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Review Article

The use of biopharmaceutic classification of drugs in drug discovery and development: current status and future extension

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Abstract

Bioavailability (BA) and bioequivalence (BE) play a central role in pharmaceutical product development and BE studies are presently being conducted for New Drug Applications (NDAs) of new compounds, in supplementary NDAs for new medical indications and product line extensions, in Abbreviated New Drug Applications (ANDAs) of generic products and in applications for scale-up and post-approval changes. The Biopharmaceutics Classification System (BCS) has been developed to provide a scientific approach for classifying drug compounds based on solubility as related to dose and intestinal permeability in combination with the dissolution properties of the oral immediate-release (IR) dosage form. The aim of the BCS is to provide a regulatory tool for replacing certain BE studies by accurate in-vitro dissolution tests. The aim of this review is to present the status of the BCS and discuss its future application in pharmaceutical product development. The future application of the BCS is most likely increasingly important when the present framework gains increased recognition, which will probably be the case if the BCS borders for certain class II and III drugs are extended. The future revision of the BCS guidelines by the regulatory agencies in communication with academic and industrial scientists is exciting and will hopefully result in an increased applicability in drug development. Finally, we emphasize the great use of the BCS as a simple tool in early drug development to determine the rate-limiting step in the oral absorption process, which has facilitated the information between different experts involved in the overall drug development process. This increased awareness of a proper biopharmaceutical characterization of new drugs may in the future result in drug molecules with a sufficiently high permeability, solubility and dissolution rate, and that will automatically increase the importance of the BCS as a regulatory tool over time.

Introduction

Bioavailability (BA) and bioequivalence (BE) play a central role in pharmaceutical product development and BE studies are presently being conducted for New Drug Applications (NDAs) of new compounds, in supplementary NDAs for new medical indications and product line extensions, in Abbreviated New Drug Applications (ANDAs) of generic products and in applications for scale-up and post-approval changes. For example, NDA BE studies may be required when comparing different formulations in pivotal clinical trials and products intended for market. The complexity and number of studies required is often driven by the fact that several dose strengths may be included in the development process. In addition, BE documentation may also be needed when comparing blinded and original comparator products in clinical trials. Thus, an NDA typically contains a multitude of BE studies.

The Biopharmaceutics Classification System (BCS) has been developed to provide a scientific approach for classifying drug compounds based on solubility as related to dose and intestinal permeability in combination with the dissolution properties of the oral immediate-release dosage form (Amidon et al 1995; CDER/FDA 2000; Yu et al 2002). The aim of the BCS is to provide a regulatory tool for replacing certain BE studies by accurate in-vitro dissolution tests. This will certainly reduce costs and time in the drug development process, both directly and indirectly, and reduce unnecessary drug exposure in healthy subjects, which is normally the study population in BE studies. The BCS is today only intended for oral immediate-release products that are absorbed throughout the intestinal tract. This is a consequence of the fact that the oral route is the most

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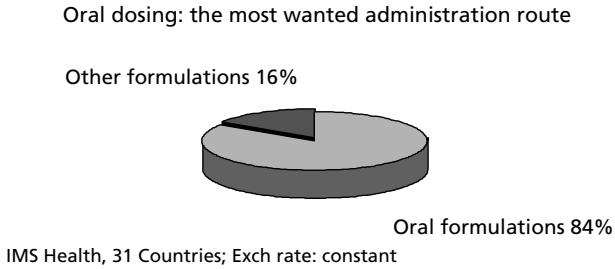

Figure 1 Percent sales of oral pharmaceutical formulations in comparison with other drug administration routes for the 50 most-sold pharmaceutical products in USA and Europe (from IMS Health 2001).

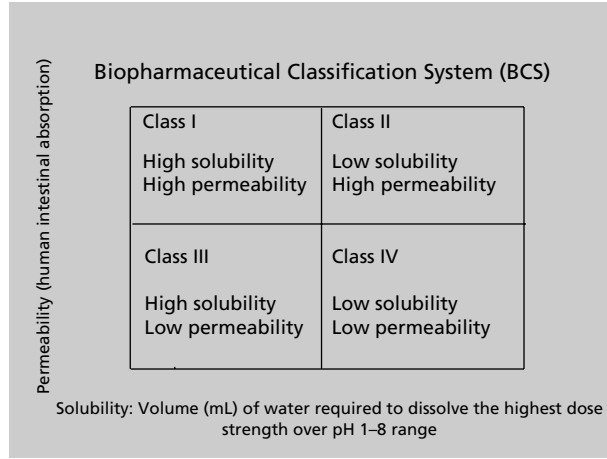

Figure 2 The Biopharmaceutics Classification System (BCS) provides a scientific basis for predicting intestinal drug absorption and for identifying the rate-limiting step based on primary biopharmaceutical properties, such as solubility and effective intestinal permeability (P_{eff}). The BCS divides drugs into four different classes based on their solubility and intestinal permeability (defined as more than 90% of the dose absorbed). Drug regulation aspects related to in-vivo performance of pharmaceutical dosage forms have been the driving force in the development of the BCS. Guidance for industry based on the BCS mainly clarifies when bioavailability/bioequivalence (BA/BE) studies can be replaced by in-vitro bioequivalence testing (www.fda.gov/cder/guidance/3618fnl.htm).

preferred drug administration route in the usage of drugs, with the majority (84%) of the 50 most-sold pharmaceutical products in the US and European markets being given orally (Figure 1). It has been reported that an application of a BCS strategy in drug development will lead to significant direct and indirect savings for pharmaceutical companies (Cook, J., oral drug delivery strategy; Veil 2004, personal communication). Furthermore, the BCS has been shown to be very useful for identifying the rate-limiting step and predicting intestinal drug absorption based on primary biopharmaceutical properties such as solubility and effective intestinal permeability (P_{eff}) (Amidon et al 1995; CDER/FDA 2000; Yu et al 2002). Accordingly, the current version of the BCS divides drug compounds into four classes based on their solubility and intestinal permeability (Figure 2). A drug is considered as highly permeable when the extent of fraction dose absorbed is complete in man, defined by the US Food and Drug Administration (FDA) as being more than 90%, whereas the European Medical Evaluation Office (EMEA) requires "complete" absorption (CDER/FDA 2000; CPMP 2001). A drug compound is currently classified as a high solubility drug when the highest clinical dose is dissolved in 250 mL buffer at all pH values within the range pH 1–7.5. This criterion is currently applied both by the European and US guidelines (CDER/FDA 2000; CPMP 2001).

