Research Paper

Theme: Facilitating Oral Product Development and Reducing Regulatory Burden through Novel Approaches to Assess Bioavailability/Bioequivalence
Guest Editors: James Polli, Jack Cook, Barbara Davit, and Paul Dickinson

An Investigation into the Utility of a Multi-compartmental, Dynamic, System of the Upper Gastrointestinal Tract to Support Formulation Development and Establish Bioequivalence of Poorly Soluble Drugs

Paul A. Dickinson,^{1,3} Ragheb Abu Rmaileh,² Lee Ashworth,² Richard A. Barker,² Wendy M. Burke,¹ Claire M. Patterson,² Nick Stainforth,² and Mohammed Yasin²

Received 25 January 2012; accepted 13 February 2012; published online 29 February 2012

Abstract. In recent years mechanical systems have been developed that more closely mimic the full dynamic, physical and biochemical complexity of the GI Tract. The development of these complex systems raises the possibility that they could be used to support formulation development of poorly soluble compounds and importantly may be able to replace clinical BE studies in certain circumstances. The ability of the TNO Simulated Gastro-Intestinal Tract Model 1 (TIM-1) Dynamic Artificial Gastrointestinal System in the 'lipid membrane' configuration to support the development of Biopharmaceutics Classification System Class 2 compounds was investigated by assessing the performance of various AZD8055 drug forms and formulations in the TIM-1 system under standard fasting and achlorhydric physiological conditions. The performance data were compared with exposure data from the phase 1 clinical study. Analysis of the AZD8055 plasma concentrations after tablet administration supported the conclusions drawn from the TIM-1 experiments and confirmed that these complex systems can effectively support the product development of poorly soluble drugs. Particularly, the TIM-1 system was able to show that AZD8055 exposure would increase in an approximately dose proportional manner and not be limited by the solubility or dissolution. Additionally, the investigations also showed that the exposure produced by a solution and a tablet would be the same. Specific instances when the TIM-1 system may not be predictive of clinical product performance have also been identified.

KEY WORDS: BCS; biopharmaceutics; biorelevant dissolution; bridging; replacement.

INTRODUCTION

The TNO Simulated Gastro-intestinal Tract Model 1 (TIM-1) is a multi-compartmental, dynamic, computer-controlled system that simulates the human upper GI tract (1,2), which was developed at TNO Nutrition and Food Research (Zeist, The Netherlands) (3). Initial applications of the TIM-1 system were in the field of nutrition including

the availability/absorption of minerals (4), vitamins (5) and food mutagens (6) in humans; the survival of ingested bacteria (7) and yeasts (8); and also bioavailability of heavy metals from soil (9).

The TIM-1 system has attracted interest as the most physiologically relevant dissolution apparatus for use in drug development (10). This is because this *in vitro* system closely simulates all of the *in vivo* dynamic physiological processes that occur within the lumen of the stomach and small intestine of humans, including secretion of enzymes, co-factors and bile salts in physiological amounts; appropriate pH control using physiologically relevant buffers; appropriate mixing and physiological transit times for each step of digestion and uniquely removal (absorption) of the products of digestion. In drug development, the study of digestion becomes the study of dissolution with the additional benefit that the removal of the dissolved drug is analogous to absorption and thus drives further dissolution that would occur *in vivo* due to drug permeation but does not occur in simpler biorelevant dissolution apparatus with fixed volumes (such as USP2). Additionally in the

¹ Clinical Science, AstraZeneca R&D, Alderley Park, Macclesfield, UKSK10 4TG.

² Pharmaceutical Development, AstraZeneca R&D, Alderley Park, Macclesfield, UK.

³ To whom correspondence should be addressed. (e-mail: paul.dickinson@astrazeneca.com)

ABBREVIATIONS: GI, Gastro-intestinal; TNO, Nederlandse Organisatie voor Toegepast Natuurwetenschappelijk Onderzoek (Netherlands Organization for Applied Scientific Research); TIM-1, TNO Simulated Gastro-Intestinal Tract Model 1.

TIM-1 system, the main parameters of interest in drug product behaviour (pH, transit time, gastric, biliary and pancreatic secretions) can be controlled to mimic various human GI (patho-)physiologies including age, different food intakes and pathological conditions (such as achlorhydria or altered GI transit such as in diabetes). Also the TIM-1 system as with other *in vitro* techniques (11) offers advantages over *in vivo* studies such as accuracy, reproducibility (no biological variation) and relatively easy manipulation.

