The utility of microdosing over the past 5 years

Graham Lappin† & R Colin Garner
†Xceleron Ltd, The Biocentre, Innovation Way, York, YO10 5NY, UK

Background: Microdosing studies (human Phase 0) are used to select drug candidates for Phase I clinical trials on the basis of their pharmacokinetic properties, using subpharmacologic doses (maximum 100 µg). There are questions as to whether pharmacokinetic data obtained at these low doses will predict those at the clinically relevant dose. Objective: To review the current literature on microdosing and assess how well microdose data have predicted the pharmacokinetics obtained at a therapeutic dose. Methods: All data published in the peer reviewed literature comparing pharmacokinetics at a microdose with a therapeutic dose were reviewed, excluding those studies aimed at imaging. Conclusions: Of the 18 drugs reported, 15 demonstrated linear pharmacokinetics within a factor of 2 between a microdose and a therapeutic dose. Therefore, data that support the utility of microdosing are beginning to emerge.

Keywords: accelerator mass spectrometry, drug development, microdosing, pharmacokinetic linearity

Expert Opin. Drug Metab. Toxicol. (2008) 4(12):1499-1506

1. Introduction

A microdose (or human Phase 0) study is performed at a very early stage of drug development to obtain preliminary pharmacokinetic data on a drug candidate before the start of the Phase I clinical trials. As its name implies, the dose administered during a human Phase 0 study is very small, the amount being defined by both the European Medicines Agency and the FDA as one-hundredth of the predicted pharmacologic dose [1,2]. These small doses are considered inherently safer than pharmacologically active doses and, therefore, the regulatory authorities accept a much reduced safety toxicology package to allow a human Phase 0 study to proceed (although these safety packages are now under review [3]). This allows the drug candidate to be administered to human volunteers earlier and with less expenditure compared to a Phase I clinical study [4,5]. Some commentators have also considered that microdosing studies may strengthen the ethical basis for Phase I clinical trials [6].

The obvious question that arises, however, is how reliable the pharmacokinetic data obtained from a microdose are when compared to those obtained at higher clinically relevant doses; or in other words, how well the pharmacokinetics obtained at Phase 0 (using a maximum dose of 100 µg) predict those at Phase I (using a clinically relevant dose).

There was some initial speculative debate about the utility of microdosing [7,8] but more recently a body of data has been acquired, albeit limited at present, from which a better assessment can be made. This review examines the current status of microdosing (excluding studies relevant to imaging) and surveys the data published so far to see what lessons can be learned.
2. The purpose of microdosing

The initial concept of microdosing was proposed as a method of obtaining pre-Phase I pharmacokinetic data that were more reliable than some of the existing methods such as in vitro and in silico modelling and allometric scaling from animal studies [7]. Allometric scaling, although commonly used in an attempt to predict the pharmacokinetics in man from animal data, does not have a particularly good track record. In a review by Grass and Sinko [9] it was noted that there was no correlation between human and animal bioavailability for any of the species that were examined (rodents, dogs and primates). For some compounds pharmacokinetic parameters might correlate but for others they may not. Physiologically based pharmacokinetic models are probably more reliable than allometric scaling but even so, there are notable limitations, for example, in cases where renal or biliary clearance dominates [10]. In the context of microdosing, the hypothesis was that data obtained in vivo in humans at low doses would be a better predictor of pharmacokinetics at therapeutic doses than allometric scaling and probably physiologically based pharmacokinetic models; in other words a dose extrapolation would be more appropriate than a species extrapolation. In 2003 there were only summary data available for one drug and so the hypothesis was untested. Five years later there are now 18 drugs in the peer reviewed literature that compare the pharmacokinetics of a microdose with a therapeutic dose and so, to a limited extent, it is possible to make some estimation as to whether the original hypothesis had any validity. To make the comparison, however, it is important to define the boundaries for which the comparison is being made. There has been some debate about how close the pharmacokinetics obtained from a microdose has to be to the therapeutic dose to be a useful predictor. It has been suggested that differences of 25 or 30% between pharmacokinetic parameters measured for the two-dose levels would be inadequate [11] but others have suggested that a prediction within a twofold of the mean observed in a Phase I study is acceptable [12]. The latter is a commonly held view, widely adopted when using allometric scaling and, therefore, this is the criterion adopted in this review as well [12].

