), estimated using the time for 90% of paracetamol absorbed. In addition, the gastric emptying period (GE) was calculated from the difference between T_C and T_L.

2.7. Caecal arrival and small intestinal transit time

The caecal arrival time (CAT) was estimated from the serum sulfapyridine data and was defined as the time taken for the pellets to reach the caecum after ingestion. Some workers considered the time taken for the first appearance of sulfapyridine in the blood as the arrival of sulfasalazine at the caecum (Kennedy et al., 1979; Kellow et al., 1986; Staniforth et al., 1987; Sommers et al., 1990). This method may, however, overestimate the CAT, because there may be a time lapse for the sulfasalazine to be cleaved by colonic bacteria, prior to the release of sulfapyridine for absorption. Hence, in the present study, the CAT was estimated by interpolating the initial ascending portion of the serum concentration curve to the time axis. On the other hand, the small intestinal transit time (SIT) was calculated from the difference between the CAT and T_L.

2.8. Estimation of theophylline absorption at various regions of gastrointestinal tract

The absorption of theophylline in the various regions of the gastrointestinal tract (namely, the stomach, the small intestine and the colon) under the fed and fasted conditions was estimated from the absorption profile with reference to the distribution of the pellets in the gastrointestinal tract. The percentage absorbed while the pellets were in the stomach was estimated from the percentage absorbed at T_{50E}. On the other hand, the percentage absorbed while the pellets were in the small intestine was estimated from the difference of the percentage absorbed at T_{50E} and the percentage absorbed at T_{50S}. The T_{50S} was defined as the time in which the sulfapyridine concentration was one half the peak serum concentration. The percent of theophylline absorbed after this time point was deemed to have been absorbed while the pellets were in the colon.

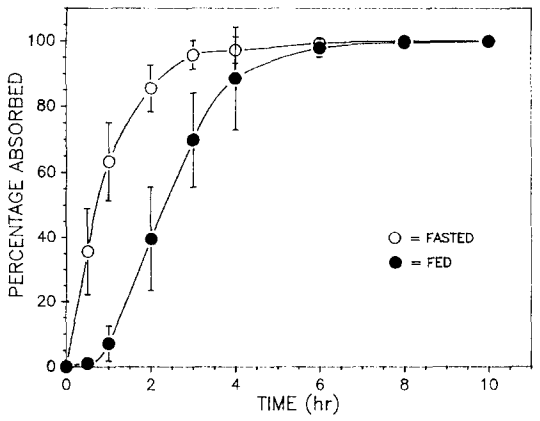

Fig. 1. Mean paracetamol absorption vs. time curves of the fed and fasted conditions. Mean ± SD, $n = 12$.

2.9. Statistical analysis

All the results are expressed as mean ± standard deviation, SD. The values of $AUC_{0-\infty}$, T_L, T_{50E}, T_C, GE, SIT, CAT and the percentage of theophylline absorbed in the stomach, the small intestine and the colon obtained under fed and fasted conditions, were analyzed using an analysis of variance (ANOVA) procedure which distinguishes effects due to group, subject/group, period and treatment (Wagner, 1975). A statistically significant difference was considered at $p < 0.05$.

3. Results and discussion

3.1. Gastric emptying

The mean paracetamol absorption–time curves obtained under fed and fasted conditions are shown in Fig. 1. No lag time in absorption was observed in the fasted curve. In contrast, a lag time in absorption was noted in the fed curve, resulting in the curve being shifted to the right.

The values of parameters $AUC_{0-\infty}$, T_L, T_{50E}, T_C and GE of fed and fasted states are given in Table 1. No statistically significant difference was observed between the fed and fasted logarithmic transformed $AUC_{0-\infty}$ values ($p = 0.3547$), indicating that the extent of paracetamol absorption was not affected by the different food conditions. The mean values of all the other parameters obtained under the fed condition were comparatively larger than those obtained under the fasted condition. A statistically significant difference was obtained between the fed and fasted values of T_{50E} ($p = 3.58 \times 10^{-6}$), T_C ($p = 1.23 \times 10^{-5}$) and GE ($p = 0.0001$), respectively. On the basis of these

Table 1
Individual numerical values of $AUC_{0-\infty}$, T_L, T_{50E}, T_C and GE