Despite these potential advantages, there are limited reports of the successful use of the TIM-1 system in the pharmaceutical field and even fewer reports that systemically documented its utility in drug product development (3,12–14). These reports have concerned highly soluble drugs (Biopharmaceutics Classification System (BCS) Class 1 compounds: paracetamol/acetaminophen and paroxetine hydrochloride) or 5-aminosalicylate which has a good absolute solubility (>1 mg/mL) (15), although the impact of controlled or timed-release formulation and prandial state have been investigated (13,14). However, for TIM-1 system to become a maximally valuable tool in pharmaceutical development, its ability to support the development of poorly soluble (BCS2 and 4 class) drugs which make up the majority of the Pharmaceutical Industry's current development portfolio needs to be investigated and confirmed.

A systematic approach to using the TIM-1 system to aid the development of BCS2 and 4 products would address the ability of the TIM-1 system to support the following activities:

1. Formulation selection/clinical performance evaluation including the likely clinical performance of standard and bio-enhanced formulations and dose linearity
2. Formulation bridging and bioequivalence
3. Physical form evaluation
4. Prediction of food effects
5. Formulation performance in disease state
6. Quality by design and IVIVC activities to allow design space definition (16)

To date, the application of TIM-1 in the pharmaceutical field has utilised the 'dialysis membrane' set up for the removal of dissolved drug and a relatively unusual gastric pH and emptying profile. These gastric conditions are most appropriate for a Dutch population consuming dairy products

(pH ~4, gastric emptying half-life=30 min) and thus are not representative of the standard fasted healthy volunteer population generally used in bioequivalence and phase 1 studies. The use of the dialysis to remove dissolved drug also raises the potential that, for poorly soluble drugs, dissolved drug removal could be become limited by saturation of the dialysis media since it is run in a closed configuration (digestion products tend to be water soluble and so this not a concern when studying digestion). This saturation does not occur *in vivo* due to plasma protein binding and drug distribution/clearance. However TNO have developed a novel lipid membrane configuration in which the dissolved drug is removed by filtration through 50-nm pore filter at a predetermined filtration rate, which has been used successfully in the investigation of lipid digestion (17). This configuration would seem to offer several potential advantages over the dialysis membrane for poorly soluble drugs including:

1. No risk that the dialysis media become saturated, thus the concentration gradient for dissolution is maintained akin to the *in vivo* situation
2. Easy ability to change the 'effective permeability/absorption rate' of the drug to that expected in the clinic by increasing or decreasing the filtration rate
3. The filtration approach is mechanistically related to USP4 dissolution apparatus and thus insights gained in the TIM-1 system may facilitate the development of more quality control type methods in USP 4 apparatus.

A comparison of the different standard operating conditions for TNO TIM-1 in the dialysis and lipidic membrane configurations is given in Table I, and a schematic and description of the TIM-1 lipid membrane set up is shown in Fig. 1. Settings not listed in Table I are identical between the two configurations.

The objective of this report is to start to investigate systematically whether TNO TIM-1 system in the lipid membrane configuration can be used to support the pharmaceutical development of poorly soluble compounds. To this end, this initial publication covers the use of TIM-1 to assess:

- The likely dose proportionality of a poorly soluble weak base
- The choice of first time in man formulation

Table I. Comparison of the Difference Between the TIM-1 Model Operating in the Lipid Membrane Set up and Dialysis Membrane Set up

	Lipid membrane configuration	Dialysis membrane configuration
Membrane type	Microfiltration	Dialysis
Pore size/mol.wt. cut off	50 nm	~10 kDa
Duodenal secretion ^a	Porcine whole bile 20% v/v (fasted) and 100% v/v (fed)	Porcine bile extract powder 0.8% w/v (fasted), 4% w/v (fed)
Typical filtration/dialysis flow rate	3.9 mL/min	10 mL/min
Jejunal secretion	Small intestinal electrolyte solution ^b containing 2% (fasted) 10% (fed) pig bile ^c	Not required
Ileal secretion	Small intestinal electrolyte solution ^d	Not required

^a Not representative of luminal composition which is dynamic throughout the experiment

^b Sodium chloride, 5 g/L; potassium chloride, 0.6 g/L; and calcium chloride di-hydrate, 0.30 g/L

