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8

PRODRUG APPROACHES TO DRUG DELIVERY

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8.1. INTRODUCTION

Most drugs, in order to produce their desired pharmacological action, have to overcome many hurdles before reaching the desired site of action. These hurdles include the intestinal barrier, the blood-brain barrier (BBB), and metabolic reactions that could render them inactive. These three subjects are covered in Chapters 2, 3, and 6, respectively, and therefore will not be discussed in detail here. Most drugs distribute randomly throughout the body, and the amount of drugs reaching the site of action is relatively small. For an effective amount to reach the site of action and not cause severe systemic side effects, a drug must possess certain physicochemical properties that make it conducive to penetration through various biological membranes (i.e., sufficiently bioavailable), to avoid metabolic inactivation by various enzymes, and to avoid retention in body depot tissues that could lead to undesirable long-lasting effects. These desired physicochemical properties are not always present in pharmacologically active compounds.

With the advance of new technologies such as combinatorial and computational chemistry, more and more compounds are being identified with extremely potent *in vitro* activity but are found to be inactive *in vivo*. They may have the optimal configuration and conformation needed to interact with their target receptor or enzyme, but they do not necessarily possess the best molecular form and physicochemical properties needed for their delivery to the site of action. Some of the problems often encountered include (1) limited solubility and poor chemical stability preventing the drug from being adequately formulated, (2) low or variable bioavailability due to incomplete absorption across biological membranes or extensive first-pass metabolism, and (3) lack of site specificity. Further structural modifications are often performed but do not always solve all the problems. Another approach that

is often effective in solving some of these delivery problems is the design of prodrugs by attaching a promoiety to the active drug.[1–3] This chapter will focus on the various prodrug approaches that have been used to overcome many of the pharmaceutical and pharmacokinetic barriers that hinder optimal delivery of the active drug.

8.2. BASIC CONCEPTS: DEFINITION AND APPLICATIONS

A prodrug by definition is inactive or much less active and has to be converted to the active drug within the biological system. There are a variety of mechanisms by which a prodrug can be activated. These include metabolic activation mediated by enzymes present in the biological system as well as the less common, simple chemical means of activation such as hydrolysis.

Prodrugs occur in nature. One example is proinsulin, which is synthesized in the pancreas and releases its active moiety, insulin, and an inactive peptide. Codeine is another example; it can be regarded as a prodrug of morphine, which is responsible for its analgesic effect.

Most synthetic prodrugs are prepared by attachment of the active drug through a metabolically labile linkage to another molecule, the "promoiety". The promoiety is not necessary for activity but may impart some desirable properties to the drug, such as increased lipid or water solubility or site specificity. Advantages that can be gained with such a prodrug include increased bioavailability, alleviation of pain at the site of injection, elimination of an unpleasant taste, decreased toxicity, decreased metabolic inactivation, increased chemical stability, and prolonged or shortened duration of action.

8.2.1. Increasing Lipophilicity to Increase Systemic Bioavailability

This is the most successful application of prodrugs. Because of the lipid bilayer nature of biological membranes, the rate of passive drug transport is affected by both lipophilicity and aqueous solubility (also called "hydrophilicity"). The rate of passive diffusion across the biological membrane will increase exponentially with increasing lipophilicity and then level off at higher lipophilicity. This is due to the fact that an increase in lipophilicity is usually accompanied by a decrease in water solubility and will eventually decrease the flux over the membrane due to poor water solubility. The design of prodrugs aims to achieve a balance between lipophilicity and aqueous solubility in order to improve passive drug transport across various biological membranes. Using data drawn from U.S. Adopted Names, the World Drug Lists, and Pfizer internal compound collections, it was concluded that, to have good membrane permeability, drugs should have a relatively low molecular weight ($\leq$500), be relatively nonpolar, and partition between an aqueous and a lipid phase in favor of the lipid phase but, at the same time, possess certain water solubility ($-1 \leq \mathrm{Log}P \leq +5$).[4] The majority of effective oral drugs obey this so-called Lipinski's rule of 5.