Subject	Fasted				Fed				
	$AUC_{0-\infty}$ (h·μg/ml)	T_{50E} (h)	T_C (h)	GE (h)	$AUC_{0-\infty}$ (h·μg/ml)	T_L (h)	T_{50E} (h)	T_C (h)	GE (h)
1	50.24	0.8	2.3	2.3	54.50	0.8	1.9	3.5	2.7
2	64.77	0.7	2.9	2.9	66.28	0.7	2.1	5.2	4.5
3	46.00	0.5	1.8	1.8	41.71	0.6	2.2	3.7	3.1
4	38.40	0.5	0.9	0.9	31.09	0.3	2.4	4.1	3.8
5	46.03	1.3	2.9	2.9	45.02	0.3	2.4	5.2	4.9
6	36.05	0.7	1.9	1.9	35.41	0.4	2.1	3.5	3.1
7	55.91	0.6	2.6	2.6	54.29	0.6	2.3	3.2	2.6
8	36.81	0.9	1.8	1.8	37.66	1.5	3.1	4.7	3.2
9	59.05	0.7	2.1	2.1	70.57	0.2	1.8	3.4	3.2
10	42.39	0.8	2.5	2.5	46.77	0.2	2.2	3.5	3.3
11	40.54	0.9	3.1	3.1	47.77	0.8	4.2	5.9	5.1
12	38.89	0.6	2.9	2.9	35.45	0.3	1.9	5.3	5.0
Mean	46.26	0.8	2.3	2.3	47.21	0.6	2.4	4.3	3.7
SD	9.40	0.2	0.6	0.6	12.35	0.4	0.7	0.9	0.9

results, it can be concluded that the gastric emptying pattern was markedly different under the fed and fasted conditions. Food not only delayed the onset of gastric emptying but also decreased the gastric emptying rate, resulting in a prolonged gastric emptying period of the pellets. These results are consistent with the findings of Yuen et al. (1993) who monitored pellets of approximately similar size range and density using gamma scintigraphy. Devereux (1987) also reported a significant increase in the fed values of T_{50E} and T_C but no statistically significant difference was observed between the fed and fasted values of T_L and GE. In the studies of Christian et al. (1980) and Davis et al. (1984), pellets dosed following a heavy meal were noted to empty more slowly compared to a light meal. Hence, although a different method was employed to study the gastric emptying process, the values obtained from the present study were found to be in good agreement with the findings of the above workers who used gamma scintigraphy.

The most common parameter used thus far to describe the gastric emptying process was T_{50E}. Devereux (1987) suggested that vast differences in the T_{50E} values between workers may be attributed to different physical properties of the pellets used for the study, such as size range and density. The pellets investigated by Devereux (1987) and Yuen et al. (1993) were closely similar to those studied here. Under the fed condition, Devereux (1987) reported a T_{50E} value ranging from 1.8–3.6 h, while Yuen et al. (1993) reported a range of 2.0–4.1 h, compared to 1.8–4.2 h obtained in the present study. For the fasted condition, the range reported by Devereux (1987) was 1.1–3.5 h and that by Yuen et al. (1993) was 0.1–2.6 h, compared to 0.5–1.3 h obtained here. Thus, the results obtained using the indirect method in the present study are comparable to those of the other workers.

3.2. Caecal arrival and small intestinal transit times

The mean serum sulfapyridine concentration–time curves of the fed and fasted conditions are shown in Fig. 2. In general, the curves obtained

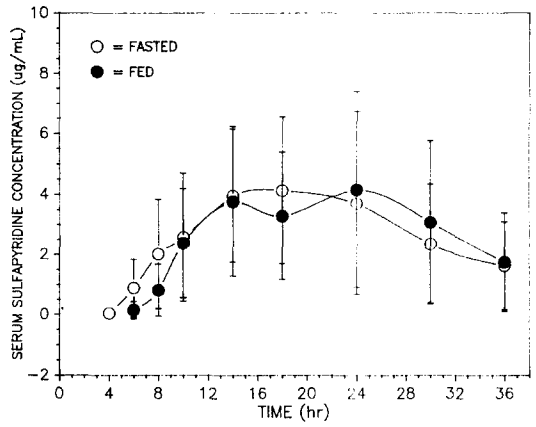

Fig. 2. Mean sulfapyridine concentration vs. time curves of the fed and fasted conditions. Mean ± SD, $n = 12$.

were not smooth, but characterized by plateau, sudden increase as well as decrease in the serum sulfapyridine concentrations. These features may be a result of a variable rate of sulfasalazine reaching the caecum or a variable quantity of sulfapyridine produced by caecal bacteria. Similar observations were reported by Staniforth et al. (1987) and Gramatte and Terhaag (1991).

