Drug Absorption

2

Alan Talevi and Carolina Leticia Bellera

2.1 Introduction

► **Definition** Absorption is a mass transfer process that involves the movement of unchanged drug molecules from the site of absorption (which often, but not always, coincides with the site of administration) to the bloodstream. We will focus on *systemic absorption*, i.e., the movement of the drug molecules to general circulation.

The extent and rate of drug absorption depend on several factors including physicochemical features of the drug, the pharmaceutical dosage form, and anatomical and physiological factors. In many cases, crossing epithelia or endothelia will become the rate-limiting step of the whole absorption process. For that purpose, drug molecules might exploit the paracellular way or, more commonly, the transcellular way. In the latter, the critical step of drug absorption will usually involve crossing cell membranes. Most drug molecules will cross the cell membrane by simple diffusion, which can be appropriately modeled using Fick's first law. Molecules resembling physiological compounds might also be absorbed by carrier-mediated transport which, to some extent, can be modeled by Michaelis-Menten equation.

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Absorption involves overcoming different biological barriers but also surviving pre-systemic chemical or biochemical modifications (e.g., hydrolysis in the gastric medium and/or first-pass metabolism). By considering that a drug molecule has been absorbed as soon as it has left the heart left ventricle with no chemical modifications (i.e., no covalent bond formation or breakage), any pre-systemic phenomenon undergone by drug molecules can be regarded as constitutive of the absorption process.

Establishing the difference between *systemic* and *topical* administrations of a drug is probably the first step to explain the importance of drug absorption. Systemic administration of a medication uses the circulatory system to distribute the drug molecules. In contrast, topical medications are applied to a particular place in the body to treat an ailment in a local manner (typically, the skin or mucous membranes, though topical medications also involve inhalational medications, eye drops, or ear drops, among others). Note that, when systemic administration is used, the whole body (and not only the site of action) is potentially exposed to drug molecules. Conversely, topical administration is intended to circumscribe drug exposure to the affected site of the body and surroundings. Frequently, adverse reactions to medications are due to the interaction of the drug with elements of the body different from the drug molecular target (i.e., off-target interactions). Adverse reactions to topical medications are often local effects, such as irritation or localized allergic reactions. *Systemic* administration is more likely to involve more severe adverse reactions than *topical* administration. Nevertheless, *topical* administration is not always viable when the molecular target is situated in difficult-to-reach organs. Special mention must be made to targeted therapies, where drug molecules are preferably delivered to the site of action through targeted delivery systems (e.g., targeted nanosystems, viral vectors). This later concept will be separately discussed in Chap. 7, which focuses on nanocarriers and drug delivery. Accordingly, this chapter will address conventional medications only.

It follows from the previous discussion that drug absorption is a crucial process whenever resorting to systemic administration, while it is not required (and often, unwanted) when using topical administration: for drugs applied at their target, such as local anesthetics, absorption often terminates the therapeutic effect (Sultatos 2011).

2.2 Factors Affecting Drug Absorption

It should be remembered that, generally speaking, drug molecules are not administered in isolation but incorporated in a pharmaceutical dosage form. However, as discussed in the previous chapter, to overcome biological barriers, the drug must be in its free form; that is, it must have been released from the pharmaceutical carrier and in solution (Sultatos 2011). Only then drug molecules will be able to initiate their movement toward systemic circulation. The previous applies for conventional dosage forms but might not be necessarily true for last-generation pharmaceutical carriers as nanocarriers. Also, note that some dosage forms may provide the drug already in solution (e.g., pharmaceutical solutions).

► **Important** When dealing with conventional dosage forms, drug release and dissolution are prerequisites for absorption to occur.

Based on the previous concept and the definition of absorption that has been provided, absorption will depend on:

- *Physicochemical properties* of the drug, fundamentally: Its aqueous solubility, its ionization constant(s), its permeability through biological barriers that it shall encounter before arriving at general circulation (which, in turns, depends mostly on molecular size and lipophilicity), and its affinity to biological systems such as enzymes or transporters that it may come across during absorption.
- The *pharmaceutical dosage form*, which directly influences drug release. Occasionally, the pharmaceutical carrier might include components capable of modulating the drug uptake and/or metabolism. For instance, it has been reported that some common excipients can modify the expression and/or activity of drug transporters and cytochrome p450 enzymes (Tompkins et al. 2010; Hodaei et al. 2014; Al-Mohizea et al. 2014).
- The *anatomical and physiological characteristics* of the site of absorption, including expression levels of drug transporters, pH, anatomical adaptations that may favor absorption, and the characteristics of the tight junctions between adjacent cells of the absorptive tissue. Note that considerable physiological changes that impact on drug absorption can be observed during pathological processes and also with age.
- The *way the medication has been administered*. For instance, the extent and rate of absorption for a drug given through the oral route might vary if the medication has been administered before, along with or immediately after a meal in comparison with administration on an empty stomach. Transdermal absorption could be enhanced by means of different physical stimuli, such as local heat or mechanical friction.

The previous factors should be jointly considered when designing dosage forms or when assessing a medication from a biopharmaceutical viewpoint. The pharmacokinetic processes undergone by a drug after administration are often described through the LADME scheme, liberation, absorption, distribution, metabolism, and excretion, which are considered in separate chapters in the first part of this book.

► **Definition** The acronym LADME refers to liberation, absorption, distribution, metabolism, and excretion. These are the pharmacokinetically relevant processes that a drug undergoes after entering the body. Whereas these processes generally follow such sequence, *they should not be regarded as discrete events*. In other words, one process is still occurring, while the next one begins, and in certain occasions (e.g., sustained release delivery systems), all five processes may be occurring simultaneously (in parallel) to different drug molecules.

Conventional drug delivery systems can only impact, in a direct manner, on the processes of Liberation and Absorption, and only indirectly in the remaining ones, by influencing the rate and extent of drug release and absorption. Last-generation delivery systems, in contrast, could possibly modify in a direct manner all the LADME processes.