^c To allow for filtration of fluid and bile

^d To allow for filtration of fluid and mimic bile salt absorption in ileum

TNO TIM-1 System Schematic

A	Stomach Compartment
B	Peristaltic Valve
C	Duodenum Compartment
D	Jejunum Compartment
E	Ileum Compartment
F	Pressure Sensor
G	Stomach Secretion
H	Duodenum Secretion
I	Jejunum Secretion
J	Ileum Secretion
K	Pre-Filter
L	Semi-Permeable Membrane
M	Filtrate Pump
N	pH Electrode
O	Level Sensor
P	Temperature Sensor
Q	Dosing Port

Fig. 1. Schematic diagram and description of TNO TIM-1 apparatus in lipid membrane set up. Each TIM-1 compartment is formed by two connected basic units consisting of a glass jacket with a flexible silicone internal wall. Water is pumped into the glass jacket to maintain physiological temperature (37°C) and to control the pressure on the flexible walls. Variation of the water pressure enables mixing of the contents (chyme) by alternate compression and relaxation of the flexible walls to simulate peristalsis. Valves separate the compartments and regulate the rate of emptying of chyme from one compartment to the next. The rate of transit can be varied depending on the physiological state being simulated. Predetermined pH profiles are maintained in each compartment by automatic secretion of acid or bicarbonate as required, and biorelevant secretions of pepsin, lipase, electrolytes, pancreatin and bile salts are introduced by computer-controlled pumps. Hollow fibre filtration membranes are fitted to the jejunal and ileal sections. Ileal and jejunal secretion are continuously pumped through these membranes at a rate of 3.9 mL/min, and the filtrate is collected and analysed for filtered drug (in solution) and hence available for absorption. The total run time of an experiment is 6 h. Material collected from the ileal effluent tube is analysed and considered to have been delivered to the colon and not available for absorption. At the end of the experiment, all the residues remaining in the model are removed and analysed for mass balance purposes. The output from TIM-1 is cumulative percent of administered dose available for absorption (mass of drug in jejunal and ileal filtrate) with respect to time

- The potential for variability under conditions of high gastric pH ($\sim\text{pH } 5$) such as commonly seen in some elderly populations (18), those with *Helicobacter pylori* infection or those taking proton pump inhibitors or other medications that affect gastric pH
- The need for a salt of the weak base
- And finally, to support the bridging from an early clinical solution formulation to a later development tablet.

The data from the TIM-1 system will be compared with the phase 1 clinical exposure data for the same compound. Where the preclinical safety profile for a compound does not support its dosing to healthy volunteers, for example often those intended for the treatment of cancer, the TIM-1 system could be particularly valuable in supporting the compound's

development as any clinical pharmacokinetic assessment of formulation would have to be performed in patients raising numerous ethical, logistical and time concerns.

The chosen test compound, AZD8055, was predicted to have less than dose proportional increases in exposure at high doses (>300 mg) due to solubility limited absorption. A TIM-1 study was initiated to assess the potential for non-linear exposure from a simple pH adjusted solution (10 mg/mL) of AZD8055 at doses of 200 and 500 mg under standard fasted conditions. A suspension formulation, representative of a 'best-case' tablet formulation (i.e. a tablet with instantaneous disintegration and dispersion), was also tested at the same doses to risk assess the future formulation switch to a solid dosage form. Additionally, the 200 mg suspension was run under achlorhydric conditions (gastric pH 5) with rapid gastric emptying ($t_{1/2}$ 20 min) to assess the potential for *in*

in vivo variability resulting from the pH sensitive solubility of this compound.

The fumarate salt of AZD8055 was also evaluated as a potential mitigation strategy for poor performance identified in any of the aforementioned conditions.

MATERIALS AND METHODS

AZD8055 is a weak base with a pK_a of 6.2 and $\log D_{7.4}$ of 3.2. As such, the compound showed greater solubility in acidic media (13 mM in simulated gastric fluid at pH 1.7) compared with intestinal media (17 μ M in pH 6.8 phosphate buffer). Based on the predicted efficacious dose (325–3,325 mg twice daily) AZD8055 was classified as ‘low solubility’ according to the Biopharmaceutics Classification System (BCS) (19). Solubility was slightly improved (71 μ M) in aspirated human intestinal fluid and fasted simulated small intestinal fluid at pH 6.5 (39 μ M) owing to the presence of bile components. Intestinal permeability was deemed high based on *in vitro* epithelial cell culture methods (MDCK, Caco 2), giving an overall tentative BCS classification of 2.