It should be borne in mind that human Phase 0 studies are likely to be performed in situations where in vitro or in silico modelling and animal studies are thought to be unreliable. On the other hand, in cases where these traditional approaches are believed to be predictive, there is probably little to be gained from a microdose study. It is often the case that several drug candidates are compared in a microdose study to select the lead and back-up candidate based on optimal pharmacokinetics. Pharmacokinetics can be of pivotal importance in the selection of the most appropriate candidate drug. For example, a given dose of drug will achieve a certain plasma concentration depending on its volume of distribution. Lower than expected plasma concentrations following oral dosing, for example, could be owing to poor performance of the formulation, poor absorption or permeability (or first-pass metabolism) or, equally, to a higher than anticipated volume of distribution [13]. Microdosing does not, however, provide any information on toxicology or efficacy per se and, therefore, not performed routinely rather than with a specific purpose and under conditions where predefined questions are being asked. For example, the question may be one of half-life or clearance to meet the requirements for a specified dosing regimen (e.g., once a day). Alternatively, the question may be one of bioavailability, particularly in situ ations where, for example, first-pass metabolism is difficult to predict in humans. On the other hand, microdosing is unlikely to be useful in predicting, for example, population pharmacokinetics as it has been pointed out that these early human Phase 0 studies are performed with small numbers of volunteers [11,14]. The potential benefits and limitations of human Phase 0 studies have been explored previously [15].

There has also been some debate about whether the prediction of a drug’s pharmacokinetics remains problematic in terms of drug selection as it has been pointed out that poor pharmacokinetics was attributed to 40% of drug attrition in 1991 but only to 10% by 2000 [14]. It has been argued, however, that drug attrition can rarely be attributed to a single factor and that there are dangers of placing reasons for failure into one particular ‘silo’ [16]. The 40% attrition rate for pharmacokinetics quoted for 1991 was heavily skewed by the contribution of several anti-infective drugs. With anti-infectives, there is a well-established relationship between pharmacokinetics and pharmacodynamics [17] and so the reason for attrition could have equally been efficacy instead of pharmacokinetics, which would have altered the figure significantly. This example, therefore, illustrates how this type of simplistic classification has to be treated with some caution.

3. Analytical modalities used in microdosing

There are essentially three analytical technologies that can be used to acquire data from a microdose study: positron emission tomography (PET), liquid chromatography mass-spectrometry/mass spectrometry (LC-MS/MS) and accelerator mass spectrometry (AMS). Very recently, a new technique called optogalvanic spectroscopy, which rivals AMS in its sensitivity, was reported [18]. Although this technique may have utility for microdosing in the future, now it is a very new technology and, therefore, is not considered further in this review.

Positron emission tomography is an imaging technique where either the drug or a specific receptor ligand is labelled with a short-lived positron emitting isotope [19]. One of the earliest receptor ligands was 11C-raclopride to measure cerebral D2-receptor occupancy [20]. Receptor ligand binding studies require drug concentrations sufficiently high to displace the ligand from its site of occupancy and, therefore, these types of PET studies are not common to microdosing. On the
other hand, using a drug substance labelled with a position emitting isotope and imaging the drug’s disposition, for example, in the brain, has been applied many times [21]. Positrons emitted from $^{11}$C (or another isotopic label), annihilate on collision with their antimatter equivalent electrons, forming $\gamma$-radiation perpendicular to the point of collision at a characteristic energy of 511 keV, which is then imaged by $\gamma$-cameras. Position emitting isotopes have short half-lifes, for example, the half-life of $^{11}$C is just 20.4 min, which precludes the acquisition of any pharmacokinetic data after ~ 1 h from dose administration. Although PET is a very valuable imaging technique, the data it acquires during a microdosing study is, nevertheless, limited in terms of determining pharmacokinetic parameters such as clearance, volume of distribution and half-life and, therefore, it is not considered in this review any further (although the reader is referred to references [21,22] for further information).