The BCS has primarily been developed for regulatory applications. However, it has several other implications in both preclinical and clinical drug development processes and has gained wide recognition within the research-based industry (Yu et al 2002; Abrahamsson & Lennernäs 2003; Polli et al 2004). The importance of drug dissolution in the gastrointestinal tract and permeability across the gut wall barrier in the oral absorption process has been well known since the 1960s but the research carried out to constitute the BCS has provided new quantitative data of importance for modern drug development, so far especially within the area of drug permeability (Lennernäs 1998; Petri & Lennernäs 2003). Another merit of the BCS in a development context is that it provides very clear and easily applied rules for determining the rate-limiting factor in the gastrointestinal drug absorption process. Thereby, the BCS framework has implications in the selection of candidate drugs for full development, prediction and elucidation of food interactions, choice of formulation principle, including suitability for oral extended-release administration, and the possibility of in-vitro/in-vivo correlation in the dissolution testing of solid formulations (Fleisher et al 1999; Polli et al 2004). Most of these aspects will be discussed and exemplified in further detail below.

The aim of this review is to present the status of the BCS and discuss its future application in pharmaceutical product development. An extension of the BCS regulatory guideline is considered to be essential for a widespread use in all development phases as the current version limits the broad use of the BCS since class I drug substances today are quite rare in pharmaceutical pipelines. For instance, the proportion of class I compounds in the development phase for oral immediate-release formulations at AstraZeneca was less than 10% in 2001 (Abrahamsson & Lennernäs 2003). On the contrary, of the 123 WHO oral drugs available in immediate-release dosage forms, about 25–30% are class I drugs (Kasim et al 2004).

Pharmacokinetic definition of the intestinal absorption (fa), presystemic metabolism (E_G and E_H) and absolute bioavailability (F) of drugs administered orally to man

The most useful pharmacokinetic variable for describing the quantitative aspects of all processes influencing the absorption (fa) and first-pass metabolism and excretion (E_G and E_H) in the gut and liver is the absolute bioavailability (F) (Rowland & Tozer 1995). This pharmacokinetic parameter is used to illustrate the fraction of the dose that

reaches the systemic circulation, and relates it to pharmacology and safety of oral pharmaceutical products in various clinical situations. The bioavailability is dependent on three major factors: the fraction dose absorbed (fa); and the first-pass extraction of the drug in the gut wall (E_G) and by the liver (E_H) (Equation 1) (Amidon et al 1995; Wu et al 1995; Lennernäs 1998; von Richter et al 2001):

$$F = fa \cdot (1 - E_G) \cdot (1 - E_H) \quad (1)$$

There exist several factors that may affect the intestinal absorption (i.e., fa) and gut-wall metabolism (i.e., E_G) of drugs. These can be divided into three general categories: pharmaceutical factors; physiochemical factors of the drug molecule itself; and physiological, genetic, biochemical and pathophysiological factors in the intestine (Amidon et al 1995; Wu et al 1995; Lennernäs 1998; von Richter et al 2001). The fa is the fraction of the dose transported (absorbed) across the apical cell membrane into the cellular space of the enterocyte according to scientific and regulatory definitions (Amidon et al 1995; Wu et al 1995; Lennernäs 1998; CDER/FDA 2000; CPMP 2001; von Richter et al 2001). Once the drug has reached the intracellular site, it may be subjected to CYP P450 (predominantly CYP3A4) metabolism, as well as other enzymatic steps. The enzymatic capacity of the small intestine to metabolize drugs can, in pharmacokinetic terms, be expressed as the extraction ratio of the intestine (E_G) (Amidon et al 1995; Wu et al 1995; Lennernäs 1998; von Richter et al 2001). It is important to realize that CYP3A4 is not expressed in the colon (De Waziers et al 1990; Nakamura et al 2002; Berggren et al 2003). Instead, drug metabolism by colonic microflora may play a crucial role in colonic drug absorption, especially with regard to drugs given in extended-release dosage forms, which may be subjected to predominantly hydrolytic and other reductive reactions (Goldin 1990). The fraction that escapes metabolism in the small intestine ($1 - E_G$) may undergo additional metabolism and/or biliary secretion in the liver (E_H) before reaching the systemic circulation. The E_H is dependent on the blood flow (Qh), protein binding (fu) and intrinsic clearance of the enzymes or transporters (Clint) (Rowland & Tozer 1995). Recently, it has also been recognized that membrane transport into the hepatocyte has to be included in the models for predicting and explaining liver extraction (Tannergren et al 2003a; Chandra & Brouwer 2004).

Biopharmaceutical classification system (BCS)

Present biowaiver for class I drug

Presently, the BCS is primarily used as a regulatory tool for identifying which drug substances in oral immediate-release formulations are suitable for in-vitro BE testing. Additional criteria that must be verified for a biowaiver, besides solubility, dissolution and permeability, include drug stability in the gastrointestinal fluids, therapeutic index classification (narrow or non-narrow), no effect of pharmaceutical excipients on the rate and extent of absorption and that the formulation has no absorption from the oral cavity. If these criteria are met, test and reference products can be compared by in-vitro dissolution testing and deemed bioequivalent if sufficiently similar results are obtained. The in-vitro dissolution testing should be done at three different pH values within the physiological range – typically pH 1.2, 4.5 and 6.8. The product dissolution must be complete (>85%) within 30 min to utilize the in-vitro BE route. The underlying rationale for this demand on product performance is to ascertain that drug dissolution is fast enough so as not to become the rate-limiting step. It is assumed that gastric emptying will control the absorption rate for class I substances in products with such a rapid dissolution and no effect on BA will be obtained for different dissolution profiles within acceptance limits. This has also been verified in-vivo by studying metoprolol tablets (a class I drug) with different in-vitro release profiles (Rekhi et al 1997).