Unless otherwise stated, reagents were supplied by Sigma-Aldrich Company Ltd, Dorset UK. Pig bile (Hendrix Slaughter House) and pancreatin (Pancreatic Enzyme for Cats and Dogs 250 g Tub, Pfizer Animal Health) was supplied

by TNO, Zeist, NL. Trifluoroacetic acid was supplied by Alfa Aesar, MA, USA. Lipase (Rhizopus lipase F-AP 15) was supplied by Amano Pharmaceuticals, Chipping Norton, Oxfordshire, UK

TNO TIM-1 Experiments

The TNO TIM-1 System Description

Each of the four compartments has a pH probe to automatically measure the pH of the solution in that compartment (see Fig. 1). The computer is able to administer a set pH curve by secreting either water or 1 M HCL into the Stomach, or either water or 1 M NaHCO_3 into the duodenum, jejunum and ileum.

The TIM-1 system was run in a ‘lipid membrane’ configuration, using microfiltration as the separation technology to mimic absorption of dissolved/solubilised drug. The filtrate pump was set to 3.9 mL/min, which produces a borderline ‘high permeability’ condition (inferred from the fact that ~80% of a freely soluble compound would be ‘absorbed’ over the course of an experiment at this flow rate, see the ‘Discussion’).

All experiments in the present study were run under fasted conditions where media and machine parameters were set to mimic the fasted state.

Fig. 2. Graph of cumulative appearance of AZD8055 in jejunal and ileal filtrates *versus* time after dosing of 200 and 500 mg AZD8055 free base as a solution and suspension. $N=1$, except 200 mg solution; $N=3$, mean $\pm$ SD

TNO TIM-1 Media

All working solutions are prepared on the day of use. All solutions are made with de-ionised water unless specified.

In the stomach compartment, fasting gastric start media comprising: 202 mg sodium chloride, 72 mg potassium chloride, 39 mg sodium bicarbonate and 10 mg calcium chloride di-hydrate, 0.01 M pH 7 citrate buffer, lipase (37.5 units) and pepsin (600 units). The required pH adjustment (to pH 2 or 5) was made using 1 M HCl and made to 300 g with potable water to represent the water volume ingested by the patient at the time of dosing.

The duodenum and jejunum contain approximately 60 and 120 g, respectively of an aqueous solution containing 1.25 g/L sodium chloride, 0.15 g/L potassium chloride and 0.075 g/L calcium chloride di-hydrate, 3.5 g/L pancreatin and 10% w/w bile. The ileum contains 120 g of aqueous solution containing 5.0 g/L sodium chloride, 0.6 g/L potassium chloride and 0.30 g/L calcium chloride di-hydrate.

TNO TIM-1 Sampling

Samples were collected at 60, 120, 180, 240, 300 and 360 min from three locations and assayed for AZD8055: the fluid that exits the ileal compartment ('effluent' analogous to drug that

would enter the colon and assumed not to be available for absorption) and the jejunum and ileal filtrates (analogous to dissolved and permeated drug that would enter the systemic circulation, known collectively as the 'bioaccessible fraction'). The entire volume from each sampling period was collected, weighed, sampled and diluted with an equal volume of acetonitrile for subsequent analysis. At the end of the experiment residual fluid remaining in the TIM-1 compartments was collected and analysed for AZD8055. The compartments were further washed and the washings assayed for AZD8055.

HPLC Analysis of AZD8055

AZD8055 concentration in the TIM-1 media, residual fluid and washings, diluted with an equal volume of acetonitrile (to precipitate protein and ensure filtered drug is in solution), was assayed by HPLC utilising an Agilent 1100 HPLC system (Agilent Technologies, Inc, CA, USA) and a Luna C18 (2) 3- μ m 4.6 $\times$ 50-mm column (Phenomenex, CA, USA) and gradient elution with a mobile phase consisting of water, acetonitrile and 0.1% v/v trifluoroacetic acid. AZD8055 concentrations were quantified against standards of known concentration by UV absorbance at 254 nm.

