itself into the systemic circulation. In the case of a tissue-targeted prodrug where the prodrug must be delivered into the systemic circulation, the promoiety must not undergo appreciable degradation during the absorption process. If the prodrug is intended to target a specific tissue or organ, it should have sufficient stability in the blood to reach that target organ or tissue. The enzymatic process required to process the prodrug and release the active drug must be efficient such that adequate concentrations of the drug are available to reach therapeutic levels.

To help in assessing the utility of a prodrug strategy and optimize the desired properties, *in vitro* and *in vivo* systems are available. *In vitro* assays evaluating stability in simulated gastric and intestinal fluids, stability in blood or plasma, stability on exposure to intestinal and liver S9 fractions, Caco-2 cell permeability and isolated enzyme stability have been used extensively in prodrug development programs. In using *in vitro* cell-based systems to evaluate intrinsic potency or any potential toxicological liabilities of a prodrug that requires an enzymatic process to release the active drug, it is important to determine that the cell line being used expresses the complement of enzymes required to cleave the prodrug of interest. Otherwise, development and evaluation of a prodrug strategy can be misleading. Ultimately, for the successful development of any prodrug, *in vivo* evaluation needs to be an integral part of the prodrug assessment process at the earliest stages of development. The real test of a prodrug strategy's utility is evaluation in animals where the fate of the prodrug and parent drug can be followed to determine if the prodrug strategy being employed is delivering the desired results when exposed to the complexity of an *in vivo* environment.

The development of antiviral therapies has seen the implementation of a wide variety of prodrug approaches to address a range of drug development problems.[4–7] The field has seen successful implementation of prodrug approaches to address membrane permeability issues, dissolution and solubility-limiting absorption, metabolic deficiencies and tissue targeting. A number of examples that highlight prodrug solutions to problems in antiviral drug discovery and development are presented here.

12.2 Prodrugs of Alcohols and Carboxylic Acids

Polar functionality such as alcohols, amines and carboxylic acids reduce the overall lipophilicity of a molecule and, therefore, can impact diffusion across biological membranes. To mask the polar nature of these functionalities, simple ester, carbonate and carbamate prodrugs are frequently employed (Figure 12.1A).[8] These prodrug constructs are usually cleaved by ubiquitous esterases in the liver, blood or other organs to release the active agent into the systemic circulation or site of action. In developing an ester-type prodrug of a poorly bioavailable molecule, a balance between lipophilicity, solubility and enzymatic cleavage kinetics is required. It should be noted that because of differences in esterase activities in preclinical species, human PK predictions can be difficult.