2.3 The Fluid Mosaic Model and Beyond

Crossing the selective barrier posed by cell membranes is in general the more relevant mass transfer process during absorption. To appreciate the role of biological membranes and tissue barriers in drug absorption, it is convenient to begin with an understanding of cell membranes. While we assume that the reader is familiarized with the generalities of the cell membrane architecture, we will overview them in order to provide a context for understanding drug absorption mechanisms. *Note that the principles governing absorption are similar to those governing distribution*. Accordingly, this and the subsequent sections will be of much value for the comprehension of both Chaps. 2 and 3.

The fluid mosaic model of the cell membrane, devised by Singer and Nicolson (1972) based on thermodynamic considerations and experimental evidence, proposes that phospholipids are the main constituent of cell membranes. This type of lipid (Fig. 2.1) generally consists of two hydrophobic fatty acid "tails" and a hydrophilic "head" consisting of a phosphate group. The fatty acids and the phosphate group are esterified with a glycerol molecule. The phosphate groups can be modified with simple organic molecules such as choline.

The phospholipids provide a mosaic structure predominantly arranged as a fluid bilayer (the matrix of the mosaic), with their phosphate groups in contact with the aqueous phase (either the extracellular medium or the cytosol), while the nonpolar fatty acid chains are sequestered away from contact with water and oriented to the inner space of the bilayer. Many integral proteins (or occasionally, glyco- or lipoproteins) can be found embedded in the membrane (more or less buried in its hydrophobic interior). Integral proteins are referred to as transmembrane if they

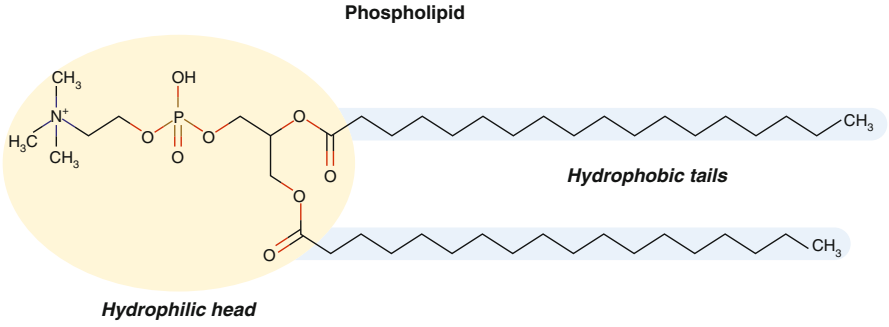

Fig. 2.1 General structure of a phospholipid, essential constituent of a cell membrane

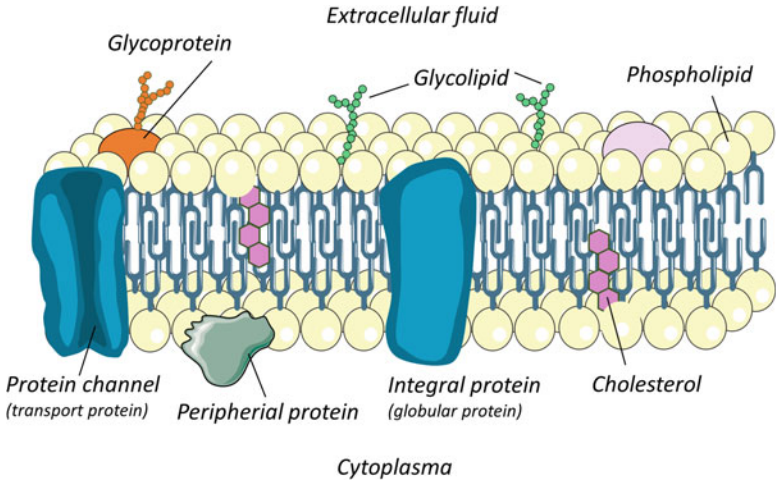

Fig. 2.2 Fluid mosaic model

extend from one side of the membrane to the other. Integral proteins are amphipathic, with highly polar groups protruding from the membrane into the aqueous phase and nonpolar groups embedded within the hydrophobic core of the bilayer. Additionally, peripheral proteins bound to the membrane by weak interactions and not strongly associated with membrane lipids can also be found. Membrane proteins present comparatively high amount of α-helical domains, in contrast with soluble globular proteins that in general exhibit a smaller fraction of α-helix in their native structure.

The arrangement of cell membranes (Fig. 2.2) acts as a bidimensional fluid that, in principle, allows free lateral diffusion of the membrane components. Cholesterol plays a much relevant role regulating membrane fluidity and conferring structural stability.

The model by Singer and Nicolson has been expanded to include important elements of the membrane architecture; some of which had already been envisaged by Singer and Nicolson themselves (Alberts et al. 2002). Among those elements, we may mention membrane asymmetry, the existence of lipid rafts (specialized areas in membranes where proteins and some lipids, primarily sphingolipids and cholesterol, are concentrated), and flip-flop movements.

The cell membrane is permeable to small nonpolar molecules, whereas molecules with high polarity (among them, charged ones), high molecular weight, and/or high conformational freedom will have difficulties crossing it. Cells have developed specialized transport mechanisms by using membrane proteins that include channels and carriers. Embedded in biological membranes, there are carriers that facilitate the entry of their substrates to the cell (*uptake transporters*) and carriers that export their substrates (*efflux transporters*). Channels and some types of carriers carry substrates down a concentration gradient, without direct or indirect ATP consumption. Other carriers require energy to transport their substrates. In some very specific cases,

molecules interacting with cell surface receptors trigger the formation of endocytic vesicles, which carry the receptor-bound molecules into the cell (*endocytosis*) or through the cell (transcytosis). This form of transport is more frequent for large molecules (e.g., some therapeutic proteins) or advanced drug delivery systems (e.g., nanoparticles). The combined contribution of all the aforementioned elements (lipid bilayer, channels, uptake and efflux carriers, endocytosis) determines the selective permeability of a given cell to a given molecule. Selective permeability is crucial to regulate the cell inner microenvironment.