The difference in dissolution between a test (T) and a reference (R) product should be evaluated by use of the f_2-test (Equation 2), where $f_2 > 50$ is the required limit for equivalence. This limit corresponds to an average difference in amount dissolved at different times (t) of less than 10%. If the dissolution is very rapid (i.e. complete dissolution within 15 min), the f_2-testing is not necessary.

$$f_2 = 50\log\left[(1 + (1/n)\Sigma_{t=1}^{n}(R_t - T_t)^2)^{-0.5} \times 100\right] \quad (2)$$

In addition to the in-vitro testing, the test and reference products must not contain excipients that could modify drug absorption in any way, except for dissolution effects. For example, the potential for permeability-enhancing effects by surface-active agents, sometimes included in solid formulations, has been identified as one potential concern. However, the reported effects of formulation excipients on efflux inhibitors are not clearly defined, especially not in-vivo (Polli et al 2004). Furthermore, the effect on gastrointestinal transit by large amounts of sugars has been highlighted as another issue, especially for liquid formulations where large amounts of such excipients may be included (Adkin et al 1995; Yu et al 2002; Basit et al 2004).

BCS in the early drug development phase

Pharmacokinetic (ADME) parameters are today considered to have a crucial role in the selection process of oral candidate drugs for product development (Van de Waterbeemd et al 2001; Van de Waterbeemd & Gifford 2003; Lajiness et al 2004). Fundamental BCS parameters, such as permeability, solubility and fraction dose absorbed, are among those ADME parameters and therefore it has been suggested that these fundamental BCS parameters should be useful in both the discovery and early development process (Amidon et al 1995; Van de Waterbeemd 1998). For instance, it is clear that new compounds with a very low permeability and/or solubility/dissolution will certainly result in low and highly variable BA, which may limit the possibilities that a clinically useful product can be developed. It is obvious that a selection of

candidates that fulfil the BCS requirement of high permeability/high solubility (class I) almost guarantees the absence of failure due to incomplete and highly variable gastrointestinal absorption. However, these BCS limits are generally too conservative to use as acceptance criteria since many useful drugs can be found in class II–III and even class IV (Kasim et al 2004).

In the preclinical development phase, various animal models are used to evaluate the absorption potential of new drugs. Especially, the rat is considered as a validated and appropriate model that predicts human intestinal absorption well (Fagerholm et al 1996, 1997, 1999; Chiou & Barve 1998; Ungell et al 1998; Corrigan et al 2003; Berggren et al 2004). Experimental methods and relevant acceptance criteria regarding permeability and solubility are also needed in the early drug discovery process (Lipinski et al 2001; Egan & Lauri 2002; Sun 2004; Volpe 2004). Such procedures have also been introduced in the industry, including solubility screens using turbidimetric measurements and automatic permeability screens based on the Caco-2 cell model. There appears to be a good correlation between in-vitro and in-vivo permeability for drugs with passive diffusion as the main transport mechanism, but there is a significant deviation for drugs absorbed through transporters (Lennernäs et al 1996; Lennernäs 1997, 2003; Sun et al 2002). It is also considered that the interpretation of the importance of efflux carrier on intestinal absorption process is overrated based on results obtained in tissue cell cultures (e.g., Caco-2 cells) (Lennernas 1998, 2003; Chiou et al 2001; Yu et al 2002; Tannergren et al 2003a, b; Petri et al 2004). For instance, based on only cell culture data (Caco-2 cells), it was shown that the in-vitro permeability of fexofenadine in the absorptive direction increased by approximately 200–300% in the presence of various P-glycoprotein (Pgp) inhibitors, such as verapamil, ketoconazole and GF 120918, and that low passive diffusion was the main reason for the incomplete and variable intestinal absorption (Figure 3A) (Petri et al 2004). Interestingly, in-vivo perfusion of the proximal part of the jejunum with our Loc-I-Gut technique showed that the in-vivo permeability was affected by neither ketoconazole nor verapamil at clinical doses (Tannergren et al 2003a, b) (Figure 3B). This clearly shows that the cell monolayer is too poorly defined to quantitatively predict drug–drug interactions at the transporter level. These cell models can only provide quantitative in-vivo predictions of drug transport if an extensive mapping of the expression of functional activity of these membrane transporters is done. Therefore, there is a need to develop in-vitro techniques with functional expressed transporter activity that is better correlated to the various regions of the human intestinal tract (Sun et al 2002; Lennernäs 2003; Mizuno et al 2003; van Montfoort et al 2003; Petri et al 2004) (Figure 4).

It is well known that the intestinal absorption potential of drugs that are mainly transported by passive diffusion may be predicted from molecular properties (Lipinski et al 2001). Accordingly, it is suggested that the fundamental BCS parameters should be based on theoretical descriptors in the future and computational approaches have also

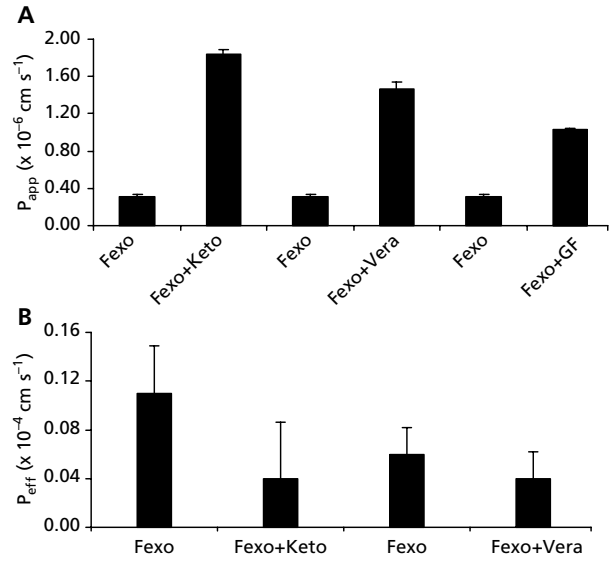

Figure 3 A. Effect of ketoconazole (Keto), verapamil (Vera) and GF 120918 (GF) on the in-vitro permeability (P_{app}) of fexofenadine (Fexo) in Caco-2 cells. B. Effect of ketoconazole and verapamil on the in-vivo permeability (P_{eff}) of fexofenadine in the single-passed perfused human jejunum. There is an important difference in the effect of the inhibitors on the fexofenadine absorptive permeability. It is obvious that the Caco-2 cells are more sensitive than the in-vivo situation. Future studies have to reveal if the cell model predicts the in-vivo situation better than the in-vivo perfusion model. Data are means ± s.e.m., from Tannergren et al (2003a, b) and Petri et al (2004).