Both LC-MS/MS and AMS will generate drug concentration data from samples taken from human volunteers over time (the samples being typically plasma and urine), which are used to calculate pharmacokinetic parameters. An advantage of microdosing is that it is relatively easy to obtain fundamental pharmacokinetics (e.g., clearance and volume of distribution) from i.v. administrations, albeit at low doses. Accelerator mass spectrometry is an isotope ratio method and, therefore, requires that the drug contains a rare isotope, typically $^{14}$C. Because of the sensitivity of AMS (see later) it is necessary to administer only very low levels of $^{14}$C to human volunteers, typically below those where regulatory approval, for the administration of radioactivity, is required. Liquid chromatography MS/MS does not require an isotopically labelled drug, which makes the study design somewhat more straightforward. Given that the dose administered in a microdose study is a maximum of 100 µg, very sensitive analytical techniques need to be applied to the determination of the drug’s concentration. The sensitivity of LC-MS/MS is now typically in the region of 10 ~ 100 pg/ml, with some methods achieving 1 pg/ml, depending on the properties of the particular drug under analysis. Depending on the specific activity of the drug administered, AMS can measure drug concentrations in the zeptomole region and, therefore, offers potentially greater sensitivity of 100,000 times or more [23]. To illustrate the analytical sensitivity required for a microdose study, take as an example a drug with a moderate volume of distribution of 500 l. An intravenous dose of 100 µg will, therefore, attain a blood concentration of 200 pg/ml at time 0. Assuming it is necessary to follow the concentration of the drug over five half-lifes to obtain a good measure of the AUC then the assay would have to have a limit of quantification of ~ 5 pg/ml. If the same drug was dosed orally without a reasonably good previous knowledge of its bioavailability, then a limit of quantification of even 5 pg/ml would probably be insufficient.

Accelerator mass spectrometry can be used to measure the total $^{14}$C content (i.e., the sum of parent drug and $^{14}$C-labelled metabolites) as well as that of just the parent drug (or selected metabolites) by means of previous separation with HPLC [24]. Although the synthesis of a $^{14}$C-labelled drug is an added complication and expense, it should be borne in mind that comparison of systemic total $^{14}$C concentration versus parent drug concentration does provide information on the extent of metabolism and a measure of first-pass effects, which LC-MS/MS alone cannot attain [25]. It should be noted, however, that at present there are very little, if any, data in the literature that examine the ability of microdosing to predict the metabolism of a drug; data are more focused on attaining pharmacokinetics of the parent drug.

4. Current microdosing literature

Not including imaging studies using PET as the analytical modality, there are 18 drugs reported in the peer reviewed literature at present [7,26-32] where the pharmacokinetics from a microdose have been compared to that of a therapeutic dose. There are almost certainly a great deal more data but unfortunately it remains unpublished owing to the commercial sensitivity of such information at an early stage of drug development. However, some microdosing data alone, without the corresponding data for the therapeutic dose, is now appearing in the literature [33].

The first microdose data to be published was in 2003 [7]; it presented summary results for an α1A-adrenoceptor antagonist administered at doses of 5, 50 or 500 µg given to six healthy male volunteers (500 µg was the proposed therapeutic dose). Plasma was collected over 7 days from each dosing and was analysed by AMS. The drug was $^{14}$C-labelled, administering ~ 1.85 kBq (50 nCi) to each volunteer. Pharmacokinetic parameters were not reported but graphical presentation of the data showed that the pharmacokinetics were linear between doses, with an estimated half-life of ~ 40 h.

The following year (2004) saw the publication of pharmacokinetic data obtained in the dog for 7-deaza-2′-C-methyl-adenosine administered at 0.02 mg/kg p.o. and i.v. and 1 mg/kg p.o. [29]. Blood samples were taken over 80 h. The drug was $^{14}$C-labelled and samples were analysed for total $^{14}$C concentration with AMS. In addition, dogs were administered 7-deaza-2′-C-methyl-adenosine 0.4 mg/kg i.v., followed by 1 mg/kg p.o., 2 weeks later. In this group the drug was not labelled and blood samples, taken over 72 h, were analysed for parent drug with LC-MS/MS. For pharmacokinetic assessments, it is not usually considered good practice to measure total $^{14}$C in blood as the measurements represent potential mixtures of parent and metabolites. In this study with 7-deaza-2′-C-methyl-adenosine, although total $^{14}$C measurements were made, comparisons between AMS and LC-MS/MS data showed that the total $^{14}$C determinations were representative of parent drug concentrations and so
the data do appear valid. Pharmacokinetics over the 50-fold dose range were shown to be linear with the AUCs attained at the 1 mg/kg dose, being on average 75% of those attained from the 0.02 mg/kg dose (when dose normalised) for both p.o. and i.v. administration. Half-lifes were calculated at 3 points in the drug plasma concentration time curves, which, for the 0.4 mg/kg i.v. dose, were 0.1, 0.8 and 17.5 h for \( t_\alpha \), \( t_\beta \) and \( t_\gamma \), respectively (although the time intervals were not reported). The equivalent half-lifes for the 0.02 mg/kg i.v. dose were 0.13, 0.71 and 22.7 h for \( t_\alpha \), \( t_\beta \) and \( t_\gamma \), respectively. It is difficult to compare these half-lifes with the literature as it is usually only the terminal half-life that is quoted.