The individual values of CAT and SIT of the fed and fasted conditions are presented in Table 2. There was a significant increase ($p = 0.0154$) in the mean CAT from 5.1 ± 2.0 h in the fasted condition to 6.5 ± 2.3 h in the fed condition. This is consistent with the findings of Davis et al. (1984) and Yuen et al. (1993).

The mean small intestinal transit times were 6.0 ± 2.3 h and 5.1 ± 2.0 h for the fed and fasted conditions. Although the mean fed value was comparatively larger, it was not significantly different statistically from the fasted value ($p = 0.0903$). Thus, the small intestinal transit time was not significantly affected by food, being consistent with the findings of Davis et al. (1984), Devereux (1987) and Yuen et al. (1993). Food was noted to cause a delay in gastric emptying, leading to a corresponding delay in caecal arrival, but the interval (small intestine time) between the two processes for both fed and fasted conditions was found to be rather consistent.

In the studies of Davis et al. (1984), Devereux (1987) and Yuen et al. (1993), the small intestinal

Table 2
Individual numerical values of CAT, T_{50S} and SIT

Subjects	Fasted			Fed		
	CAT (h)	T_{50S} (h)	SIT (h)	CAT (h)	T_{50S} (h)	SIT (h)
1	0.8	5	0.8	1.5	11	0.7
2	5.2	6	5.2	5.5	15	4.8
3	3.8	3	3.8	7.5	11	6.9
4	3.2	7	3.2	3.0	10	2.9
5	5.8	9	5.8	7.5	11	7.2
6	5.2	13	5.2	7.0	13	6.6
7	7.8	12	7.8	9.0	18	8.4
8	4.2	6	4.2	7.9	12	6.4
9	5.4	10	5.4	5.0	9	4.8
10	5.6	3	5.6	7.5	8	7.3
11	8.4	13	8.4	7.5	19	6.7
12	5.4	9	5.4	9.0	12	8.7
Mean	5.1	8	5.1	6.5	12	6.0
SD	2.0	4	2.0	2.3	3	2.3

transit time was obtained from the difference between the $T_{50\%}$ values for caecal arrival and gastric emptying. However, in the present study, the small intestinal transit time was described by the difference between the start of the caecal arrival and gastric emptying. As a result, the small intestinal transit times obtained in the present study were found to be relatively longer than those reported by Devereux (1987) and Yuen et al. (1993), for pellets of approximately similar size range and density. This could be attributed to a time lapse between the arrival of sulfasalazine in the caecum and the hydrolysis/absorption of sulfapyridine.

3.3. Absorption of theophylline in relation to the distribution of the drug pellets in the gastrointestinal tract

The bioavailability of the theophylline pellets under fed and fasted conditions was found to be comparable in a previous paper (Peh and Yuen, 1996). In this part of the present study, an attempt was made to relate the absorption of theophylline with the transit properties of the pellets in the gastrointestinal tract. Figs. 3 and 4 show the mean serum theophylline concentration and in vivo absorption profiles obtained under the fed and fasted conditions, together with the distribution of pellets in the gastrointestinal tract. The different shaded areas under the curves denote the estimated time the pellets spent in the stomach, the small intestine and the colon. The two boundaries represent the T_{50E} and T_{50S}, respectively. The T_{50S} was used instead of the CAT because the latter only represents the start of arrival of the pellets at the colon. Perhaps a better parameter may be the time for 50% sulfapyridine absorption. However, it could not be calculated because of insufficient data points, especially at the terminal portion of the serum sulfapyridine curve. Hence, the T_{50S} was chosen to approximate 50% sulfapyridine absorption. Individual values of T_{50E} and T_{50S} are given in Tables 1 and 2, respectively. The theophylline absorption was generally slow when a large proportion of pellets remained in the stomach, especially in the fed condition. An increase in the drug absorption rate can be observed when the pellets began to empty into the small intestine. In the small intestinal region, the drug absorption rate appeared to be comparable between the fed and fasted conditions, as demonstrated by the parallel nature of the fed and fasted absorption curves in this region (Fig. 4). Thus, the drug absorption rate in the small intestinal region was not affected by the presence of food.