2.4 How Can Drugs Be Absorbed?

In principle, any substrate being absorbed could potentially use the paracellular and/or the transcellular transport (Fig. 2.3). The paracellular transport refers to the transfer of substances across the epithelium or endothelium by passing through the intercellular space between adjacent cells. Paracellular transport is exclusively passive; which substances can use this route of transport depends on the tight junction characteristics for the considered epithelium or endothelium. For instance, particularly tight junctions at the blood-brain barrier preclude the paracellular route.

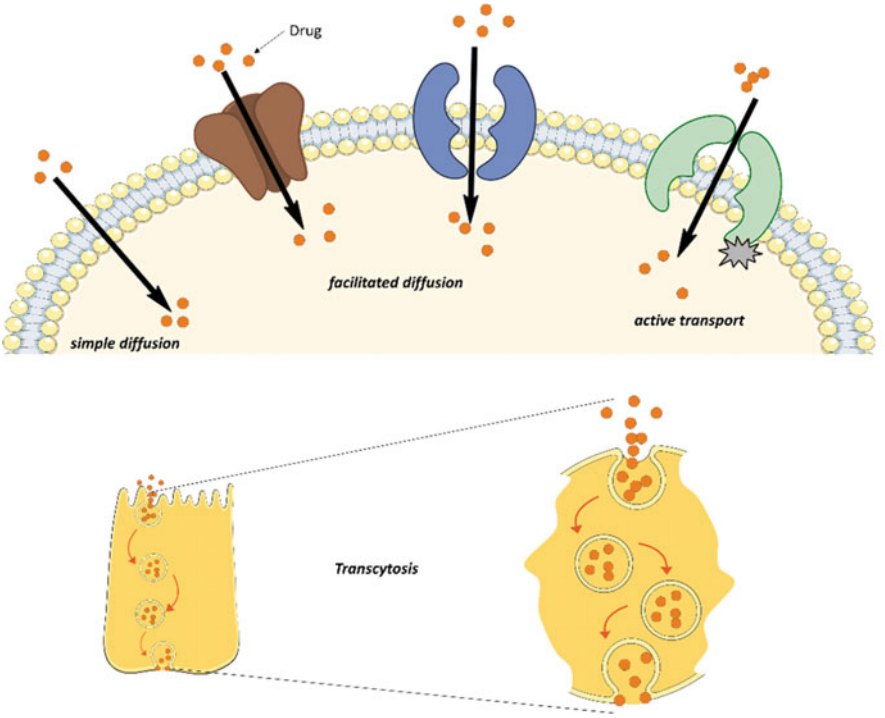

Fig. 2.3 Ways that a drug might employ, depending on its physicochemical and biochemical properties, to cross epithelia or endothelia

In contrast, other tissues present "leaky" tight junctions, thus favoring the paracellular route. When the paracellular route is available, small polar molecules are preferably transported through it.

► **Definition** The tight junctions are protein structures which form a selective barrier in the paracellular space between epithelial and endothelial cells, limiting movement of ions, solutes, and water. Tight junction strands represent an impermeable barrier, as a "wall," but a series of permeable aqueous "pores" perforate such wall.

Whereas the structure of the tight junctions is complex and dynamic and varies among different epithelia and capillary beds (González-Mariscal et al. 2003; Bazzoni 2006), evidence points to tight junction-associated claudins as the basis for the selective size, charge, and conductance properties of the paracellular pathway (Van Itallie and Anderson 2004).

Tight junction permeability can be significantly affected by pathological stimuli, but it is also actively investigated as a possible way to enhance drug delivery of hydrophilic drugs (González-Mariscal et al. 2017).

Conversely, the transcellular route offers a wide spectrum of possibilities that expand the diversity of the substrates that may be uptaken by the cells. These possibilities include *simple diffusion* (passive and without intervention of any membrane protein), *facilitated diffusion* (down the concentration gradient either through protein channels or carrier proteins), active transport (up the concentration gradient with energy consumption), and transcytosis.

► **Important** By far, the most frequent transfer mechanism involved in drug transport through cell membranes is passive diffusion. To diffuse, drugs should be sufficiently hydrophobic to partition into the lipid bilayer of the plasma membrane. Since most drugs are either weak acids or bases, the ability of a given drug to partition into the membrane will be highly modified by the pH conditions.

2.5 Simple (or Free) Diffusion

In simple diffusion, drug molecules will use the transcellular route spontaneously, without involvement of any membrane protein and down the concentration gradient. Since crossing the cell membrane is usually the slowest step of the whole absorption process (thus governing its kinetics), we will focus on the concentration gradient through the cell membrane. The kinetics of simple diffusion can be adequately modeled using Fick's first law. For the sake of mathematical simplicity, let us imagine the absorption site as a well-defined compartment (which it is usually not) and that a certain amount Q of the drug is homogeneously dispersed (in solution) at the absorption site. In this way, the only diffusion process that we will pay attention to is the movement of the drug molecules from the site of absorption to the inside of

the cells. In other words, we will study the drug flux in the orthogonal direction to the epithelium (or endothelium) surface (a direction that we will arbitrarily name x direction). The *instantaneous* rate of absorption corresponding to the change in Q (dQ) during infinitesimal time period dt will be thus given by the following expression:

$$\frac{dQ}{dt} = D \times P \times S \times \frac{dC}{dx} \tag{2.1}$$

D alludes to the coefficient of diffusion through the membrane, and it is expressed in units of surface over units of time (e.g., cm^2/s). It depends on molecular features such as molecular size and shape. For instance, large molecules will diffuse more slowly than small ones, being thus associated to a smaller D. P represents the partition coefficient of the drug between the cell membrane and the surrounding aqueous environment. S refers to the total absorption surface, and dC/dx is the concentration gradient (concentration/distance units). Equation (2.1) shows that, at any moment, the (instantaneous) rate of absorption will be directly proportional to the concentration gradient, which is the driving force of the process.