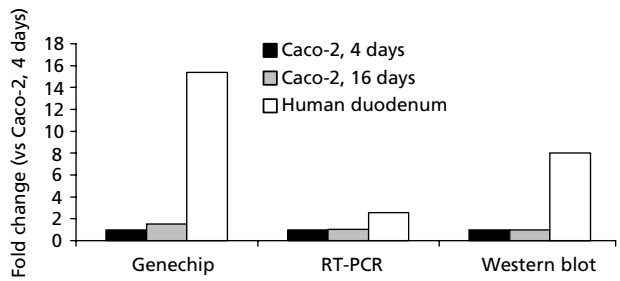

Figure 4 Comparison of Genechip, RT-PCR and Western blot for the human di-and tri-peptide transporter (hPepT1) in human duodenal tissue and Caco-2 cells cultured for 4 and 16 days. The peptide transporter expression was very low in Caco-2 cells, but its expression in human duodenum was 15-fold higher (Sun et al 2002).

been developed for permeability and solubility determinations (Amidon et al 1995; Van de Waterbeemd 1998; Winiwarter et al 1999, 2003; Bergström et al 2003; Fichert et al 2003; Kasim et al 2004). If further refinement can be achieved for these methods, such as quantitative structure activity relationship (QSAR) models to optimize ADME properties, it may be possible in the future to displace cell-based permeability screens and early solubility estimates (Lipinski et al 2001; Sun 2004). However, simple physicochemical descriptors of solubility and permeability must

probably be assembled into, and validated in, more physiological-based models to be able to accurately predict human intestinal absorption (Agoram et al 2001; Grass & Sinko 2002; Rowland et al 2004). Recently, Lindahl et al (2004) demonstrated that in-vitro permeability values could be used to simulate the absorption from various intestinal segments in man. An intermediate approach for permeability determination, between physicochemical- and physiological-based methods, is the use of artificial membranes (Bermejo et al 2004; Obata et al 2004). Such techniques provide a pure estimate of transcellular permeability without influence from transporters or the unstirred water layer present in cell-based models, which could be useful feedback to chemists in the screening phase. Finally, it is important to understand that even if absorption properties are acceptable the drug may have a limited clinical use due to an extensive first-pass extraction in the gut or liver.

The BCS in the pharmaceutical development phase

The BCS may be used to make the normally time-constrained pharmaceutical development process more efficient. If a drug is classified as having low solubility, it is obvious that the rate and extent of absorption may be improved by the use of formulation principles that increase the dissolution rate or drug solubility. There are several different formulation principles of varying complexity, ranging from selecting a suitable solid-state form or salt to the use of technologically more advanced formulation principles. Although their application could be limited by several practical factors, such as poor drug stability, excessive size due to the need for large amounts of excipients in relation to the dose, technical manufacturing problems and the high cost of goods, it is believed that many poorly soluble compounds with good pharmacological properties could be "saved" by such approaches. One of the most recent advances in this area is the utilization of nanoparticles, which can be obtained by milling or precipitation methods (Hu et al 2004; Patravale et al 2004). One favourable feature of this approach is the relatively small amount of additional excipients needed and very significant improvements in oral BA has been reported. Microemulsions and cyclodextrin complexes are example of other formulation approaches that have proven to increase BA several times for low solubility drugs.

Low permeability compounds are less suitable for formulation improvements of BA. Despite extensive research in the area of permeation enhancers, very few products using such principles have entered the market. Significant increases in drug permeability found in in-vitro models have often not provided corresponding responses in-vivo (Fagerholm et al 1998; Schipper et al 1999; Lennernäs et al 2002a; Kato et al 2003; Mahato et al 2003; Mrestani et al 2003). Another limitation of used enhancers has been their unspecific nature, leading to a risk of increased permeability to toxins and other undesirable molecules (Uchiyama et al 1999).

The use of pro-drugs has proven a more successful approach to increase the BA of low permeability compounds (Ettmayer et al 2004). Melagatran, a direct thrombin inhibitor, is a recent and illustrative example of how intestinal permeability is increased and the development potential of a drug is increased (Eriksson et al 2002, 2003). It is produced by adding protecting chemical groups to its prodrug ximelagatran (H376/95), which results in greater BA and less interindividual variability in the pharmacokinetics of melagatran. The two protecting groups change the pK_a values, producing a prodrug which is uncharged when its pH is above 6.2. Accordingly, the octanol–water partition coefficient and in-vitro permeability increased 170 and 80 times, respectively (Eriksson et al 2002, 2003). The BA of melagatran increased from 5% to 20% with significant less interindividual variability when ximelagatran, instead of melagatran itself, was given orally.

Targetting the carrier-mediated transport process is another successful prodrug strategy that has been shown to enhance intestinal absorption (Thomsen et al 2003; Ettmayer et al 2004; Steffansen et al 2004). For example, oligopeptide transporters are responsible for the active absorption of β-lactam antibiotics, the ACE inhibitor enalapril, valaciclovir and valganciclovir. The BA of valaciclovir was 5 times higher than that of aciclovir itself due to transport by the intestinal oligopepetide transporter PEPT1 (Beauchamp et al 1992; Swaan & Tukker 1997; Han et al 1998).