In 2006, Balani et al. published data comparing the pharmacokinetics of three drugs in the rat using up to five dose levels [26]. In this study, all measurements were done using LC-MS/MS and so the drugs were not \(^{14}C\)-labelled. The three drugs were fluconazole, tolbutamide and a drug identified as MLNX. Doses were given orally at between 0.001 and 10 mg/kg (although doses varied between compounds). Samples of plasma were collected over 24 h. Essentially, linear pharmacokinetics was demonstrated for fluconazole (5,000-fold dose range) and tolbutamide (1,000-fold dose range). For fluconazole dose normalised AUCs differed by ∼ 1.6-fold between the top (5 mg/kg) and bottom (0.001 mg/kg) doses. For tolbutamide dose normalised AUCs differed by ∼ 2-fold between the top (1 mg/kg) and bottom (0.001 mg/kg) doses. Half-lifes for fluconazole ranged from 4.6 to 7.3 h and for tolbutamide from 6.8 to 10.1 h. MLNX demonstrated essentially linear pharmacokinetics between 0.01 and 1 mg/kg, with the dose normalised AUCs being virtually the same. The AUC for the top dose (10 mg/kg) was, however, nearly 3.8-fold different to the lowest dose when dose normalised. MLNX, therefore, demonstrated pharmacokinetic nonlinearity at the top dose but how relevant this would be to therapeutic dose levels in humans is difficult to ascertain.

A consortium of pharmaceutical-based companies funded a five-compound human microdose study in 2004. The Consortium for Resourcing and Evaluating AMS Microdosing (CREAM Trial) published their findings in 2006 [27]. The five drugs were warfarin, ZK253, diazepam, midazolam and erythromycin. The drugs were \(^{14}C\)-labelled (7.4 KBq, 200 nCi administered per volunteer) and analysis was with AMS following HPLC separation of parent drug. With the exception of ZK253, male volunteers were used but ZK253 was administered to postmenopausal women.

Erythromycin was, in retrospect, a poor choice of drug as it is known to be very acid labile [34] and it was likely that the oral microdose degraded rapidly in the acidic environment of the stomach. The study was not designed to compare a microdose with a therapeutic dose using the intravenous route of administration and, therefore, no comparison of pharmacokinetics could be made. It is worth pausing here for a moment to explain a much misunderstood aspect of intravenous microdosing. A microdose (human Phase 0) is based on the administration of a maximum 100 µg of drug. There is another study design, explored in the CREAM trial, whereby a \(^{14}C\)-labelled tracer intravenous dose (maximum 100 µg) is administered along with a therapeutic nonlabelled dose given by an extravascular therapeutic route (typically orally). This latter study design is not a microdose study as a therapeutic dose has been administered to the volunteers. Intravenous tracer dose studies are performed in a Phase I setting, as the extravascular dose is administered at the therapeutic dose level and, therefore, the necessary toxicology studies are required to justify the safety of the dose for the extravascular route to be used in humans. In addition, in a microdose study the resultant drug plasma concentrations are only relevant to the microdose administered. In intravenous tracer dose studies, the plasma drug concentrations for the intravenous dose are overwhelmed by the systemic concentrations of the drug from the extravascular route (assuming it is bioavailable). The shape of the plasma drug concentration–time curve obtained for the intravenous dose (and thus the values for clearance and volume of distribution) are, therefore, governed by the total drug concentration in the plasma. As the study design demands that the extravascular administration is given at a clinically relevant therapeutic dose the drug plasma concentrations are therefore by definition, equivalent to those achieved by therapeutic dose administration [13,35]. It has been incorrectly claimed, for example, that the tracer dose of erythromycin given in the CREAM trial (\(^{14}C\)-labelled 100 µg i.v. dose given simultaneously with a 250 mg nonlabelled oral dose) showed linear pharmacokinetics with a therapeutic dose [11]. In fact, no comparison can be made as no intravenous microdose of erythromycin was administered in that study (only a \(^{14}C\) intravenous tracer dose along with a therapeutic oral nonlabelled dose). For these reasons, erythromycin is not included in the 18 drugs now reported in the literature that compares pharmacokinetics at a microdose and therapeutic dose.