If we call the thickness of the membrane δ (Fig. 2.4) and we assume that the concentration falls linearly through such thickness, by integrating expression (2.1), we get to:

$$\frac{dQ}{dt} = D \times P \times S \times \frac{\Delta C}{\delta} \tag{2.2}$$

If we now call A the concentration of the drug in the left compartment (representing the site of absorption or donor compartment) and we call B the drug concentration in the right (acceptor) compartment, Eq. (2.2) now takes the form:

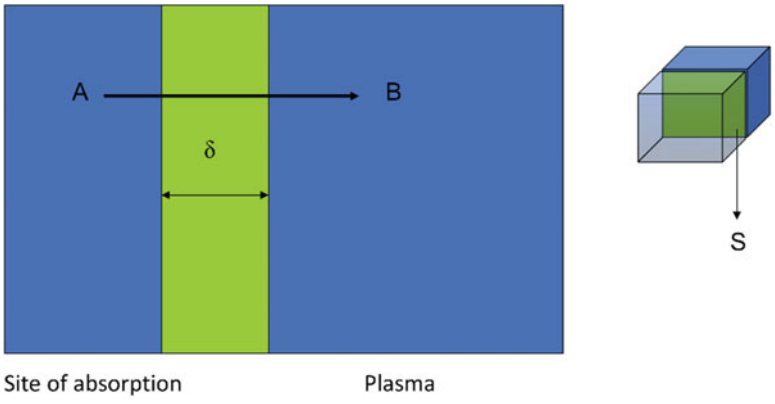

Fig. 2.4 A simple model to describe drug absorption. The site of absorption corresponds to the left (donor) compartment, whereas the right compartment represents the acceptor compartment (that contains absorbed drug)

$$\frac{dQ}{dt} = D \times P \times S \times \frac{(B - A)}{\delta} \tag{2.3}$$

Dividing both sides of the equation by V, the volume in the site of absorption:

$$\frac{dQ}{Vxdt} = \frac{D \times P \times S}{V \times \delta} \times (B - A) \tag{2.4}$$

Note that the factor $(B - A)$ determines if expression (2.4) acquires a negative or positive sign (the remaining factors on the right-hand side of the equation are positive constants at a given pressure and temperature conditions).

Questions

Is the $(B - A)$ factor in Eq. (2.4) smaller or greater than zero?

Answers

Provided that, according to the example in Fig. 2.4, the diffusion is proceeding from the left compartment (the one that has been assimilated to the site of absorption or donor compartment in the example) to the right compartment, A must necessarily be greater than B. Accordingly, the alluded factor is smaller than zero.

Questions

When will the diffusion end?

Answers

In principle, if the considered system were a close system, diffusion would end when A equals B. Take into consideration that, at that point of dynamic equilibrium (identical concentrations at both sides of the diffusion barrier), the mass units of the drug might well be very different in both compartments, since they depend on the compartments' volumes. If for a moment we consider the intravascular fluid as the acceptor compartment, it can be accepted that the volume of the acceptor compartment is much larger than the one of the donor compartment, thus accepting, in a hypothetic equilibrium condition, much more mass units of the drug than the donor compartment.

Moreover, neither the "acceptor compartment" nor any biologic system can be thought of as close systems. As we will discuss in brief, this condition will contribute to an efficient absorption of the administered dose.

Let us express $(B - A)$ as $-(A - B)$, and let us focus on the variation in concentration at the absorption site, instead of the variation in mass units:

$$\frac{dQ}{Vxdt} = \frac{dA}{dt} = -\frac{D \times P \times S}{V \times \delta} \times (A - B) \tag{2.5}$$

dA/dt has, undoubtedly, negative sign (D, P, S, V, δ are positive quantities, and we have already established that $A > B$). This is reasonable (for the diffusion to occur in the hypothesized sense, the concentration at the site of absorption will be diminishing as time goes by). Also, observe that the instantaneous rate of absorption is directly proportional to the absorptive surface area. This explains the importance of some anatomical adaptations that favor absorption in certain organs by increasing the surface area (i.e., Kerckring valves, villi, and microvilli at the small intestine). The concept is also useful for the design of transdermal dosage forms (see the example below).

Example

Nicotine replacement therapy is one of the most frequent treatments for smoking cessation. It is available in a diversity of dosage forms, including gum, sublingual tablets, nasal spray, inhaler, and transdermal patches.

Table 2.1 shows the impact of nicotine transdermal patches surface area on nicotine bioavailability (the table includes mean data from six healthy male smokers ± the standard deviation). The bioavailability is measured through the area under the curve in a plot of drug concentration in blood plasma versus time (AUC) and the peak plasma concentration (C_{max}). AUC (from zero to infinity) is an indicator of the total amount of drug that has reached general circulation (total drug exposure over time); C_{max} reflects both the quantitative and kinetic aspects of bioavailability. T_{max} represents the time at which C_{max} is observed, and it speaks of the kinetic component of bioavailability (the smaller T_{max}, the higher the absorption kinetics). The data shown in the table has been extracted from Sobue et al. (2006). The reader may visit Chap. 10 for an extensive discussion on bioavailability.

