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In vitro dissolution testing of oral controlled release preparations in the presence of artificial foodstuffs. I. Exploration of alternative methodology: microcalorimetry

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Summary

The possibility of using a microcalorimeter as an analytical method to monitor dissolution in complex media (to simulate fed and fasted patients) has been explored. The dissolution of Phyllocontin continus tablets (controlled release aminophylline, 225 mg) has been studied by conventional USP methodology under the following conditions: (i) buffer at pH 2.5; (ii) buffer at pH 2.5 followed by pH 5.6; (iii) halved tablet at pH 5.6; (iv) halved tablet in Ensure/buffer mixture at pH 5.6. The results showed dissolution to be independent of pH and almost independent of tablet integrity. The use of Ensure to simulate a fed patient produced a change in the first stage of the tablet dissolution kinetics. A microcalorimeter was used to monitor tablet dissolution with studies being undertaken on placebo and active tablets. Dissolution in buffer, buffer/Ensure, and buffer/Intralipid mixtures was studied. Where it was possible to effect comparisons it was clear that changes observed in the USP method were similar to those seen by microcalorimetry, the microcalorimetric results showing that Ensure altered the first, but not the second stage of dissolution kinetics, and that Intralipid altered the second but not the first stage. A major media-induced difference observed by microcalorimetry, was the timing of the change between the two processes. A tentative explanation of these results has been proposed. Microcalorimetry seems to be a useful tool by which dissolution into complex media (which due to the presence of factors such as colour and suspended matter cannot be used with conventional analysis techniques without labour intensive separation/dialysis techniques) can be monitored. The results provide an indication of mechanism of release/interaction. This could be a useful aid to formulators and could provide a convenient in vitro initial indicator of the effects of food on drug release, without recourse to expensive animal experiments.

Introduction

According to Ganderton (1987), in vitro dissolution testing requires "an enormous expenditure of technical effort using a test which is inherently variable, showing great sensitivity to the source material and the test methodology." "As a result, opinion on the value of the test varies from those who see the dissolution test as an effective predictor of the in vivo performance of solid dosage forms to those who regard it as no more than a crude indicator of batch to batch uniformity." This paradox has been arrived at because despite occasional high correlations between in vitro and in vivo release profiles there are also

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cases of almost zero correlation. It is often true that a zero correlation may be expected, for example when membrane transport rather than dissolution is the rate-limiting step. Equally, dissolution experiments are often carried out in only acidic conditions (or even worse when pH is not controlled) where many drugs show poor solubility compared to that observed at higher pH values; the significance of this may be expounded by the fact that most products move from the stomach to the intestine after a period of about 0.5–3 h (depending upon the patient and the extent of food intake). Most drugs are absorbed in the intestine rather than the stomach. Another possible reason for failed in vivo/in vitro correlations is that even when pH is controlled, and indeed when it is varied to 'mimic' elements of gastrointestinal transit, its in vitro composition is almost without exception unlike that which will be observed in vivo. The argument that is used to defend this position returns to the dissolution test paradox outlined above, that is that the in vitro dissolution test should be a simple, reproducible, official test and thus not complicated by diverse media. This is a view for which we have sympathy, particularly when the test is to be used in a quality control environment, and when results obtained from one laboratory are to be related to those obtained by another. The official dissolution tests must always be used as the standard to which reference should be made. The acceptance of official tests does not, however, preclude the use of unofficial dissolution tests which may well be of value to aid in understanding the drug release process(es).

