A second method available for transporting a compound across a cellular barrier is active transport.[12] The gastrointestinal tract is, of course, designed to absorb nutrients using a variety of active transport system, and drug compounds that can take advantage of one of these systems enter the systemic circulation using these transport systems. Active transport systems in other types of cells can also be exploited for the transport of a compound across a biological membrane. In the majority of cases, a transmembrane protein is involved and energy, typically in the form of ATP, must be expended in order for transport to occur. This method is far less common than passive diffusion.

It is also possible for compounds to move across biological barriers via paracellular transport and endocytosis, but these mechanisms are far less common than passive diffusion. Endocytosis requires compounds to be trapped in a membrane vesicle on the outer surface of the cell, transported through the cell membrane, and then released on the opposite side when the vesicle reopens.[13] Paracellular transport, on the other hand takes advantage of the space between cells in certain types of membranes.[14] Compounds move through the pores between cells of the GI tract, kidney cells, or other "leaky" membranes, thus avoiding the lipophilic environment of the cell membrane. Hydrophilic compounds incapable of diffusing into the lipid layer of a cellular membrane may be transported in this manner, provided they are small enough to fit in the space between the cells (typically 8 Å). Larger compounds (e.g., proteins) are too large to undergo paracellular transport.

Passive diffusion, active transport, endocytosis, and paracellular transport generally act to move a compound from one side of a biological barrier to the other, often moving down a concentration gradient, but not always (active transport). There are, however, systems in place designed to protect the body from xenobiotics. These efflux transporters generally move compounds against a concentration gradient, require the expenditure of energy (e.g., ATP), and their action decreases the ability of a compound to cross a biological barrier. In simple terms, compounds entering the cell membrane are captured by an efflux system protein, and are then ejected from the biological barrier on the same side as they entered. The net effect is an overall decrease in compound permeability through the cell membrane in question.

P-glycoprotein (Pgp) efflux proteins, a member of the ATP-binding cassette family of transporters, are a prototypical example.[15a,b] Unlike many other proteins, Pgps are capable of acting upon a broad range of substrates. They are heavily expressed in tissues such as the brain, liver, kidneys, intestines, and uterus. In principle, their action could decrease passage of compounds from the GI tract into the systemic circulation, but high concentrations generated by oral dosing often far exceed Pgp capacity in the gut. The impact of Pgp and other transport systems is more significant in areas of lower drug concentration, such as the blood–brain barrier. In this scenario, plasma drug concentration is unlikely to exceed Pgp expression and their substrates can