Note that the bioavailability for a surface area of 40 cm^2 roughly doubles that for a surface area of 20 cm^2

Table 2.1 Comparison of nicotine bioavailability between 20 and 40 cm^2 nicotine transdermal patches

	AUC (ng h/mL)	C_{max} (ng/mL)	T_{max} (h)
Transdermal patch 20 cm^2	155.13 ± 29.51	9.15 ± 1.79	13.3 ± 3.4
Transdermal patch 40 cm^2	280.96 ± 31.08	17.25 ±−1.60	8.2 ± 5.2

Mean ± SD

Let us go back to Eq. (2.5). As previously stated, P represents the partition coefficient between the majorly hydrophobic cell membrane and the aqueous phase. A lipophilic drug will display a high affinity to the membrane and will be linked to a high rate of absorption (however, an extremely lipophilic drug will possibly tend to remain "trapped" in the membrane, which is termed *membrane retention*). Beside this issue, also consider that a drug must be in solution to be absorbed: as it is discussed in other chapters, drugs with low solubility will tend to have dissolution problems. Accordingly, adequate hydrophilic-lipophilic balance is often pursued when designing new drugs intended for oral administration. D and P vary from drug to drug. For a given drug and at a given moment, D, P, S, δ, and V are constants and can then be grouped together and replaced with a single constant that we will refer as the absorption rate constant through the lipid bilayer $K_{\text{a,memb}}$. $K_{\text{a,memb}}$ has units of s^{-1}:

$$\frac{dA}{dt} = -K_{a,\text{memb}} \times (A - B) \tag{2.6}$$

▶ **Important** The lipid bilayer of the cell membrane is often considered as the fundamental barrier that a drug must overcome to reach the intravascular space. Such assumption is in general well-founded, though the number of diffusional barriers that the drug must actually overcome greatly exceeds the bilayer. For instance, if we think of the intestinal epithelium, it is covered by a mucus layer rich in mucins, with gel-like properties (Johansson et al. 2013). After crossing the apical membrane, the drug must also cross the cytosol, the basolateral membrane, the connective tissue, the interstitial tissue, and the endothelium of the capillary bed. The drug will then be carried to the heart, and it will finally reach general circulation.

It can be observed that the aforementioned (sub)processes are serial: if any of them is slower than the others, it will be the limiting-step and govern the kinetics of the whole absorption process. In general, it is safe to assume that crossing the cell membranes is such a rate-limiting step (the hydrophobic core of the cell membrane is 100–1000 times more viscous than water!).

2.5.1 Sink Condition and Absorption

The diffusion processes linked to absorption will conclude once the drug has reached the capillary beds irrigating the site of absorption. As blood is circulating and removing drug molecules from the site of absorption, and taking into consideration that a substantial amount of the drug in plasma is complexed with plasma proteins (as further discussed in the next chapter), the term B in Eq. (2.6) will in general tend to be small in comparison with A, and, in most scenarios, it can be neglected. This condition is known as *sink condition* and allows simplifying Eq. (2.6):

$$\frac{dA}{dt} = -K_{a,\text{memb}} \times A \tag{2.7}$$

This simple step has very important implications. The resulting expression (2.7) clearly represents first-order kinetics: the instantaneous rate at which the concentration of drug diminishes in the site of absorption is at any moment proportional to the remaining concentration of the drug in the site of absorption. The sink condition positively impacts on the extent of absorption, since it helps sustaining a concentration gradient (driving force of the diffusion process) and thus favors a complete absorption of the administered dose.

There are some conditions for which the sink condition may not apply, consequently reducing the extent of absorption. For instance, during physical exercise, the viscera perfusion is reduced, negatively impacting on the bioavailability of some drugs (the less permeable ones).

Figure 2.5 represents the instantaneous rate of absorption versus the concentration of drug in the site of absorptions: absorption by simple diffusion can be regarded, doubtlessly, as a linear process. A similar behavior would be found experimentally when performing permeability models if charting the initial rate of absorption versus initial concentration of the drug.

► **Important** Simple diffusion is a linear and non-saturable transport mechanism.

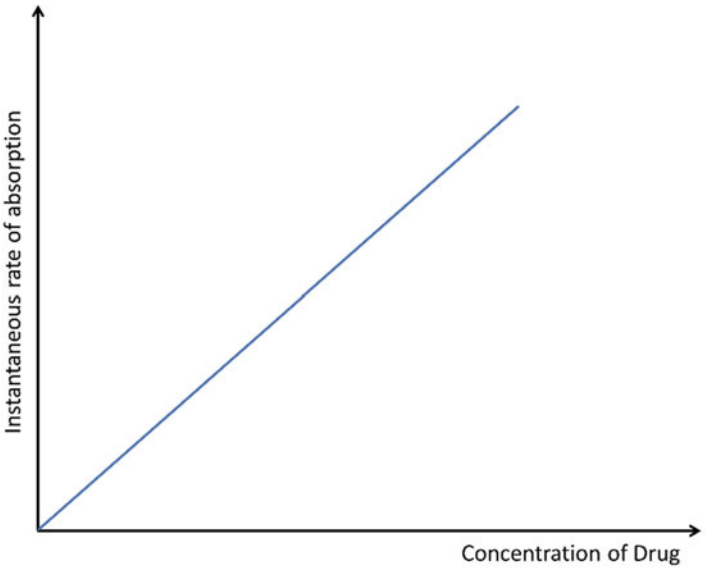

Fig. 2.5 Chart of instantaneous rate of absorption versus the (instantaneous) concentration of drug in the site of absorption, for a drug being absorbed through a linear process

2.5.2 The Impact of pH on Drug Absorption

As said, most of the known drugs are weak acids or bases. For a weak acid (for instance, aspirin, $pK_a = 3.5$), the ionization equilibrium can be written as

$$HA(aq) + H_2O(l) \rightleftarrows H_3O^+(aq) + A^-(aq)$$

Notice that, following Le Chatelier's principle, in an alkaline media that consumes H_3O^+, the equilibrium will shift to the right, thus increasing the concentration of the charged species A^-. Conversely, in acid media the equilibrium will shift to the left, increasing the concentration of the non-charged species HA. Interestingly, while the solubility in an aqueous media will be higher for charged species, the permeability of charged species through the cell membrane by passive diffusion will be low. Moreover, the electrical potential gradient will also impact on the transport of charged species, in which case the Nernst-Planck equation will be useful to determine the flux across the barrier in question.