Fig. 3. Graph of effect of gastric pH on the cumulative appearance of AZD8055 in jejunal and ileal filtrates *versus* time after dosing of 200 mg AZD8055 free base as suspension. $N=1$ for gastric pH 2; $N=2$, mean $\pm$ range for gastric pH=6

TNO TIM-1 Experiments

The TIM-1 study was performed in four stages to assess the following:

1. The likely dose proportionality of this poorly soluble weak base from a simple solution (intended for early clinical studies) and a suspension of the free base intended to replicate the performance of a best case tablet (i.e. a tablet with instantaneous disintegration and dispersion).
2. The potential for variability under conditions of high gastric pH (~pH 5) and rapid gastric emptying for the solution and suspension.
3. The need for a salt of the weak base.
4. Bridging required to change from an early clinical solution formulation to a later development tablet.

Unless otherwise stated starting stomach pH was 2, and this was maintained throughout the experiment and gastric emptying half-life was 30 min.

1. Dose proportionality—a solution of the AZD8055 in unbuffered pH adjusted water (pH 4) was prepared at

10 mg/mL. The suspension was prepared at 10 mg/mL free base in HPMC/Polysorbate 80 (0.5% w/v /0.1% w/v). Dose volumes of 20 and 50 mL were introduced to the gastric compartment to give total doses of 200 and 500 mg, respectively. The volume of water added to the gastric start media was adjusted to maintain total gastric starting contents of 300 g for each study.

2. Impact of high gastric pH—the experiment was performed using the solution and suspension dosed at 200 mg as described in the dose proportionality experiment, except that gastric start media was adjusted to pH 5 and HCl secretion into the stomach was prevented throughout the experiment, gastric emptying half-life was also reduced to 20 min.
3. Salt evaluation—the experiment was performed using a suspension of AZD8055 fumarate (10 mg/mL in HPMC/Polysorbate 80 (0.5% w/v /0.1% w/v) dosed at 200 mg free base equivalents as described in the high gastric pH experiment.
4. Bridging solution to tablet—20 and 100 mg tablets of AZD8055 fumarate were introduced to the gastric compartment at doses of 120 or 240 mg free base equivalents. Evaluation was carried out under

Fig. 4. Graph of effect of gastric pH on the cumulative appearance of AZD8055 in jejunal and ileal filtrates *versus* time after dosing of 200 mg AZD8055 free base or AZD8055 fumarate as suspension. $N=1$ for base gastric pH 2 and fumarate pH 5; $N=2$, mean $\pm$ range for base gastric pH=5

normal conditions (gastric pH of 2; half-life, 30 min) as well as under the high pH conditions (pH 5; half-life, 20 min)

CLINICAL STUDY

AZD8055 was dosed to patients in a phase I open label, ascending dose clinical trial (NIH registry: NCT00731263) (20). Cohorts of three to six patients received a single dose of AZD8055 followed 1 week later by continuous, twice-daily dosing. Dose levels of 10 mg up to 120 mg were evaluated. Initially, a solution formulation was used at the 10-, 20- and 40-mg dose level. The formulation was switched to the tablet formulation as soon as it became available and was evaluated at dose levels of 40, 60, 90 and 120 mg. Plasma samples were collected for pharmacokinetic analysis following the single dose and where possible on the 29th day of multiple dosing.

RESULTS

TNO TIM-1 Experiments

The results of the dose proportionality study are shown in Fig. 2. Both formulations produced high bioaccessible fractions and no difference was detected between the fraction

available for absorption from a solution or suspension at 200 mg. Similarly, no difference was detected between the two formulations at the 500 mg dose or indeed between the 200 and 500 mg doses. The results of the impact of gastric pH on bioaccessible fraction are shown Fig. 3. Under achlorhydric conditions the fraction available for absorption from the AZD8055 200 mg free base suspension was substantially reduced (~40% cf ~80%) and variability increased. The effect of presenting AZD8055 as the fumarate salt is shown in Fig. 4. Presenting AZD8055 as the fumarate salt removed the decrease in bioaccessible fraction seen under achlorhydric conditions resulting in good and similar bioaccessibility regardless of gastric condition. The impact of dosing AZD8055 fumarate as a tablet on bioaccessible fraction is shown in Fig. 5, the tablet produced similar performance to the solution regardless of gastric pH.

Clinical Study

The data showed that exposure (area under the plasma concentration-time curve) increased broadly proportionally with increasing dose, although inter-patient variability was high (Figs. 6 and 7).