One of the main uses of unofficial dissolution testing is to monitor release of sparingly soluble drugs which would exceed the requirement for maintenance of 'sink conditions', and are often not sufficiently soluble to fully dissolve in one litre of aqueous dissolution medium. One way around this problem is to use non-aqueous media or co-solvents to aid solubility; such methods have limitations which will not be discussed here. The second method often considered is the use of flow-through systems; here the volume of dissolution medium is theoretically infinitely large and need not be recirculated, thus preventing concentration build up. The most cited flow-through technique is that of Langenbucher (Langenbucher and Rettig, 1977). Ganderton (1987) summarized the current methodology relating to flow-through systems as follows: 'the capital costs are too high, flow may not be sufficiently uniform, test duration is too high and, above all, the test is very labour intensive'. The test that we will outline in this work, that is the use of microcalorimetry, is not open to all these criticisms; indeed test duration need not be long and the process is certainly not labour intensive. It should be noted that this work describes a novel use of a technique that has far wider application than has a dissolution apparatus; we would not necessarily suggest that a microcalorimeter should be purchased to perform dissolution experiments, but rather that the technique, which is the subject of considerable interest within the pharmaceutical academic and industrial worlds, could be so used for research and development purposes to yield information not currently available by simple in vitro methodology.

Within the scope of this work, the main advantages of the microcalorimeter are the ability to respond in the presence of complex media (i.e. colour and suspended matter will not interfere with the operation of the instrument), unlike traditional analytical methods (e.g. UV spectrophotometers) which require the removal of 'interfering' components; consequently this instrument can be used to investigate dissolution in the presence and absence of food, also the effect of individual components of food such as metal ions, fibre, fat, etc. can be studied for their effect on dissolution rate and the existence of drug/food interactions. Physical drug/drug interactions can also be studied either by admitting both drugs to the microcalorimeter in solution form, or by letting one product undergo dissolution in the presence of a solution of the other drug. This paper will deal with the study of dissolution rates of a commercially available aminophylline product (Phyllocontin continus) in the presence and absence of various simulated foodstuffs. The experimental procedure for study of drug/food and drug/drug interactions will be presented in subsequent submissions.

In its simplest form, the dissolution process is

described by the Noyes–Whitney relationship, which expresses the rate of change of concentration (c) with time (t) as a function of diffusion coefficient (D), surface area (A), saturation solubility (C_s) and the diffusion layer thickness (h), thus:

$$\frac{dc}{dt} = \frac{DS}{h}(C_s - c) \quad (1)$$

For 'sink conditions' C_s must be much larger than c (normally if c rises to more than 10% of C_s sink conditions are said to have been exceeded). It should be noted that c refers to the concentration of drug in solution, and thus does not account for drug which may have dissolved and been adsorbed to stomach contents or absorbed into the blood stream, thus the presence of food could remove drug from solution (lower c) and thus increase dissolution rate. It is also true that the diffusion coefficient in the Noyes–Whitney equation refers to the diffusion coefficient at the tablet surface, thus if fatty components of food adsorb to the tablet surface the value, and hence dissolution rate, will be different to the situation of dissolution in aqueous media. In terms of official dissolution tests, regulatory authorities have not looked favourably upon the use of adsorbents or partitioning processes. There does, however, seem to be a need for a predictive dissolution test to study the effect of food on drug release profiles as a forerunner to animal experimentation. If food is present during dissolution, the possibility for drug adsorption (i.e. drug/food interaction) is significant. Drug/food interaction can have two possible effects, it can either increase dissolution rate by decreasing the c term in Eqn. 1 thus possibly increasing absorption rate, or it can remove drug from solution in such a way as to reduce the rate and/or extent of absorption. There is clear value in obtaining an understanding of how drug release rate is altered in the presence of food and to be able to assess the magnitude of interaction if binding to foodstuffs does occur.

Historically, the choice of dissolution medium has largely been a compromise between some biological simulation and the need for simplicity and reproducibility. Finholt and Solvang (1968) demonstrated the value of adding a surfactant as a method of improving comparability to the in vivo situation, although it is clear that a concentration below that which will result in solubilisation is of value even this modification is seldom reported as having been used. Macheras et al. (1987) suggested the use of low-fat milk as a suitable food simulating dissolution medium, it was noted that the use of milk slowed the release rate of drug from certain controlled release theophylline preparations. This work used a complex dialysis method to extract the dissolved drug from the milk. The use of milk as a standard dissolution medium is perhaps a problem since despite quality control, and defined fat content, the nature origin may cause large variation in content. One alternative that has been used in the work presented below is a diluted Intralipid emulsion which is more standard in content than milk.