A successful development of oral extended-release formulations requires a strategy where the BCS today could provide guidance. It is well known that not all drugs would clinically benefit from being given in an extended-release formulation due to unfavourable human intestinal absorption properties along the intestine. If clinical advantage should be obtained, the extended-release formulation most often requires drug release between 12 and 24 h. Thus, since the small intestinal transit of formulations is only 3–4 h, a significant part of the dose in an extended-release product will be delivered to the colon, which means that the absorption has to be classified as high in the entire intestine. Class I drugs should therefore be the best candidates for extended-release product development, which is borne out by virtue of several extended-release formulations being based on class I compounds (Corrigan 1997). Metoprolol is such a drug; it has been shown to be a high permeability drug in the human jejunum, as determined with the Loc-I-Gut technique, and its BA did not differ after administration by intubation in different regions of the intestine (Figure 5) (Lindahl et al 1996). Consequently, metoprolol has a complete fraction dose absorbed and also high permeability in the colon. It is our experience that if passive diffusion is the dominating membrane transport mechanism, an intestinal permeability equal to, or higher than, that of metoprolol is an indication of a useful extended-release candidate. On the contrary, a low permeability drug will not even be completely absorbed in the small intestine given as a solution or an immediate-release tablet and, accordingly, the amount absorbed in the colon region will be even lower. This statement is based on experimental data demonstrating that the colonic membrane has an even lower permeability for drugs already classified as low permeability drugs in the small intestine. For instance, human and rat intestinal specimens mounted in the Ussing chamber model (in-vitro) have shown that the

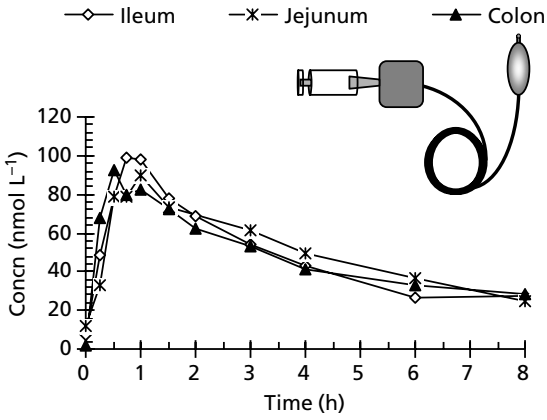

Figure 5 Mean plasma drug concentrations of metoprolol in healthy subjects (n = 8) after administration of metoprolol tartrate 25 mg, as a solution, to different parts of the gastrointestinal tract.

permeability of class III–IV drugs is even slower in the colon than in the small intestine, whereas class I–II drugs show a slightly higher permeability in the colon when passive diffusion is the dominating membrane transport mechanism (Corrigan 1997; Ungell et al 1998; Connor et al 2001; Berggren et al 2003). Consequently, it will not be possible to control the rate of absorption by an extended-release formulation for low permeability drugs. In addition, a large part of the dose will not be absorbed, leading to a low and uneven variability, and no increased duration of effect will be achieved. This is exemplified in extended-release tablets of amoxicillin. This high solubility drug (4.0 mg mL^{-1}) is classified as a low P_{eff}, even if it is transported across the intestinal barrier via the oligopeptide carrier (PepT1) (Oh et al 1993; Lennernäs et al 2002b). Essentially no absorption occurred for an extended-release tablet when it entered the colon, as determined by intubation technique and gamma scintigraphy, which is also in accordance with its slow passive permeability due to hydrophilic properties (PSA = 154.4, log $D_{6.5} = -1.7$ (Barr et al 1994; Gottfries et al 1996; Winiwarter et al 1999). However, recent advances in the area of gastric retention formulations hold some promise that extended-release formulations of low permeability compounds may be possible in the future, insofar as the need for colonic drug absorption is no longer critical since the product stays in the upper gastrointestinal tract during the entire period of drug release (Klausner et al 2003).

Class II drugs are also frequently used in extended-release formulations. Felodipine extended-release tablets are an example of a low solubility compound being included in a useful extended-release product. It has been possible to administer felodipine, which has a water solubility in the physiological pH range of about 1 μg mL^{-1}, as a once-daily product in doses of up to at least 20 mg without any reduced BA compared with an oral solution (Blychert et al 1990). This successful absorption is indeed dependent on the use of a dissolution-enhancing formulation principle where the drug is given in a solubilized form. Another class II compound, nifedipine (10 μg mL^{-1}; doses up to about 100 mg), is absorbed from the colon when released as a micronized drug (Grundy & Foster 1996).

A special consideration may be made regarding classification of low solubility compounds in extended-release forms. The standard classification is based on the idea that the drug should be completely dissolved in the gastric fluid, which has been estimated to be 250 mL (Amidon et al 1995). However, this way of classifying drugs may be less relevant for extended-release formulations since only a very small part of the dose is made available for dissolution in the stomach. The dose is generally spread over the entire gastrointestinal tract, making the effective water volume available as a dissolution medium for the drug probably larger than the 250 mL used in the original BCS. Furthermore, the drug permeability of these compounds is often much faster than the drug release, further preventing solubility limitations for class II drugs in extended-release formulations (Bonlokke et al 2001). Thus, increased understanding of total available intestinal fluid volumes and influence of factors other than pH on colonic drug solubility/dissolution are needed before scientifically based solubility criteria can be proposed in the future. The dynamic processing of lipids and bile acids along the small intestine is also a challenging area, which needs to be further considered for understanding and predicting drug solubility limitation of absorption along the intestine (Kossena et al 2004; Porter et al 2004).

In-vitro/in-vivo correlation (IVIVC)

In-vitro dissolution tests are important for the successful development of solid pharmaceutical products and in batch quality controls. The BCS is useful for predictions when IVIVC could be expected for solid immediate-release products. These in-vitro dissolution tests can only model the release and dissolution rates of the drug and it is only when these processes are rate limiting in the overall absorption that IVIVC can be established. For class I drugs, the complete dose will be dissolved already in the stomach and, provided that the absorption in the stomach is negligible, the gastric emptying will be rate limiting and therefore IVIVC is not expected. Thus, in-vitro dissolution testing can be expected to be "over-discriminating" for those drugs (i.e., tablets showing different in-vitro dissolution profiles will provide the same rate and extent of BA) (Rehki et al 1997).