Fig. 5. Graph of cumulative appearance of AZD8055 in jejunal and ileal filtrates *versus* time after dosing of 200 and 500 mg AZD8055 free base as a solution to pH 2 stomach and AZD8055 fumarate tablets 120 and 240 mg free base equivalents to pH 5 and 2 stomach, respectively. $N=3$, mean $\pm$ SD except 500 mg solution and 240 mg tablet

DISCUSSION

Evaluation of AZD8055 and formulations in the TNO TIM-1 multi-compartmental, dynamic, system of the upper gastrointestinal tract allowed the project to move forward with confidence that exposures in the early clinical evaluation stage, when there was a desire to investigate safety and tolerability across a reasonable exposure range, would not be limited by formulation performance. This was shown by the proportional performance of both solution and suspension in TIM-1 system at doses of 200 and 500 mg. These data showed that neither dissolution rate nor solubility are limiting factors to the absorption of AZD8055 under standard conditions. An apparent paradox exists as the maximum bioaccessible fraction is about 80%, indicating some limitation to absorption. This can be explained by the fact that the bioaccessible fraction is a composite value made up of dissolution rate/solubility and filtration rate (permeation). A filtration rate of 3.9 mL/min was chosen which results in an 'effective permeability' that is at the upper end of BCS low permeability categorisation and commonly used as the low/high permeability cut off used in a discovery setting, that is a permeability that results in 80% or greater of the dose being absorbed. This is slightly lower than the BCS categorization of high permeability which requires a fraction absorbed of 90% or greater (19). The filtration rate of 3.9 mL/min results in a bioaccessible fraction for a highly soluble drug dosed as a solution (when dissolution/solubility is not limiting) of 80%. Therefore solution and suspension 200 and 500 mg curves

(Fig. 2) are actually 'permeability' limited. An example of a dissolution rate-limited profile was observed when the free base suspension is dosed under high gastric pH (Fig. 3).

Also noteworthy in this experiment is the low variability observed for the 200-mg solution data when a larger number of replicates has been performed. McAllister (21) has previously noted that TIM-1 publications tend to have a low replicate number due to the time taken to run each replicate and this is a perceived weakness of the system. Although low replicates are a reality it is not really a weakness, as, through the fine control of the system resulting from dynamic feedback to the computer produces a low inherent variability (all sources of variability including analytical=3%) (AstraZeneca, data on file). This means that low or even a single replicate is considered sufficient to base early development decisions on. For the important bridging decision (200 mg solution/120 mg tablet) three replicates were performed confirming the low variability. For sub-optimal formulations or compounds with poor biopharmaceutic properties, the variability can be higher (for instance, see Fig. 3 and the performance of the free base suspension in achlorhydric conditions). However, under these circumstances the bioaccessible fraction is also low so there is limited risk of concluding that a formulation is good even if there is some variability about the mean performance.

The performance of the suspension in TIM-1 system suggested that an immediate release tablet would provide acceptable AZD8055 exposure for long term development in standard subjects. However when tested in the TIM-1 system

Fig. 6. Plot of individual and geometric mean AZD8055 area under the plasma concentration time curve *versus* dose and formulation

under the condition of achlorhydria suggested there was likely to be halving of exposure. This was particularly relevant for AZD8055 as it was being developed in the Oncology arena and there was thus an increased probability that a reasonable proportion of the patient population would be achlorhydric (age related) and/or taking medication that increases gastric pH such as proton pump inhibitors (for symptomatic relief). These data strongly indicated that the formulation development should aim to reduce the variability in exposure due to variability in patient stomach pH, which led to the engagement of product development to develop a mitigation strategy (e.g. salt selection or formulation approaches). Several enhanced formulation approaches could be considered to achieve this aim, but would substantially add to development time, cost and risk. The TNO TIM-1 experiments were able to show that fumarate salt of AZD8055 provided good exposure regardless of stomach pH and should be developed (Fig. 4). Thus immediate release tablets of AZD8055 fumarate were developed and manufactured and these were shown to produce greater than 79% bioaccessible fraction, similar to the oral solution already being dosed in the clinic and stomach pH-independent performance (Fig. 5).