From the previous equilibrium, the corresponding acid ionization constant is:

$$K_a = \frac{[H_3O^+][A^-]}{[HA]} \tag{2.8}$$

Taking the logarithm of both sides of the equation easily leads to the Henderson-Hasselbalch equation:

$$\log_{10} K_a = \log_{10} [H_3O^+] + \log_{10} \frac{[A^-]}{[HA]} \tag{2.9}$$

Then:

$$-pK_a = -pH + \log_{10} \frac{[A^-]}{[HA]} \tag{2.10}$$

Finally:

$$pH = pK_a + \log_{10} \frac{[A^-]}{[HA]} \tag{2.11}$$

A very similar development can be achieved for a base:

$$B(aq) + H_2O\ (l)\ \rightleftarrows BH^+(aq) + OH^-(aq)$$

Here, the base ionization constant is:

$$K_b = \frac{[OH^-][BH^+]}{[B]} \tag{2.12}$$

This leads to:

Table 2.2 Negative effect of lansoprazole coadministration on atazanavir bioavailability

	AUC_{0-24} (μM · h)	C_{max} (μM)	T_{max} (h)
Atazanavir administered alone	16.3 ± 9.0	3.2 ± 1.7	2.8 ± 0.75
Atazanavir administered after lansoprazole	0.85 ± 1.8	0.13 ± 0.19	3.1 ± 1.0

Mean ± SD

$$\mathrm{pOH} = \mathrm{p}K_b + \log_{10}\frac{[\mathrm{BH}^+]}{[\mathrm{B}]} \tag{2.13}$$

Have in mind that pH + pOH equals 14.

Whereas the effects of pH variations on solubility and permeability are relatively straightforward for drugs having a single ionizable group within the physiologically relevant range of pH, the analysis gets more complicated when addressing drugs with many ionizable functions, where such ionizable groups will gain protons in a serial manner when pH gradually drops and vice versa. Accordingly, when a drug has many ionizable groups, the number of ionization states gets multiplied, and even zwitterionic species with no net charge can appear.

Example

Clinically, significant drug-drug interactions involving antiretroviral medications occur in up to 40% of HIV patients on treatment; the consequences of such interactions include fluctuations in antiretroviral plasma levels leading to toxicity, diminished efficacy, and increased risk of drug resistance (Lewis et al. 2016). Acid-reducing agents are commonly used co-medications by HIV-1-infected patients receiving antiretroviral treatment. Furthermore, acid-reducing therapy medications are widely used, and many of them are available over the counter in most countries.

Several reports demonstrate significant drug-drug interactions between acid-reducing medications and antiretrovirals. For instance, Tomilo et al. (2006) have reported a mean reduction of 94% in atazanavir bioavailability in nine healthy volunteers receiving such antiretroviral drug coadministered with lansoprazole (Table 2.2). The loss in solubility for atazanavir at increased pH values is considered responsible for this effect.

Studies suggest that some of the interactions between antiretroviral medications and acid-reducing agents may be mitigated by temporal separation of dose administration (Falcon and Kakuda 2008).

2.6 Facilitated Diffusion

If we pay close attention to expression (2.4), it is clear that simple diffusion will not be favored for certain chemical compounds. That is the case with large or highly hydrophilic molecules. How does the cell manage to transport such molecules (e.g., glucose from the intestinal lumen) through the cell membrane, if required?

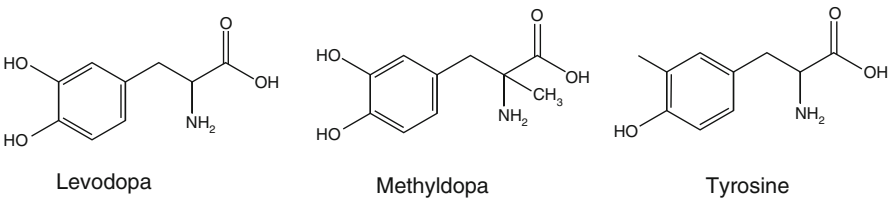

Fig. 2.6 The chemical similarity between the therapeutic agents levodopa and methyldopa explains why these drugs can use amino acid or peptide carriers for their absorption

Facilitated diffusion (also known as passive-mediated transport) is the process of spontaneous passive transport of molecules or ions across a biological membrane via specific transmembrane integral proteins.

There are essentially two types of protein structures facilitating diffusion. *Channel proteins* form open pores in the membrane, allowing small molecules of the appropriate size and charge to pass freely through the lipid bilayer. As previously stated, channel proteins also permit the passage of molecules between adjacent cells connected at tight junctions. In contrast, *carrier proteins* bind specific molecules to be transported and then undergo conformational changes that allow the molecule to pass through the membrane and be released on the other side. They are responsible for the facilitated diffusion of sugars, polar amino acids, and nucleosides across the plasma membranes of most cells.

▶ **Important** Since uptake carriers have evolved to transport very specific substrates with physiologic role through the membrane, only drug resembling such substrates will be able to exploit carriers as transport mechanism. Some classical examples of drugs that use uptake carriers are 5-fluorouracil, levodopa (antiparkinsonian), and methyldopa (antihypertensive). Notice the resemblance of levodopa and methyldopa to the amino acid tyrosine (Fig. 2.6).

A very important difference between facilitated diffusion and free diffusion is that the number of substrate molecules that can be transported by time unit depends on the number of channel or carrier copies available per cell. Accordingly, this kind of transport is saturable. The temperature dependence of facilitated transport is substantially different due to the presence of an activated binding event, as compared to free diffusion where the dependence on temperature is mild. Usually, variation of transport rate upon temperature changes will be used as experimental evidence of mediated transport.

The kinetics of mediated transport can be described by the Michaelis-Menten equation:

$$\frac{dA}{dt} = -\frac{V_m \times A}{K_m + A} \tag{2.14}$$

where V_m is the maximum rate of transport, which depends on the level of expression and the turnover number of the transporter, and K_m stands for the Michaelis-Menten constant, which is inversely related to the binding affinity of the substrate (note that it equals the substrate concentration at which half of the V_m is observed). At very low substrate concentrations (far from the saturation condition), A can be neglected versus K_m, thus resulting in apparent first-order kinetics (the *apparent* term denotes that the kinetics is not strictly linear over the entire concentration range but only at small substrate concentrations):

$$\frac{dA}{dt} = -\frac{V_m \times A}{K_m} \tag{2.15}$$

Apparent first-order kinetics will be observed under therapeutic conditions whenever diffusion is facilitated by channels, since the number of available channels is usually very large.