Materials and Methods

Conventional dissolution experiments were carried out using the USP paddle method at 37 °C stirring at 100 rpm. The pH was controlled using 'universal' buffer and was changed from 2.5 to 5.6 after 3 hours. The detail of the work and the results obtained from similar experiments have been reported previously (Buckton et al., 1988). The work described here relates to the controlled release aminophylline product Phyllocontin continus (aminophylline, 225 mg).

Attempts were made to use dissolution media in the USP paddle set up that would more closely resemble a fed patient, to this end use was made of Intralipid 10%, diluted 1 : 6 with universal buffer at pH 2.5 (changing to pH 5.6 after 3 h). Intralipid is an emulsion containing fractionated soya oil and glycerol. Further experiments were undertaken using Ensure diluted 1 : 6 of universal buffer. Ensure is a nasogastric foodstuff containing fat (3.7%), protein (3.7%), carbohydrate (14.5%), with minerals and vitamins. Experiments were carried out on whole and halved tablets. Placebo tablets of Phyllocontin were also studied.

It was not possible to separate Intralipid from the buffer, to allow the use of UV spectropho-

tometers, by methods of centrifugation, separation or precipitation; however, it was possible to remove Ensure by acidification and then centrifugation. We have not taken steps to demonstrate whether such a separation process will alter the concentration of free drug that is subsequently assayed.

As well as attempting to obtain dissolution profiles using complex media in the USP equipment, experiments were undertaken to study the dissolution process using microcalorimetry. The microcalorimeter (LKB 10700) measures heat changes that accompany reaction/interaction, giving an output of power as a function of time. The instrument is able to detect 0.1 μW (equivalent to 0.1 μJ/s). Assuming a moderate enthalpy change, it is probable that the microcalorimeter will be capable of monitoring the dissolution of the tablet (not just the drug) and any subsequent interaction that takes place between the product and the media.

The method of operation was to secure half a tablet in the cell of the calorimeter (volume of about 1.5 ml). It was necessary to halve the tablets in order to locate them in the cell. In future we propose to engineer a larger cell that will remove the need for halving the tablet. An inlet tube was positioned at the bottom of the cell and an outlet was available at the top. The outlet was fitted with a fine mesh to prevent the removal of undissolved material. This cell, which was where dissolution took place, was equilibrated to 37 ± 10^{-4} °C. The dissolution medium was passed from, and returned to, a one litre flask by use of a peristaltic pump. The one litre vessel was situated outside of the calorimeter and was thermostated to 37 ± 0.5 °C, stirring was by magnetic stirrer. Where possible (i.e. when just buffer was used as the dissolution medium) the outflow from the one litre vessel into the cell was passed through a flow-through UV spectrophotometer (Cecil) to allow assessment of rate of drug release to be compared with the calorimetric trace.

Results

USP dissolution studies

The rate of dissolution of Phyllocontin continus was not subject to change with alteration of pH. The results showed similar trends to those obtained for the Napp theophylline product Uniphyllin (Buckton et al., 1988), in that the dissolution in universal buffer followed a process described by two first-order regions separated by a mixed-order region; the transition was at about 100–120 min. The rate constants for the two first-order processes are presented in Table 1 for experiments at: (i) constant pH of 2.5; (ii) pH 2.5 followed by pH 5.6; and (iii) for halved tablets at pH 2.5. The results which are averages of at least 6 determinations, clearly show the process is not pH dependent and that the halving of a tablet only results in minor (almost insignificant) changes in release rate (as would be expected for such a matrix product).