The similar performance of the tablets to the solution in TIM-1 system gave the project team confidence that the tablet could be switched into the clinical programme without dose adjustment. That is, for the same dose, the tablet would not produce an increase in exposure so that the safety data already generated at the dose for the solution was relevant. Secondly, the exposure would not decrease so that the patient

had the same probability of receiving benefit from treatment with the tablet as from the solution. This was important for AZD8055 project, because AZD8055 was aimed at an Oncology target and the risk benefit analysis precluded dosing AZD8055 to healthy volunteers. Initially a solution was chosen to allow dose flexibility but as the clinical study and dose exposure knowledge increased it was desirable to introduce a tablet which offered patient convenience/compliance benefits. As exposure could not be confirmed in healthy volunteers the tablet was introduced into the clinical study at the highest dose of the solution, based on the equivalent performance in the TIM-1 system. Subsequent analysis of the AZD8055 plasma concentrations after tablet administration support the conclusions drawn from the TIM-1 experiments:

- That exposure would increase in an approximately dose proportional manner and not be limited by the solubility or dissolution of AZD8055.
- That exposure produced by the solution and tablet would be comparable.

However, it should be noted that the doses explored in the clinic are somewhat lower than anticipated initially.

This is the first time that a multi-compartmental, dynamic, system of the upper gastrointestinal tract (in particular the TNO TIM-1 system) has been shown to be useful in supporting the formulation development of a poorly soluble compound. At AstraZeneca the TNO TIM-1 system has been used to support successfully the formulation development of numerous projects of poorly soluble com-

Fig. 7. Plot of individual and geometric mean dose normalised AZD8055 area under the plasma concentration time curve versus dose and formulation

pounds. These projects have spanned several therapy areas and formulation types. Formulation performance in TIM-1 system was matched to either performance in dog or the clinic when that data existed. The success of the TNO TIM-1 system has meant that it has almost entirely replaced the *in vivo* dog model in the formulation development cascade.

However, there are some issues:

- For a small number of compounds, extensive plastic binding has been observed so that drug recovery is low and data interpretation is difficult or impossible
- In two projects, no difference in product performance was observed when tested in TIM-1 system, but there were differences observed in product performance when tested in the clinic

In both these cases, the differential clinical performance was probably a consequence of differential stomach dissolution or gastric emptying. Developments are ongoing at TNO to deliver a stomach with improved hydrodynamics and particularly the ability to mimic antral grinding (TIM_{psf}). Consequently in our opinion the TIM-1 system is best suited for rapid disintegrating products which have less of a dependence on stomach hydrodynamics.

CONCLUSIONS

The TNO TIM-1 system has demonstrated an ability to support the development of poorly soluble drug products using the 'lipid membrane' set up and captures dissolution under dynamic physiologically relevant conditions. Uniquely it also mimics the concurrent 'absorption' step thus providing greater insight into formulation performance than static systems, in which dissolved drug is not removed thus curtailing further dissolution that would occur *in vivo*. The design of the system means that it can quickly provide insight into the impact of GI physiological variability such as variable gastric pH. Although there are few publications describing the use of dynamic complex dissolution apparatus to characterise the clinical performance of formulations of poorly soluble drugs consideration should be given to the use of such apparatus to replace clinical bridging and/or bioequivalence studies. Areas where initially it may be most appropriate to consider dynamic complex dissolution apparatus as a surrogate for a clinical bioequivalence studies are situations when:

- Access to patient populations is difficult or healthy volunteer studies are not practical and/or
- A correlation exists between product performance in the complex apparatus and clinical exposure.

However, the TNO TIM-1 system in its current form should be used with caution in products that show variable/longer disintegration and/or which may be highly dependent on gastric emptying/dispersion.