Class II drugs are expected to have a dissolution-limited absorption and an IVIVC should be possible to establish using a well-designed in-vitro dissolution test. One way to investigate and establish such a correlation is to study formulations containing drug particles with different surface areas. For example, the different peak plasma levels in a human BA study obtained between candersartan cilexetil (water solubility < 1 μg mL^{-1}) tablets 8 mg containing drug particles with an average diameter of 4 μm and 9 μm, respectively, was predicted by a difference in in-vitro dissolution in the standard USP II paddle method containing a buffer medium with added surfactant (Figure 6). It has also been reported that, provided an appropriate composition is chosen for the dissolution test, the USP paddle apparatus can be used to reflect variations

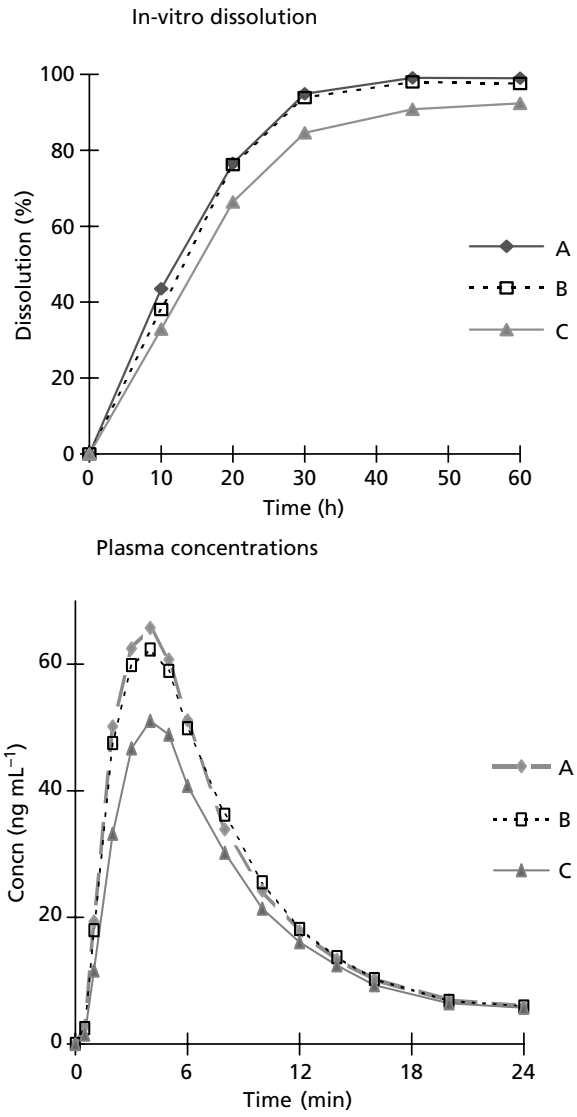

Figure 6 Mean in-vitro dissolution profiles and corresponding mean plasma concentration of three different tablet formulations of candesartan cilexetil containing different drug particle sizes: 4 μm (A), 6 μm (B) and 9 μm (C).

in hydrodynamic conditions in the upper gastrointestinal tract (Scholz et al 2003).

Two cases can be identified for class II drugs when the establishment of simple IVIVC is not feasible. Firstly, there are a number of formulation principles that could enhance the dissolution rate and solubility of low solubility compounds. It may be possible to achieve such a rapid and complete dissolution of a class II drug that the gastric emptying becomes rate limiting (i.e. the BA of the solid dosage forms equals that of an oral solution). In such a case, the prerequisites for IVIVC will be identical to the situation for class I drugs (i.e. no correlation will be obtained as long as the dissolution rate is significantly faster than the gastric emptying). It has also been discussed that these drugs and drug products should fall into a special intermediate class II, where a biowaiver should be applicable (Yu et al 2002; Polli et al 2004).

The second situation in which IVIVC is not likely for class II drugs is when absorption is limited by the saturation solubility in the gastrointestinal tract rather than by the dissolution rate. In this situation, the drug concentration in the gastrointestinal tract will be close to the saturation solubility and changes in the dissolution rate will not affect the plasma concentration profile or the in-vivo BA. Standard in-vitro dissolution tests are carried out under "sink conditions", at concentrations well below the saturation solubility. Thus, only effects related to the dissolution rate can be predicted in-vitro. If more physiologically relevant dissolution media are used, which do not necessarily provide "sink conditions", the possibility for IVIVC could be improved as indicated by recent work using simulated intestinal medium (Kostewicz et al 2002).

The absorption of class III drugs is limited by their intestinal permeability and no IVIVC should be expected. However, when the drug dissolution becomes slower than the gastric emptying, a reduction of the extent of BA will be found at slower dissolution rates, since the time during which the drug is available for transport across the small intestinal barrier will then be reduced.

Food–drug interactions

Food intake may affect the plasma exposure of a drug, which might influence the efficacy of a drug, and it is therefore crucial from a safety aspect to predict such effects at an early stage. However, an accurate prediction model is a challenge since several factors are involved in food–drug interactions, including physico-chemical effects such as increased solubility, binding to secretory or food components, physiological effects in the gastrointestinal tract such as altered flow rates and gastric emptying, mechanical effects on formulations due to different motility patterns, permeability effects due to interactions with active transporters or effects on the membrane and altered first-pass metabolism. An extensive review of different mechanisms for food–drug interactions can be found elsewhere (Fleisher et al 1999).