TABLE 1

Apparent first-order rate constants for the dissolution of Phyllocontin continus in the USP dissolution apparatus

	First stage (min^{-1})	Second stage (min^{-1})
Buffer at pH 2.5	0.0174	0.0021
Buffer at pH 2.5 then 5.6	0.0173	0.0022
Halved tablet in buffer at pH 5.6	0.0215	0.0019
Halved tablet in Ensure buffer mixture at pH 5.6	0.0635	0.0023

Microcalorimetric dissolution studies

The microcalorimeter will detect the full dissolution process as well as any interactions that take place between the product and the dissolution fluid. To investigate the contribution of the tablet matrix, experiments were undertaken using placebo tablets. It was observed (Fig. 1) that the calorimetric output, expressed as deviation from a baseline response, was approximately 3 times the size for the active product compared to the placebo; thus the major part of the response was due to the drug dissolving.

It is likely that the rate constants obtained from the calorimetric experiments will be different to those obtained from the beaker experiments, as

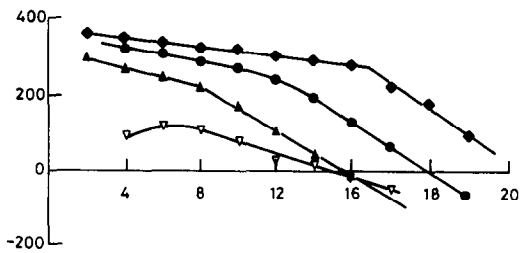

Fig. 1. Dissolution results obtained by microcalorimetry, presented as ln(deviation from basline) as a function of time. Axes: x = time (hours); y = ln(deviation). Dissolution of Phyllocontin continus tablets in: (◆) Intralipid/buffer mixture; (●) buffer; and (▲) Ensure/buffer mixture. Dissolution of placebo Phyllocontin continus tablets: ▽ buffer.

the flow method will provide a very different hydrodynamic regime. Equally, the method of calculation for the apparent rate constants from the calorimetric data is different to that used for the beaker method, in that the gradient of the ln(displacement from baseline) as a function of time plot has been used. It follows that direct numerical comparison of the results obtained from the two methods is not worthwhile; however, trends observed within any one method should be reproduced within the other.

The gradients of the two first-order sections of the ln(displacement) as a function of time plot are presented in Table 2 for Phyllocontin continus tablets with, diluted Ensure, diluted Intralipid, and buffer.

TABLE 2

Apparent first-order rate constants from the ln(displacement) as a function of time plots obtained from the microcalorimeter

	Apparent first-order rate constants		Time of onset of second stage (min)
	First stage (min^{-1})	Second stage (min^{-1})	
Placebo in buffer	–	0.41	540
Active in buffer	0.148	0.47	720
Active in buffer/Ensure	0.216	0.46	480
Active in buffer/Intralipid	0.129	0.61	1050

Discussion

Recently Macheras et al. (1987) have demonstrated (by a dialysis method) that milk seems to slow the dissolution of certain controlled release theophylline preparations. There is other evidence in the literature to support the findings of Macheras et al. (1987), for example, Pedersen and Steffensen (1986) studied the absorption of a single dose of a controlled release theophylline preparation (Somophyllin) in vivo and found that the mean time taken to reach mean serum level was 3.7 h in the fasted and 5.6 h in the fed children; it is possible that this delayed absorption in the presence of food is due to increased residence time in the stomach as a result of food. However, the other factor to be considered is that the release from the product maybe slower in the presence of food, thus compounding the effect of increased gastric residence time.

The problems with the method used by Macheras et al. (1987) are that milk is not a reproducible dissolution medium and the process of extracting all the released drug by dialysis presents difficulties; furthermore, interaction between the drug and the food will not be recorded. Drug/food interaction is important as dissolved drug which is complexed with gastric contents will not necessarily be available for absorption to the systemic circulation. Eshra et al. (1988) have reported an in vivo study that demonstrated that ketoprofen absorption is significantly reduced, both in terms of extent and rate, by the presence of food. The use of the microcalorimetric technique as an in vitro technique to assess interactions between drug and gastro-intestinal contents will be considered in a later publication.