REFERENCES

1. Minekus M, Marteau P, Havenaar R, Huis in't Veld HJ. A multicompartamental dynamic computer-controlled model simulating the stomach and small intestine. *Atla*. 1995;23:197-209.
2. Havenaar R, Minekus M. *In vitro* model of an *in vivo* digestive tract. JP, US, European Patent 1994: PCT/NL93/00225
3. Blanquet S, Zeijdner E, Beyssac E, Meunier JP, Denis S, Havenaar R, Alric M. A dynamic artificial gastrointestinal system for studying the behavior of orally administered drug dosage forms under various physiological conditions. *Pharm Res*. 2004;21:585-91.
4. Larsson M, Minekus M, Havenaar R. Estimation of the bioavailability of iron and phosphorus in cereals using a dynamic gastrointestinal model. *J Sci Food Agric*. 1997;74:99-106.
5. Verwei M, Arkbage K, Havenaar R, Van den Berg H, Witthöft C, Schaafsma G. Folic acid and 5-methyl-tetrahydrofolate in fortified milk are bioaccessible as determined in a dynamic *in vitro* gastrointestinal model. *J Nutr*. 2003;133:2377-83.
6. Krul CAM, Luiten-Schuite Baan AR, Verhagen H, Mohn G, Feron V, Havenaar R. Application of a dynamic *in vitro* gastrointestinal tract model to study the availability of food mutagens, using heterocyclic aromatic amines as model compounds. *Food Chem Toxicol*. 2000;38:783-92.
7. Marteau P, Minekus M, Havenaar R, Huis in't Veld HJ. Survival of lactic acid bacteria in a dynamic model of the stomach and small intestine: validation and the effects of the bile. *J Dairy Sci*. 1997;80:1031-7.
8. Blanquet S, Meunier JP, Minekus M, Marol-Bonnin S, Alric M. Recombinant *Saccharomyces cerevisiae* expressing a P450 in artificial digestive systems: a model for biotransformation in the human digestive environment. *Appl Env Microbiol*. 2003;69:2884-92.
9. Oomen A, Hack A, Minekus M, Zeijdner E, Cornelius C, Schoeters G, Verstraete W, Van de Wiele T, Wragg J, Rompelberg CJM, Sips AJAM, Van Wijnen JH. Comparison of five *in vitro* digestion models to study the bioaccessibility of soil contaminants. *Environ Sci Tech*. 2002;36:3326-34.
10. Cardot J-M, Beyssac E and Alric M. *In vitro-in vivo* correlation: importance of dissolution in IVIVC. *Dissolution Tech*. 2007; pp 15-19.
11. Polli JE. *In vitro* studies are sometimes better than conventional human pharmacokinetic *in vivo* studies in assessing bioequivalence of immediate-release solid oral dosage forms. *AAPS J*. 2008;10:289-99.
12. Naylor TA, Connolly PG, Martini LG, Elder DP, Minekus M, Havenaar R and Zeijdner E. Use of a gastro-intestinal model and Gastroplus™ for the prediction of *in vivo* performance. *Industrial Pharm*. 2006; 9-12.
13. Souliman S, Blanquet S, Beyssac E, Cardot J-M. A level A *in vitro/in vivo* correlation in fasted and fed states using different methods: applied to solid release oral dosage form. *Eur J Pharm Sci*. 2006;27:72-9.
14. Tenjatla S, Romasanat V, Zeijdner E, Villa R, Moro L. Release of 5-aminosalicylate from MMX Mesalamine tablet during transit through a simulated gastrointestinal tract system. *Adv Ther*. 2007;24:826-39.
15. Fadda HM, Merchant HA, Arafat BT, Basit AW. Physiological bicarbonate buffers: stabilisation and use as dissolution media for modified release systems. *Int J Pharm*. 2009;382:56-60.
16. Selen A, Cruaños MT, Müllertz A, Dickinson PA, Cook JA, Polli JE, Kesisoglou F, Crison J, Johnson KC, Muirhead GT, Schofield T, Tsong Y. Conference report: applied biopharmaceutics and quality by design for dissolution/release specification setting: product quality for patient benefit. *AAPS J*. 2010;12:465-72.
17. Reis PM, Raab TW, Chuat JY, Leser ME, Miller R, Watzke HJ, Homberg K. Influence of surfactants on lipase fat digestion in a model gastro-intestinal system. *Food Biophys*. 2008;3:370-81.
18. Morihara M, Aoyagi N, Kaniwa N, Kojima S, Ogata H. Assessment of gastric acidity of Japanese subjects over the last 15 years. *Biol Pharm Bull*. 2001;24:313-5.
19. Waiver of *in vivo* bioavailability and bioequivalence studies for immediate-release solid oral dosage forms based on a Biopharmaceutics Classification System. U.S. Department of Health and Human Services, Food and Drug Administration, Center for Drug Evaluation and Research (CDER) <http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/ucm070246.pdf>. Accessed 18 Jan 2012.
20. Naing A, Aghajanian C, Raymond E, Kurzrock R, Blanco M, Oelmann E, Grinstead L, Burke W, Kaye S, Banerji U. First results from a phase I trial of AZD8055, a dual mTORC1 and mTORC2 inhibitor. *Mol Cancer Ther*. 2011;10 (11 Suppl): Abstract nr A168.
21. McAllister M. Dynamic dissolution: a step closer to predictive dissolution testing? *Mol Pharm*. 2010;7:1374-87.