The BCS can be used to predict these mechanisms and the magnitude of food effects related to solubility, dissolution, gastric emptying and permeability. It is not possible to predict or describe the clinically important interaction with grapefruit juice and other food components that affects first-pass metabolism by using the BCS (Edgar et al 1992). The most severe cases of food–drug interactions due to factors considered in the BCS are generally found in the group of poorly soluble compounds (class II and IV drugs) given in high doses, as they approach the saturation solubility in the gastrointestinal lumen. The BA of griseofulvin (class II or class IV) has been reported to be increased up to five times by food, and dosing recommendations requiring concomitant intake of the drug with a meal are often used in these cases (Charman et al 1997). The saturation solubility will be significantly improved by food due to solubilization in mixed micelles, including bile acids, lecithin and monoglycerides obtained from the dietary fat intake, and dissolution into emulsified nutritional

lipids, and accordingly the amount of drug available for absorption will significantly increase (Porter et al 2004). An additional effect of the increased plasma exposure might be the relatively large fluid volume available in the stomach after a meal. The effect of just increasing the volume of water administered with a tablet from 200 mL to 800 mL increased the BA by 50% in a human study of danazol (water solubility 0.001 mg L^{-1}) tablets 250 mg (Sunesen 2003).

Proteolytic drugs with a pK_a within the physiological pH range will also be affected by the food-mediated pH changes in the stomach. A protein-rich meal could increase the pH from the fasting value of about 1.5–3 to a close to neutral pH (Dressman et al 1990; Lindahl et al 1997). Basic drugs, which are often freely dissolved in the acidic stomach, could thereby experience a reduced BA with food, whereas it may be the other way around for low solubility acids.

For class II drugs given in lower doses, the dissolution rate rather than the saturation solubility is the limiting factor. An increase in dissolution rate due to in-vivo solubilization mediated by food intake could theoretically be obtained but this is not always found in-vivo. For example, food does not affect the rate or extent of BA for candersartan cilexetil, a very poorly soluble compound given in a low dose (Gleiter & Morike 2002). An in-vitro dissolution and solubility study of this compound in simulated intestinal media provided a potential explanation: it was revealed that the solubility increased as a function of bile concentration as expected, whereas the dissolution rate was not increased by the higher bile concentrations being representative for the fed state. The slower gastric emptying of dissolved drug might also counterbalance increases in dissolution rate in the fed state. Thus, although intestinal solubility most often will be increased in the fed state for class II drugs, this will not always lead to a more rapid dissolution.

For class I drugs, a slower rate of absorption would be expected after concomitant food intake (reduced Cmax and prolonged tmax) due to the decreased gastric emptying rate induced by a meal. Gastric emptying is totally controlled by the two patterns of upper gastrointestinal motility – the inter-digestive and the digestive motility patterns. The inter-digestive pattern, termed the migrating motor complex (MMC), dominates in the fasted state and is organized into alternating phases of activity and quiescence (Sarna 1985). The gastric emptying in the fed state varies significantly depending on the motility pattern and meal composition, including factors such as energy content, osmolality and pH. A gastric emptying half-life of about 10 min is obtained in the fasting state, whereas a half-life of approximately 45 min has been reported for fluids when measured under non-fasting conditions (Oberle et al 1990; Ziessman et al 1992). It was also clearly shown that a smaller volume (50 mL) was more sensitive to the motility phase than large volumes (200 mL). Even during phase I of the MMC, the emptying rate was faster for the 200-mL volume (Oberle et al 1990). In addition, it is important to understand that drugs with a shorter half-life will be more sensitive to changes in gastric emptying. Recently, it was reported that in-vivo results appear to support the hypothesis that rapidly dissolving immediate-release solid oral products containing a BCS class I drug are likely to be bioequivalent under fed conditions (Yu et al 2004).

Class III drugs are perhaps those least sensitive to food intake. None of the effects induced by food, such as slower gastric emptying and increased solubilization capacity, will be of any relevance. It is also well-established that the transit from the main absorption region for a class III drug (i.e. small intestine) would not be affected by food and, generally, no effect on drug permeability is expected with food (Davis et al 1986; Ungell et al 1998; Berggren et al 2003). For example, no interaction with food was obtained for the low permeability compound ximelagatran (Eriksson et al 2002). In addition, Yu et al (2004) supported this by suggesting that BCS class III drugs may have the potential to be bioequivalent under fed conditions.

A plausible mechanism for interactions between food and drugs could occur with drugs that have carrier-mediated transport in the intestine, especially if nutritional carriers are involved. The two most important nutrient absorption carriers for drugs are the oligopeptide carrier (hPepT1) and the amino acid transport family. These carrier proteins have a high transport capacity in the human small intestine, and they seem less likely to be involved in direct food–drug interactions, unless high doses are given together with a protein-rich meal. For instance, it has been reported that a protein-rich meal does not affect the BA of levodopa, a drug that is transported by the several amino acid carriers in the human intestine, and this has been interpreted that the amino acids are absorbed rapidly, the risk for interaction is reduced and the nutritional transporter has a high capacity (Lennernäs et al 1993). The nutritional status could also cause transcriptional activation of the PepT1 gene by selective amino acids and dipeptides in the diet (Shiraga et al 1999). It has also been reported that the integrated response to certain stimuli may increase PepT1 activity by translocation from a preformed cytoplasm pool (Thamotharan et al 1999). This short-term regulation of drug transport requires further research.

Potential future extensions

Class I drug substances today are quite rare in pharmaceutical development, which limits the broad use of the BCS in drug development. For instance, the proportion of class I compounds in the development phase for oral immediate-release formulations at AstraZeneca was less than 10% in 2001. It has also been recognized that the present application represents a deliberately conservative approach and proposals for extensions have been discussed since original publication of the BCS (Amidon et al 1995; Yu et al 2002). For example, in a recent paper it was suggested that the requirement of highest pH for the solubility measurements could be changed from 7.5 to 6.8 since the latter is more relevant for the pH in the upper gastrointestinal tract (Yu et al 2002). This revision would