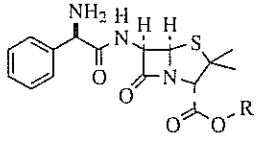
Since most drugs are either weak acids or weak bases, they are often given and present in the salt form under relevant physiological conditions. Therefore, dissociation constants also affect membrane permeability, and thus bioavailability. It is generally accepted that the neutral, unionized and thus most lipophilic form of an acidic or basic drug is absorbed far more efficiently than the ionized species. In these cases, the distribution between the ionized and neutral form depends strongly on pH. The effective partition coefficient for a dissociative system (Log*D*) gives the correct description of such complex partitioning equilibria.

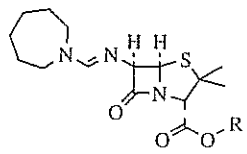
$$\text{Log}D = \text{Log}P_{HA} - \log(1 + 10^{(\text{pH}-\text{p}Ka)}) \quad \text{for an acid}$$

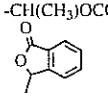
$$\text{Log}D = \text{Log}P_{B} - \log(1 + 10^{(\text{p}Ka-\text{pH})}) \quad \text{for a base}$$

where P_{HA} and P_B are the intrinsic partition coefficients of the weak acid and weak base, respectively. Programs such as ACD/Log*P* and cLog*P* are available to calculate with reasonable accuracy the Log*P* and Log*D* values using a structure-fragment approach as well as internal structure databases. To illustrate the principles discussed in this chapter, examples will be given with their Log*P* and/or Log*D* values calculated using the Advanced Chemistry Development (ACD) Software.

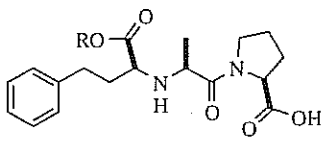
Many prodrugs feature the addition of a hydrophobic group in order to increase their lipid solubility to improve their gastrointestinal absorption. Bacampicillin (**2**), pivampicillin (**3**), and talampicillin (**4**) are more lipophilic esters of ampicillin (**1**), and pivmecillinam (**6**) is a more lipophilic ester prodrug of mecillinam (**5**), all with improved oral bioavailability. For example, absolute oral bioavailability in horses was 39%, 31%, and 23% for bacampicillin, pivampicillin, and talampicillin, respectively, compared to only 2% for ampicillin sodium.[5] Esterification of carboxylic acid in ampicillin (**1**) resulted in an increase of 0.8-1.4 unit in Log*P*. More significant are the increases in Log*D* values for the prodrugs when ionization of the amino group is taken into consideration; as much as a 4-unit difference in Log*D* is estimated at pH values in the intestines where the prodrugs are believed to be absorbed. Other prodrugs of antibiotics include esters of carbenicillin (for urinary tract infection), cefotiam, and erythromycin.





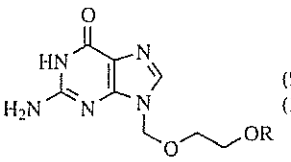
			ACD/Log*P*	ACD/Log*D* pH 7	ACD/Log*D* pH 1
(1) Ampicillin	R = H		1.350±0.320	−1.54	−1.72
(2) Bacampicillin	R = -CH(CH$_3$)OCOOC$_2$H$_5$		2.172±0.894	1.99	−0.93
(3) Pivampicillin	R = -CH(CH$_3$)OCOC(CH$_3$)$_3$		2.552±0.884	2.37	−0.55
(4) Talampicillin	R =		2.789±0.882	2.61	−0.31
(5) Mecillinam	R = H		1.493±0.866	−1.01	−1.58
(6) Pivmecillinam	R = -CH(CH$_3$)OCOC(CH$_3$)$_3$		3.453±0.921	1.77	0.35

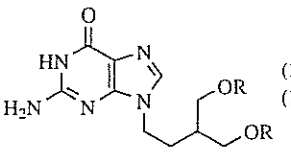
Enalapril (**8**) is an ester prodrug of enalaprilat (**7**). The latter binds tightly to the angiotensin-converting enzyme (ACE) but is transported with low efficacy by the peptide carrier in the gastrointestinal tract. The prodrug enalapril has a higher affinity for the peptide carrier[6] and is much better absorbed, with about 60% oral bioavailability.[7,8] As a matter of fact, all ACE inhibitors except captopril and lisinopril are administered as prodrugs; other commercialized ACE inhibitor prodrugs include perindopril, quinapril, ramipril, cilazapril, benazepril, spirapril, and trandolapril, all based on esterification of the same carboxylic acid group.[9] The esters are hydrolyzed *in vivo*, after absorption, to the corresponding active but poorly absorbed dicarboxylate forms.