Table 3 shows the estimated percentage of drug absorbed in the stomach, the small intestine and the colon under the fed and fasted conditions. In spite of a longer gastric residence time, the mean percentage of drug absorbed while the pellets were in the stomach was only 9% in the fed condition, being significantly lower ($p = 0.0292$) than the 13% in the fasted condition, where the gastric residence time was relatively shorter. This may be due to a reduced drug release/dissolution in the presence of food. In contrast, the mean percentage of drug absorbed in the fed mode was significantly higher ($p = 0.0097$) than that of the fasted mode when the pellets were in the small intestine, being 72% and 57%, respectively. The higher percentage of drug absorbed under the fed condition may be explained by the relatively longer residence time of the pellets in the small intestine

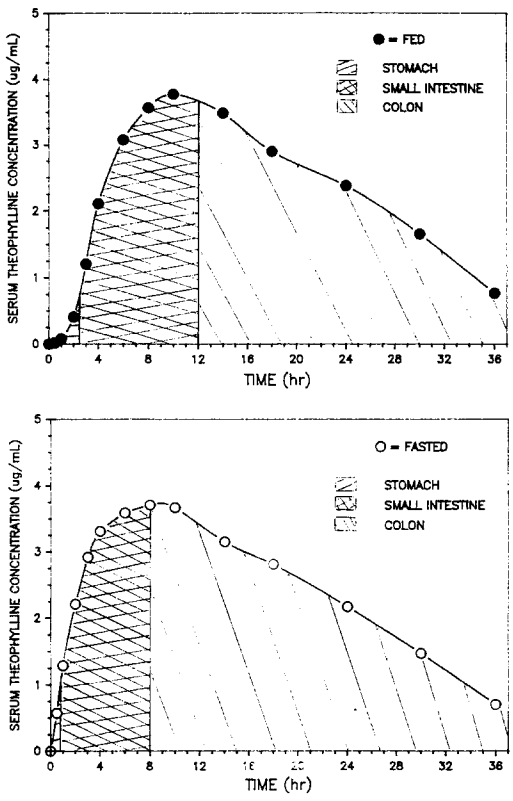

Fig. 3. Mean serum theophylline concentration vs. time curves of the fed and fasted conditions, together with the distribution of pellets in the gastrointestinal tract.

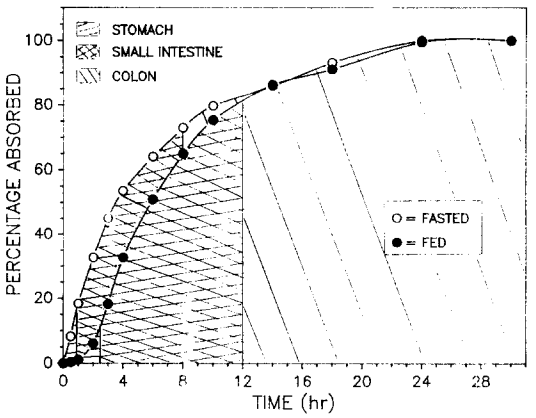

Fig. 4. Mean theophylline absorption vs. time curves of the fed and fasted conditions, together with the distribution of pellets in the gastrointestinal tract.

when dosed with food as shown in Table 2. Although the small intestinal transit time in the fed condition was not significantly different from that of the fasted condition, it was, on average, longer by approximately 1 h.

From the plots in Fig. 4 as well as Table 3, it can also be observed that a considerable amount of drug was absorbed while the pellets were in the colon. An average of 19% of the drug was absorbed in this region when the preparation was dosed with food and 29% when dosed in the fasted condition, being significantly different statistically ($p = 0.0247$). Other workers have also demonstrated that theophylline was absorbed in the colon when administered as controlled-release products. Sommers et al. (1990) and Yuen et al. (1993) found that approximately 40% of the drug was absorbed in the colon with the controlled-release theophylline preparations investigated.

4. Conclusion

In summary, the gastric emptying and caecal arrival of the pellets were significantly delayed by food while the small intestinal transit time remained rather consistent under the different food conditions. The results obtained using the indirect method appeared to be comparable to those of other workers who used gamma scintigraphy.

Table 3
Percentage of drug absorbed in the stomach, the small intestine and the colon

Subject	Fasted			Fed		
	Stomach (%)	Small intestine (%)	Colon (%)	Stomach (%)	Small intestine (%)	Colon (%)
1	14	44	42	1	84	15
2	20	44	36	14	76	10
3	14	49	37	7	71	22
4	12	54	34	14	57	29
5	12	66	22	7	65	28
6	10	84	6	6	81	13
7	16	70	14	5	90	5
8	14	63	23	5	83	12
9	12	61	27	8	59	33
10	19	27	54	18	59	23
11	9	61	30	12	67	21
12	9	65	26	14	70	16
Mean	13	57	29	9	72	19
SD	4	15	13	5	11	8

Moreover, theophylline absorption was found to occur while the pellets were in the colon, although highest absorption occurred while the pellets were in the small intestine.

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