Of the other four drugs compared in the CREAM trial, one (orally administered warfarin) showed nonlinear pharmacokinetics, with the plasma concentration arising from the microdose (100 µg) initially falling more rapidly and then more slowly than following a therapeutic dose (5 mg). This had also been previously observed in the rat and was attributed to the uptake of the drug onto a high-affinity, low-capacity binding site, coupled with a low volume of distribution [36]. Pharmacokinetics seen between microdoses (100 µg in all cases) and therapeutic doses of diazepam (10 mg), midazolam (7.5 mg) and ZK253 (50 mg) were essentially linear within a factor of twofold.

Fexofenadine was orally administered as a microdose (100 µg) and as a therapeutic dose (60 mg) to human volunteers by Yamane et al. (2007) [31]. Plasma samples were collected for 12 h following dosing and analysed by LC-MS/MS. This particular publication focused on the development and validation of suitable LC-MS/MS methods to measure the drug concentrations and very little was reported on the pharmacokinetics of the drug. Nevertheless, the plasma
concentration–time plots for both doses, when dose-normalised were very similar and the pharmacokinetics was reported as being linear between doses.

Vuong et al. compared the pharmacokinetics of an oral dose of zidovudine (520 ng) with previously published data for a therapeutic dose (between 134 and 600 mg) [30]. The study was performed in human volunteers using 14C-labelled drug and AMS analysis following UPLC separation of parent drug and the main metabolite. Comparison of the AUC for the microdose with those previously reported were within 1.6-fold and the half-life was marginally greater than 2-fold (literature 1.03 – 1.43 h, microdose 0.68 h). The pharmacokinetics of the main metabolite of zidovudine (GZDV) was also similar between the microdose and previously published data for a therapeutic dose, with the AUC being within 2.1-fold and the half-life 1.8-fold.

The pharmacokinetics of five drugs given orally to the rat at doses ranging from 1.67 to 1670 µg/kg were compared by Ni et al. (2008) [28]. The drugs were antipyrine, carbamazepine, atenolol, digoxin and metoprolol. Analysis of plasma samples taken up to 24 h from dosing was by means of LC-MS/MS. For antipyrine, carbamazepine, atenolol and digoxin, the pharmacokinetics were essentially linear between the 1.67 and 1670 µg/kg dose within a factor of 2, with the dose normalised AUCs for the microdose being within 70% of those for the highest dose, over a 10,000-fold dose range. It was reported that metoprolol showed a nonlinear relationship for the AUC but was linear for the Cmax. This latter compound, however, was difficult to measure owing to limitations of the LC-MS/MS analysis and data for doses lower than 16.7 µg/kg were not attainable.

Also in 2008, Madan et al. reported the pharmacokinetics of diphenhydramine and a structurally related drug NBI-1 in human volunteers administered via both the oral and intravenous dose routes (100 µg) [32]. The drugs were 14C-labelled and AMS following HPLC separation was used to measure the plasma concentration of parent drug. Pharmacokinetic data were compared to the literature and to a previous Phase I clinical trial for NBI-1. Pharmacokinetics obtained from microdose of diphenhydramine compared well to literature values for a dose 500-fold larger (clearance 24 l/h, volume of distribution 313 l, half-life 9.0 h and absolute bioavailability 35%). By comparing the AUC of the parent drug with the AUC for the total 14C, it was shown that diphenhydramine was extensively metabolised with ∼ 93% of the AUC for total 14C representing metabolites. Pharmacokinetics for NBI-1 administered as a microdose were also consistent with those obtained from a 50 mg p.o. dose (a Phase I clinical study not previously reported). The AUC for a 50 mg p.o. dose of NBI-1 was 422 ng.h/ml and the dose normalised AUC for the 100 µg microdose was 292 ng.h/ml (1.4-fold difference). The results of the microdose studies were compared to those obtained by allometry (mouse, rat, dog, monkey) and were found to generally overpredict within a factor of 3-fold.

