The “well-stirred” model of hepatic drug clearance was first proposed by Gillette (1971) and established by Rowland et al. (1973) and Wilkinson and Shand (1975). In the form that it is commonly used, net hepatic drug clearance based on whole-blood drug concentration (CLH) is derived as a function of hepatic blood flow (QH,B), the free fraction of drug in blood (fuH), and the intrinsic metabolic clearance in the liver based on unbound drug concentration (CLuint,H):

\[
CL_{H,B} = \frac{Q_{H,B} \cdot fu_H \cdot CL_{un,H}}{Q_{H,B} + fu_H \cdot CL_{un,H}}
\]

This assumes that the drug is distributed instantly and homogeneously throughout liver water and that the unbound concentrations in plasma and liver water are identical. Effectively, this means that drug distribution into the liver is perfusion-limited with no diffusion delay and that no active transport systems are involved. The latter possibilities have generally been disregarded in most applications.

A key feature of this model is that it equates whole-blood drug clearance, rather than plasma drug clearance, to liver blood flow, because the organ is potentially capable of extracting the drug from both plasma and blood cells. Most reported clearance values are referenced to plasma rather than blood, since it is more common to measure drug concentration in plasma. If plasma drug clearance (CLH) is to be estimated, then eq. 1 must be modified to take into account the fraction free in plasma (fu) and the total blood to total plasma drug concentration ratio (Cu/Cp):

\[
CL_H = \frac{Q_{H,B} \cdot fu_B \cdot CL_{un,H}}{Q_{H,B} + fu_B \cdot CL_{un,H}}
\]

As expected conceptually, eq. 1 indicates that hepatic blood clearance cannot exceed liver blood flow. However, from eq. 2, it is possible that hepatic plasma clearance may exceed hepatic blood flow (and hepatic plasma flow) when fuCLuint,H >> QH,B and Cu/Cp > 1 (Hinderling, 1997). Some confusion over the definition of fuH and a failure to appreciate the conversion between blood and plasma clearances are evident in some of the recent literature, confounding the ability to extrapolate between in vitro and in vivo drug clearance.

**The Definition of fuH**

The free fraction of drug in blood is given by eq. 3:

\[
fu_H = \frac{Au_H}{A_H}
\]

where AuH is the amount of unbound drug in whole blood and A_H is the total amount of drug in blood. Thus:

\[
CL_H = \frac{Q_{H,B} \cdot fu_H \cdot CL_{un,H}}{Q_{H,B} + fu_H \cdot CL_{un,H}}
\]

In the special case of the drug not associating with blood cells at all, it is preferable to report plasma, rather than blood, clearance, using fu and hepatic plasma flow (QH,H-B).

**Estimation of In Vivo Hepatic Clearance from In Vitro Data**

Values of CLun,H derived using human liver microsomes, hepatocytes, or recombinant enzymes are often scaled to in vivo hepatic clearance using the well-stirred model. However, in deriving hepatic plasma clearance, several recent studies have used eq. 8 instead of the correct form (eq. 2) (Blanchard et al., 2004; Foti and Fisher, 2004; Blanchard et al., 2005; de Graaf et al., 2006; Mohutsky et al., 2006; Strelevitz et al., 2006):

\[
CL_H = \frac{Q_{H,B} \cdot CL_{un,H}}{Q_{H,B} + CL_{un,H}}
\]
Estimation of Intrinsic Hepatic Clearance from in Vivo Data

To assess predictions of intrinsic hepatic drug clearance from in vitro data, estimates are often compared with values back-calculated from in vivo values of hepatic drug clearance using the well-stirred model. In this context, studies (e.g., Takanaga et al., 2000) have employed eq. 9 by rearranging eq. 8, instead of the correct form (eq. 11) for the estimation of hepatic clearance:

\[
CL_{in,H} = \frac{Q_{HB} \cdot CL_H}{fu \cdot (Q_{HB} - CL_H)}
\]  

\[
CL_{in,H} = \frac{Q_{HB} \cdot CL_H}{fu \cdot (Q_{HB} - CL_H(Cp/Cp))}
\]

Estimation of Hepatic Availability from in Vivo Data

Another common application of the well-stirred model is to estimate the fraction avoiding “first-pass” metabolism (hepatic availability; \(F_H\)) after oral drug administration from systemic clearance after intravenous administration. Assuming negligible extrahepatic clearance, several reports have used plasma drug clearance after intravenous administration (\(CL_{iv}\)) and eq. 11 (Lau et al., 2006; Strelevitz et al., 2006) or eq. 12 (Kharasch et al., 2004a,b) to estimate \(F_H\):

\[
F_H = 1 - \frac{CL_{iv}}{Q_{HB}}
\]

\[
F_H = 1 - \frac{CL_{iv}}{Q_{HB}(1 - Hc)}
\]

Eq. 11 is only valid if \(Cp/Cp\) is unity, and eq. 12 is not valid unless \(Cp/Cp\) is equal to \((1 - Hc)\) (i.e., no drug associates with the cells). A more appropriate equation, based on the original assumptions of the well-stirred model, is given by eq. 13:

\[
F_H = 1 - \frac{CL_{iv}}{Q_{HB}(Cp/Cp)}
\]

The different implications of eqs. 11 through 13 for the estimation of \(F_H\) are explored in Fig. 1.

Errors in the estimation of \(F_H\) may be further propagated into estimates of the fraction avoiding first-pass loss across the gut wall (\(F_G\)) from the relationship \(F_G(faF_H)\), where \(F_G\) is net oral bioavailability and \(fa\) is the fraction of the dose entering the intestinal wall (Takanaga et al., 2000; Kharasch et al., 2004a,b).

In summary, we emphasize that the mixing of plasma free fraction and hepatic blood flow is inadvisable and illogical when using the well stirred liver, or indeed any model of hepatic drug clearance, for in vitro or in vivo extrapolation.

References


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