Above saturation conditions (where, at a given time point, every copy of transporter will be interacting with a substrate molecule and the system will be working at maximum velocity), K_m could be neglected, and the transport process assumes apparent zero-order kinetics:

$$\frac{dA}{dt} = -V_m \tag{2.16}$$

Both parts of the Michaelis-Menten saturation curve are shown in Fig. 2.7. It should be emphasized that, strictly speaking, Michaelis-Menten equation does not fully represent facilitated diffusion, which is intrinsically dependent on substrate concentrations on both sides of the plasma membrane. The approach is, however, useful to describe apparent transport properties under specific conditions (e.g., initial uptake rates) (Panitchob et al. 2015). More complex mechanistic models are required to capture transporter behavior under diverse physiological conditions.

2.7 Active Transport

Generally speaking and from a mathematical perspective, active transport does not differ of the previously discussed description for facilitated diffusion, since it may be adequately modeled through a Michaelis-Menten kinetics. Some points should be considered, though.

First, active transport is mediated by carrier proteins that undergo important conformational changes to move its substrate(s) across membranes.

Second, it consumes energy in a direct or indirect manner to move its substrate (s) against the concentration gradient. Primary active transport utilizes energy in the form of ATP to transport molecules across a membrane against their concentration gradient. ATP-powered pumps contain one or more ATP-binding sites, which are present on the cytosolic face of the membrane. Conversely, secondary active

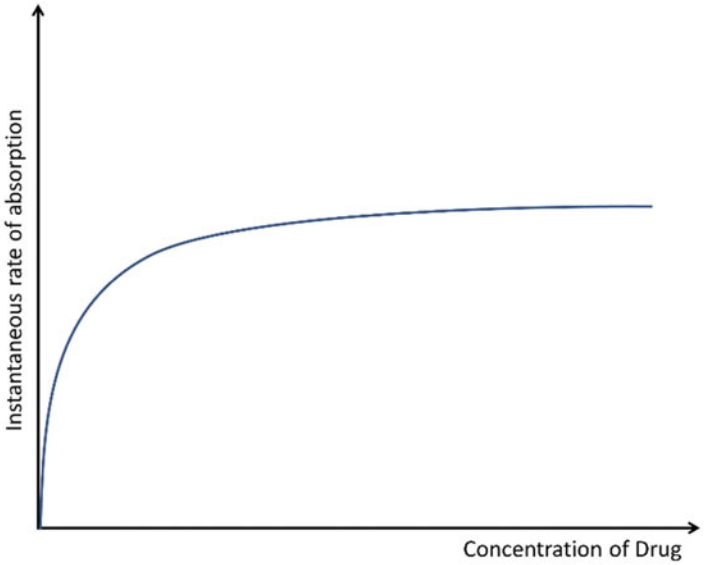

Fig. 2.7 Michaelis-Menten saturation curve

transport couples the movement of an ion (typically Na^+ or H^+) or molecule down its electrochemical gradient to the uphill movement of another molecule or ion against a concentration/electrochemical gradient. Thus, energy stored in the electrochemical gradient is used to drive the transport of another solute against a concentration or electrochemical gradient. The cotransported species may move in the same direction (symporters) or in opposite direction (antiporters). For example, transport of amino acids makes use of sodium-dependent symporters, of proton-motive force, and also of the gradient of other amino acids (Broër 2008).

Third, there are transporters that collaborate with drug absorption, whereas others oppose it. The former tends to display narrow substrate specificity and the latter, in contrast, wide substrate specificity. Consider that uptake transporters have evolved to help move specific molecules of physiologic value through membranes; conversely, efflux transporters have evolved to protect cells (or the organism) from strange, nonphysiologic compounds. We will discuss uptake and efflux transporters in a specific chapter of this volume.

Also, note that drug absorption might occur by a combination of transport mechanisms acting in parallel, e.g., active transport plus simple diffusion. In such case, the instantaneous rate of absorption will result from the addition of the mathematical equations explaining each of the individual processes involved in drug absorption.

Figure 2.8 illustrates the instantaneous rate of absorption versus concentration at the site of absorption, whenever the transport occurs by a combination of simple diffusion plus active transport or facilitated diffusion. Note that, as absorption proceeds, the concentration of substrate at the site of absorption will diminish, and

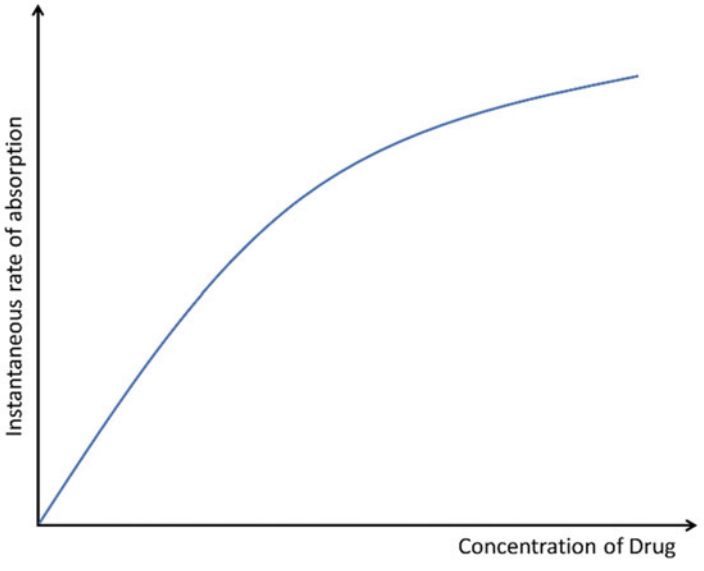

Fig. 2.8 Relationship between the instantaneous rate of absorption and drug concentration at the absorption site, for drug transport occurring by simple diffusion plus active transport or facilitated diffusion

the instantaneous rate of absorption will also go down. In other words, the rate of absorption will fall as the remaining (not yet absorbed) substrate at the site of absorption goes down.