5. Lessons on microdosing from the current literature

The 18 drugs where a microdose has been compared to a therapeutic dose have diverse pharmacokinetic properties (Table 1). Half-lifes range from 1 to >40 h, clearance from 0.2 to 100 l/h, volume of distribution from 8.4 to 1,300 l, bioavailability from virtually 0 to 100% and solubility from virtually insoluble to 170 g/l. There is, therefore, no obvious pharmacokinetic parameter that would cause concern in terms of the predictability of microdosing; for example, a drug with a short half-life would be equally as likely to be predictive in a microdosing study compared to one with a long half-life. Of the 18 drugs tested so far, 15 showed essentially linear pharmacokinetics, which means that within this small population, 83% were predictive. Of the 3 drugs that showed nonlinear kinetics, one (MLNX) was only nonlinear at the highest dose level, another (metoprolol) was suspect because of questions about the sensitivity of the LC-MS/MS assay, leaving one (warfarin) that was unambiguously nonlinear.

It must be recognised that 18 drugs is not a large number on which to completely evaluate the utility of microdosing and more compounds need to be added to the database over time before a reasonably firm conclusion can be drawn. Nevertheless, microdosing has moved from a mere working hypothesis to gaining some validity based on 18 drugs published in the peer reviewed literature over the past 5 years. Furthermore, microdosing does appear to be gaining industry acceptance (AAPS Magazine February 2007).

What are the prospects of improving the predictability of microdosing further and are there any methods by which confidence could be added to the pharmacokinetic linearity across doses? It is possible, for example, to examine the pharmacokinetics of a drug at a range of dose levels in animal models before a human microdose study. Although the pharmacokinetic parameters obtained from animals may not be predictive of those in the human, at least it would test the likelihood of pharmacokinetic linearity. The assumption is that any mechanism that would lead to a nonlinearity in humans would be detected in the animal model. This is perhaps a reasonable assumption and evidenced by this review, several animal microdose studies have been successfully performed. How useful this would be in practice is very difficult to judge in the absence of a more thorough investigation but the concept clearly has some merit.

In 2005, a research study that examined the pharmacokinetics obtained between a microdose and a therapeutic dose of seven drugs in human volunteers was initiated. The research was funded by the European Community under the Framework-6 programme and was known as the European Microdosing AMS Partnership Programme (EUMAPP). The seven drugs and the reason for their choice are given in Table 2 (also at [37]). Results of EUMAPP are under evaluation at the time of writing and published results can be expected in 2009. Part of the objective of this project was to investigate...
The utility of microdosing over the past 5 years

6. Expert opinion

There is a growing body of evidence to support the utility of microdosing although there is still some way to go to fully assess this technique. In particular a better understanding is needed for those properties of a drug that might lead to significant nonlinearities in the pharmacokinetics seen between a microdose and a therapeutic dose. Anecdotally, microdosing is being used within the pharmaceutical industry but very few studies are being published, probably because ways of integrating human microdose data into physiologically based pharmacokinetic models and from the preliminary results, it appears that this may indeed lead to better pharmacokinetic predictions, although the data have yet to be peer reviewed and published.

Table 1. Pharmacokinetic parameters in humans at therapeutic doses for various drugs where the pharmacokinetics have been compared between a microdose and a therapeutic dose.