		ACD/Log*P*	ACD/Log*D* pH 7	ACD/Log*D* pH 1
(7) Enalaprilat	R = H	2.102±0.574	−1.45	−0.92
(8) Enalapril	R = Et	2.983±0.580	−0.12	−0.10

Valacyclovir (**10**)[10,11] and famciclovir (**12**) are ester prodrugs of acyclovir (**9**) and penciclovir (**11**), respectively, for the treatment of viral infections. Both acyclovir and penciclovir exhibit site-specific conversion to the active triphosphate species by viral thymidine kinase. They show remarkable antiviral selectivity and specificity. However, their oral bioavailability is quite low, 15–20% of an oral dose being absorbed in humans for acyclovir and 5% for penciclovir.[12] Both valacyclovir and famciclovir have no intrinsic antiviral activity, and both are rapidly hydrolyzed to acyclovir and penciclovir by esterases present in the liver and gut wall. Valacyclovir displays a mean absolute bioavailability of 54%, a threefold increase in oral bioavailability over acyclovir, while famciclovir has an absolute bioavailability of 77% in humans.[13–15] Famciclovir's better bioavailability could be explained by the increase in lipophilicity; the high oral bioavailability of valacyclovir was also partly attributed to the involvement of an active transport mechanism through PEPT1.[16] Therefore, in addition to increasing lipophilicity, prodrug design can utilize active transport mechanisms as a means of enhancing bioavailability.

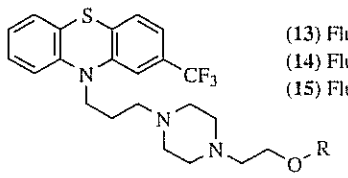




			ACD/Log*P*	ACD/Log*D* pH 7	ACD/Log*D* pH 1
(**9**) Acyclovir	R = H		−1.760±0.489	−1.76	−3.76
(**10**) Valacyclovir	R = L-Valyl-		0.040±0.577	−0.78	−4.76
(**11**) Penciclovir	R = H		−2.031±0.584	−2.03	−4.44
(**12**) Famciclovir	R = CH$_3$CO-		−0.088±0.265	−0.09	−3.27

3.2.2. Sustained-Release Prodrug Systems

Antipsychotic drugs are the mainstay treatment for schizophrenia and similar psychotic disorders. Long-acting depot injections of antipsychotic drugs are extensively used as a means of long-term maintenance treatment. The duration of action for many antipsychotic drugs with a free hydroxyl group can be considerably prolonged by the preparation of long-chain fatty acid esters with very high Log*P* values (usually 7 or above). Fluphenazine enanthate (**14**) and fluphenazine decanoate (**15**) were the first of these esters to appear in clinical use and are longer-acting, with fewer side effects than the parent drug. The ability to treat patients with a single intramuscular injection every 1–2 weeks with the enanthate or every 2–3 weeks with the decanoate esters means that problems associated with patient compliance with the drug regimens and with drug malabsorption can be reduced.[17] Esterification of antipsychotic drugs with decanoic acid yields very lipophilic prodrugs which are dissolved in a light vegetable oil such as Viscoleo or sesame oil. Intramuscular injection creates an oily depot from which the prodrug molecules slowly diffuse into the systemic circulation, where they are hydrolyzed quickly by esterases to the active moieties. These depot forms allow these drugs to be given only once or twice a month, permitting the long-term treatment of schizophrenia. Antipsychotic drugs available in depot formulation include fluphenazine (**13**), flupenthixol, haloperidol, and zuclopenthixol in their enanthate or decanoate esters.