Example

Parkinson's disease is one of the most common neurodegenerative diseases, characterized by motor and non-motor symptoms. It is characterized by the loss of dopaminergic neurons, with the consequent impairment of dopaminergic neurotransmission. Levodopa, an oral dopamine precursor, is used to restore dopamine levels at the central nervous systems. It is, so far, the most effective treatment for Parkinson's disease patients. Levodopa is rapidly metabolized to dopamine in the gastrointestinal tract by amino acid decarboxylase. This is not desired for two reasons: one, dopamine cannot penetrate the blood-brain barrier; two, peripheral conversion of levodopa to dopamine causes significant gastrointestinal side effects, such as nausea and vomiting. For these reasons, levodopa is commonly coadministered with peripherally acting amino acid decarboxylase inhibitors such as carbidopa and benserazide, which limit levodopa conversion to dopamine to the central nervous system.

The patients are likely to show reduced response to levodopa and develop motor complications such as dyskinesia and motor fluctuations (abrupt shifts between "on" and "off" states in which the symptoms are controlled and

uncontrolled, respectively). The motor fluctuations in patients in advance stages become closely connected with the rapid rise and fall in levodopa concentration after each dose. Fluctuating motor performance is the major source of disability in Parkinson's disease patients and drastically impairs their quality of life. Therefore, maximizing the therapeutic efficacy of levodopa is of utmost importance.

Levodopa, which closely resembles some dietary amino acids, is absorbed by a saturable facilitated large neutral amino acid transport system. Such transport is also responsible for the penetration of levodopa to the central nervous system, where it exerts its therapeutic action. The presence of large neutral amino acids from the diet either in the intestinal lumen or in blood may reduce levodopa absorption and distribution to the brain, in that order, since they compete with levodopa for the transporter.

Accordingly, protein-restricted diets have shown effective in ameliorating motor fluctuations (Wang et al. 2017; Barichella et al. 2017). Such diets include low-protein diets (in which protein consumptions are restricted to 0.5–0.8 g protein/per kilogram of body weight per day) and protein-redistribution diet (in which the patient consumes low-protein food such as cereal products, fruits, and vegetables for breakfast and lunch and are allowed high-protein food including eggs, legumes, fish, and meat for dinner). This maximizes daytime motor function, since it is considered that diurnal fluctuations limit the quality of life more than fluctuations at night.

Case Study

A 67-year-old patient with a 23-year history of Parkinson's disease and type 2 diabetes presented to an emergency department on New Year's Day. She had hosted a large New Year's Eve party the preceding evening. She was on levodopa/carbidopa therapy as well as two dopamine agonists (pramipexole and amantadine) and metformin. She had taken her medication at 10 pm on New Year's Eve, although it was unclear whether she had taken her tablets at 11 am that morning. A large protein-rich Chinese meal is consumed shortly before her last confirmed dose of treatment. On New Year's Day, she had gone to sleep at 11 am after cooking lunch. At 2 pm her son could not rouse her and called an ambulance. Blood sugar was 9.0 mmol/l. Paramedics arranged transfer to the hospital. She had a Glasgow coma scale score of 5/15. Her blood pressure was 130/78 mmHg, pulse 84 beats per minute, and blood oxygen saturation 98% on room air. There was no focal neurological deficit. A tomographic scan revealed a mild degree of cerebral atrophy but was otherwise normal. Routine hematology and biochemistry investigations were normal. Peripheral white cell count was normal. Treatment with intravenous acyclovir was commenced to cover possible viral encephalitis. Shortly prior to admission, she had experienced an increased frequency of falls. The patient

(continued)

recovered very rapidly following reinstitution of dopaminergic therapy via nasogastric tube.

Considering that severe motor "wearing off" phenomena can be life-threatening, in extreme cases resembling neuroleptic malignant syndrome with confusion, hyperthermia, and rhabdomyolysis discusses possible causes to the patient's condition. Take into consideration the results of hematology and biochemistry investigations and other tests performed on the patient. Based on a case report by Arulanantham et al. (2014).

2.8 Conclusions

- Drug molecules must be in solution to be absorbed (with the exception of drugs included in some last-generation pharmaceutical carriers, such as nanocarriers).
- The vast majority of the drugs are absorbed by simple diffusion and the use the transcellular way to cross epithelia and/or endothelia.
- The same mass transfer mechanisms studied in this chapter will explain the drug distribution process.
- For a drug to be absorbed by carrier-mediated transport, it must closely resemble the natural susbtrates of the involved carrier, i.e., compounds of physiological value (e.g., amino acids, sugars, nucleotides). The number of therapeutically relevant small molecules that use uptake carriers is relatively small.
- Simple difussion can be modeled using Fick's first law. Facilitated transport, in contrast, can be modeled by Michaelis-Menten kinetics, though in some cases more complex models will be required.
- Simple difussion is a non-saturable (linear) process. Facilitated transport and active transport require transmembrane proteins and are saturable processes. In many therapeutic situations, though, facilitated transport will be regarded as an apparent first-order kinetic process.
- Since most drugs are either weak acids or weak bases, their solubility and permeability will be highly dependent on the pH of the absorption site. Moreover, the electrochemical gradient should also be taken into consideration to study the absorption of charged species.
- Ionic species with net charge tend to be more soluble than neutral species; conversely, charged species are less permeable.

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Further Reading

The reader is referred to the wonderful volume by Carsten Ehrhardt and Kwan-Jin Kim for additional insight on the absorption process with a focus on experimental and computational models of absorption (*Drug Absorption Studies.* In Situ, In Vitro *and* In Silico *models*, Springer, 2008). The *Molecular Biopharmaceutics* volume by Bente Steffansen et al. is also highly recommended to study experimental models of drug absorption (*Molecular Biopharmaceutics*, Pharmaceutical Press, 2010)