The results presented here show certain differences in the dissolution performance of Phyllocontin continus when exposed to the different dissolution fluids. Changes in dissolution rate are most clearly observed over the initial first-order release period. This is a fact that is noted from both sets of results, viz. the beaker method where the sample tested in Ensure/buffer mixture dissoluted with an initial apparent rate constant of 0.0635 min^{-1} compared with an apparent rate of 0.0215 min^{-1} in buffer alone, whilst during the

second first-order process the rate constants were similar for both (approx. 0.002 min^{-1}); for the experiments carried out in the microcalorimeter the initial rate constant is far more variable with the different media than is the second rate constant (Table 2). It is also true that the largest change in rate due to halving the tablets was observed during the first of the two apparent first-order processes. The results suggest that the initial depot of drug that dissolves is either poorly controlled by the matrix (compared to the second release depot) or else the first dissolution process is more susceptible to changes in surface area and the presence of food (media composition). The continus controlled release system consists of two components, an aliphatic alcohol and a hydroxyalkylcellulose. Drug release is obtained by dissolution of the aliphatic alcohol and diffusion through hydration of the hydroxyalkyl cellulose, it is probable that these two components of the dissolution process produce the two first-order release profiles and that it is the dissolution of the alcohol that is most susceptible to the dissolution fluid composition and tablet integrity.

The calorimetric data shows that not only does the initial apparent first-order release rate alter as a function of the presence of different simulated foodstuffs, but also the time taken for the second apparent first-order process to start is considerably different. This may be due to a hindering of diffusion onset by preventing adequate access of an aqueous environment to the surface, either due to the composition of the fluid or due to the changes exhibited in the dissolution of the alcohol component. It is interesting that only the experiments carried out using Intralipid show a marked change in release rate during this second rate process (Table 2), indicating that different dissolution fluids can alter the release by different mechanisms.

Attention is drawn to the comments made in the introduction relating to the Noyes–Whitney equation, particularly with respect to local effects produced by adsorption of components of the dissolution media to the tablet surface and subsequent alterations in diffusion.

Conclusions

Phyllocontin continus was selected for study because the release profile is not pH dependent, thus simplifying the experimental considerations.

Data in the literature show that oral controlled release bronchodilator products can produce different blood levels when taken with and without food. Such results are probably due, in part, to reduced gastric transit time perhaps slowing drug access to intestinal absorption sites, although dissolved drug (as liquid) should pass from the stomach fairly easily. The other possible reason for changes in drug absorption is the influence of gastrointestinal contents on drug release and availability.

In this work it can be seen, by use of in vitro methodology, that different (artificial) foodstuffs can alter the release profiles from a controlled release product in a variety of ways. The Napp continus dosage forms are known to release by dissolution of an aliphatic alcohol and diffusion through hydration of a hydroxyalkylcellulose. It is tempting to suggest, for example, that Ensure aided the dissolution of the aliphatic alcohol in the product, thus increasing the initial dissolution rate, but did not alter the diffusion of drug from the hydroxyalkylcellulose. Conversely, Intralipid seems to have slowed the initial and increased the subsequent dissolution processes. Furthermore the time of onset of the second (perhaps the diffusion) process varied from 480 to 1050 min when tested in the various fluids, from this observation it can be argued that Ensure, by aiding the initial dissolution process, has allowed more rapid hydration of the cellulose component and thus more rapid onset of (but not rate of) release from the second component. Intralipid could have adsorbed to the product thus slowing the hydration, and thus onset of the release from the cellulose; however, when diffusion started the rate constant suggests that the presence of the Intralipid aided the release process.

Such in vitro methodology will not replace in vivo testing, but may be of value in understanding poor in vitro/in vivo correlations and in providing

a way of anticipating problems that occur if the product is taken with food.

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