<table>
<thead>
<tr>
<th>Drug</th>
<th>Water solubility (mg/l)</th>
<th>Mol. Wt</th>
<th>Pharmacokinetics</th>
<th>Biotransformation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>F (%)</td>
<td>CL (l/h)</td>
</tr>
<tr>
<td>A-1A adrenoceptor</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Fluconazole</td>
<td>1</td>
<td>302</td>
<td>1</td>
<td>1.13</td>
</tr>
<tr>
<td>Tolbutamide</td>
<td>109</td>
<td>270</td>
<td>80</td>
<td>1.1</td>
</tr>
<tr>
<td>Warfarin</td>
<td>17</td>
<td>308</td>
<td>100</td>
<td>0.2</td>
</tr>
<tr>
<td>Midazolam</td>
<td>24</td>
<td>325</td>
<td>45</td>
<td>0.2</td>
</tr>
<tr>
<td>Diazepam</td>
<td>50</td>
<td>284</td>
<td>100</td>
<td>1.6</td>
</tr>
<tr>
<td>ZK-253</td>
<td>ND</td>
<td>502</td>
<td>&lt; 1</td>
<td>30</td>
</tr>
<tr>
<td>Fexofenadine</td>
<td>Poor</td>
<td>501</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Zidovudine</td>
<td>30,000</td>
<td>267</td>
<td>60</td>
<td>100</td>
</tr>
<tr>
<td>Diphenhydramine</td>
<td>3,000</td>
<td>255</td>
<td>70</td>
<td>26</td>
</tr>
<tr>
<td>NBI-1</td>
<td>ND</td>
<td>267</td>
<td>40</td>
<td>22</td>
</tr>
<tr>
<td>Antipyrine</td>
<td>50,000</td>
<td>188</td>
<td>22</td>
<td>300</td>
</tr>
<tr>
<td>Carbamazapine</td>
<td>Very poor</td>
<td>252</td>
<td>70</td>
<td>50</td>
</tr>
<tr>
<td>Metoprolol</td>
<td>170,000</td>
<td>267</td>
<td>40</td>
<td>60</td>
</tr>
<tr>
<td>Atenolol</td>
<td>135,00</td>
<td>266</td>
<td>58</td>
<td>10</td>
</tr>
<tr>
<td>Digoxin</td>
<td>Insoluble</td>
<td>780</td>
<td>70</td>
<td>1.4</td>
</tr>
</tbody>
</table>

Source: Data are from various sources.
CL: Clearance; F: Bioavailability; ND: No data; \( t_{1/2} \): Half-life; V: Volume of distribution

Table 2. The drugs tested in the European Microdosing AMS Partnership Programme.

<table>
<thead>
<tr>
<th>Drug</th>
<th>Reason for choice</th>
</tr>
</thead>
<tbody>
<tr>
<td>Paracetamol (acetaminophen)</td>
<td>Extensive Phase II metabolism</td>
</tr>
<tr>
<td>Phenobarbital</td>
<td>Long plasma half-life</td>
</tr>
<tr>
<td>Fexofenadine</td>
<td>Pgp substrate</td>
</tr>
<tr>
<td>Sumatriptan</td>
<td>Cytosolic metabolism</td>
</tr>
<tr>
<td>Propafenone</td>
<td>Known dose-dependent kinetics</td>
</tr>
<tr>
<td>S-19812</td>
<td>Formation of an active metabolite</td>
</tr>
<tr>
<td>Clarithromycin</td>
<td>Pgp and CYP3A4 substrate</td>
</tr>
</tbody>
</table>
of commercial sensitivities at an early stage of drug development. Like any scientific method, microdosing has to be applied intelligently and studies should be carefully designed to address specific questions for development of a particular drug.

Bibliography

Papers of special note have been highlighted as either of interest (*) or of considerable interest (**) to readers.

1. EMEA, Position Paper on non-clinical safety studies to support clinical trials with a single microdose. Position paper CPMP/SWP/2599, 23 June 2004
3. ICH Topic M3 (R2): note for guidance on non-clinical safety pharmacology to address specific questions for development of a particular drug. Like any scientific method, microdosing has to be
   • The first paper where microdose data were presented.
   • A review of allometric scaling.
   • The first paper presenting full microdose data in humans.

Declaration of interest

G Lappin and R Colin Garner are employed by and own shares in Xceleron Ltd, a commercial company specialising in the use of AMS in drug development and microdosing.
The utility of microdosing over the past 5 years


37. The website of the European Microdosing AMS Partnership Programme, a research project funded by the European Commission under the Framework Programme 6. Available from: www.EUMAPP.com

Affiliation
Graham Lappin† PhD & R Colin Garner‡ DSc
†Author for correspondence
‡Head of Research and Development
Xceleron Ltd, The Biocentre, Innovation Way, York, YO10 5NY, UK
Tel: +44 01904 561567; Fax: +44 0 1904 561560;
E-mail: graham.lappin@xceleron.com
§Chief Executive Officer
Xceleron Ltd, The Biocentre, Innovation Way, York, YO10 5NY, UK