thus somewhat relax the requirements for basic drugs. It has also recently been suggested that for acidic drugs the boundaries for solubility are too restrictive (pH 1.0–7.5) and might be narrowed down to between pH 5.0 and pH 7.4 (Yazdanian et al 2004). Yazdanian et al (2004) reported that, based on the current definition, 15 of 18 acidic NSAIDS were classified as class II compounds. If only a neutral pH was used, 15 of the NSAIDS would be classified as class I drugs, which may suggest the boundaries are too restrictive for solubility. Based on these findings, the authors suggested that there might be a need for an intermediate solubility classification for highly permeable ionizable compounds, especially when given in low doses (Yazdanian et al 2004). Another proposal in the paper by Yu et al (2002) was to reduce the high permeability definition from 90% to 85% fraction absorbed based on observations that many drugs that are considered completely absorbed provide experimental values below 90% (i.e. 90% seems to be too rigid a criteria considering the precision of the experimental methods). Other, more radical, relaxations of criteria, such as including class III drugs or class II drugs using simulated intestinal media with bile acids or increasing volumes and further reducing the pH interval in solubility measurements, will most probably require further research and analysis of past experiences (Bonlokke et al 2001; Kostewicz et al 2004; Porter et al 2004). For instance, to better predict the effects of food on the BA/BE of drugs and drug products from in-vitro data, a dissolution medium that simulates the initial composition of the postprandial stomach has been developed (Klein et al 2004).

It has been suggested by Blume & Schug (1999) that the waiver of in-vivo BE studies be extended to class III drugs. For immediate-release formulations of class III drugs (high solubility, low permeability), it is considered that the handling of the dosage form in-vivo be similar to that of an oral solution. Accordingly, intestinal membrane permeability is expected to be rate limiting in the overall drug absorption process. Therefore, the rate and extent of intestinal absorption is expected to be controlled by drug molecule properties and physiological factors, rather than by properties related to the pharmaceutical formulation, given that none of the pharmaceutical excipients used in the formulation have any relevant influence on gastrointestinal transit and permeability of drug (Yu et al 2002). However, what kind of in-vitro dissolution requirements should be set to ensure that the drug release has no significant impact on in-vivo BA is still unknown, and data available for supporting biowaivers of Class III drugs are still limited (Yu et al, 2001). In a recent study by Cheng et al (2004), a biowaiver was suggested by the in-vitro dissolution profiles and was justified by the in-vivo BE data; the demonstration of BE between two immediate-release tablets of the class III drug metformin in healthy Chinese subjects serves as an example for supporting biowaivers for such cases. Yu et al (2004) also showed that that a BCS class III drug, hydrochlorothiazide, had a much narrower 90% confidence interval than BCS class II drugs and therefore may be a better candidate for biowaiver. However, important concerns for BCS class III drugs are the effect of excipients on gastrointestinal transit and permeability (both passive and carrier-mediated uptake and efflux transport (Yu et al 2004). Further support for biowaiver of BCS class III drugs is recently available from Vogelpoel et al (2004), who showed that atenolol is a candidate for biowaiver, provided that the formulation contains well-known excipients.

It has also been proposed that dissolution rate should be a more relevant variable than saturation solubility for BCS since the in-vivo absorption process is rather a dynamic rate-controlled process than a process at equilibrium conditions (Yu et al 2004). The rotating disc method, where the pure drug is compressed into a rotating disc holder to provide a constant area exposed to the dissolution medium, is the method of choice for such investigations (USP). It was shown in a limited set of drug substances that, at a rotation speed of 100 rev min^{-1}, class I and III drugs had a dissolution rate faster than 0.1 mg min^{-1} cm^{-2} (Yu et al 2001).

Extensions of the BCS beyond the oral immediate-release area have also been suggested (e.g., to apply the BCS in the extended release area). However, this will provide a major challenge since the release from different formulations will interact in different ways with in-vitro test conditions and the physiological milieu in the gastrointestinal tract. For example, the plasma concentration–time profile differed for two felodipine extended-release tablets for which very similar in-vitro profiles had been obtained, despite the fact that both tablets were of the hydrophilic matrix type based on cellulose derivates (Abrahamsson et al 1994). This misleading result in-vitro was due to interactions between the test medium and the matrix-forming polymer of no in-vivo relevance. The situation for extended-release formulations would be further complicated by the need to predict potential effects of food on the drug release in-vivo.

Although the present use of the BCS is limited, extensions of applications should clearly be made without jeopardizing the quality of products on the market. This would more likely be achieved within the area of oral immediate-release than for extended-release formulations.

Conclusion

In this review, we have discussed and emphasized the importance of the fundamental factors in the BCS, solubility and intestinal permeability for oral drug absorption. The main regulatory impact today is the use of the BCS as a framework for identifying drugs for which in-vitro dissolution testing could replace in-vivo studies to determine BE. Extensions of this approach to cases other than immediate-release formulations of the rather rare class I drugs would significantly enhance the impact of the BCS. Some drugs that fall into class II and III would be able to be regulated by the BCS rather than traditional BE testing. In a recent analysis of the 123 WHO oral drugs available in immediate-release dosage forms, it was reported that they were classified as BCS class I, class II, class III and class IV drugs using dose number and Log P as follows: 24.4% class I, 17.1% class II, 29.3% class III

and 12.2% class IV. These results suggest a satisfactory BE test for over 60% of the drug products of the WHO (class I and III drugs) and clearly demonstrate a very practical usefulness of the BCS (Kasim et al 2004).

In expansion into extended-release dosage forms, the product quality assurance must not be jeopardized and it must be carefully examined. It is clear that more basic research must be focused on the interplay between gastrointestinal physiology, dosage form performance and human intestinal absorption.

The future application of the BCS is most likely increasingly important when the present framework gains increased recognition, which will probably be the case if the BCS borders for certain class II and III drugs are extended. The future revision of the BCS guidelines by the regulatory agencies in communication with academic and industrial scientists is exciting and will hopefully result in an increased applicability in drug development. Finally, we emphasize the great use of the BCS as a simple tool in early drug development to determine the rate-limiting step in the oral absorption process, which has facilitated the information between different experts involved in the overall drug development process. This increased awareness of a proper biopharmaceutical characterization of new drugs may, in the future, result in drug molecules with a sufficiently high permeability, solubility and dissolution rate, and that will automatically increase the importance of the BCS as a regulatory tool over time.

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