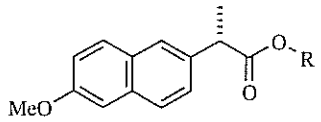
		ACD/Log*P*
(**13**) Fluphenazine	R= H	4.841±0.460
(**14**) Fluphenazine enanthate	R = -CO(CH_2)$_5$$CH_3$	8.034±0.453
(**15**) Fluphenazine decanoate	R = -CO(CH_2)$_8$$CH_3$	9.628±0.453

Anabolic steroids such as nandrolone and testosterone, anti-inflammatory glucocorticoids such as methylprednisolone, and contraceptives such as estradiol and levonorgestrel all have slow-release formulations of their ester prodrugs in the market.

3.2.3. Improving Gastrointestinal Tolerance

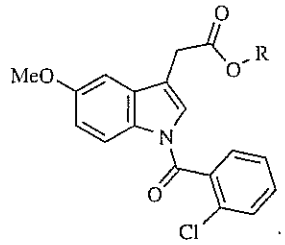
Temporary masking of carboxylic acid groups in nonsteroidal anti-inflammatory drugs was proposed as a promising means of reducing gastrointestinal toxicity resulting from direct mucosal contact mechanisms. Morpholinoalkyl esters (**17** and **19**, HCl salts) of naproxen (**16**) and indomethacin (**18**) were evaluated *in vitro* and *in vivo* for their potential use as prodrugs for oral delivery.[18] The prodrugs were freely soluble in simulated gastric fluid and pH 7.4 phosphate buffer and showed a minimum of a 2000-fold increase in solubility over the parent drugs. The prodrugs were more lipophilic than the parent drugs and were quantitatively hydrolyzed to

their respective parent drugs *in vivo*. The prodrugs were 30–36% more bioavailable orally than the parent drugs following a single dose in rats. They were significantly less irritating to gastric mucosa than the parent drugs following a single dose as well as chronic oral administration in rats.



(**16**) Naproxen (R = H)

(**17**) R = $-(CH_2)_n-N$ O

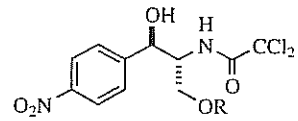


(**18**) Indomethacin (R = H)

(**19**) R = $-(CH_2)_n-N$ O

8.2.4. Improving Taste

Oral drugs with a markedly bitter taste may lead to poor patient compliance if administered as a solution or syrup. The prodrug approach has been used to improve the taste of chloramphenicol (**20**), clindamycin, erythromycin, and metronidazole.[19] A prodrug such as chloramphenicol palmitate (**21**), with Log*P* of around 10, does not dissolve in an appreciable amount in the mouth and, therefore, does not interact with the taste receptors.

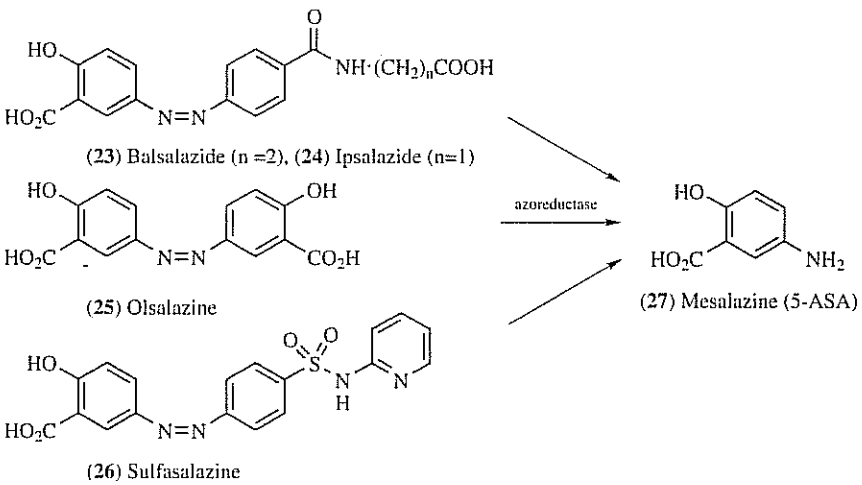


		ACD/Log*P*	ACD/Log*D* pH 7	ACD/Log*D* pH 1
(**20**) Chloramphenicol	R = H	1.018±0.321	1.02	1.02
(**21**) Chloramphenicol palmitate	R = -CO(CH_2)$_{14}$$CH_3$	9.920±0.756	9.92	9.92
(**22**) Chloramphenicol sodium succinate	R = -$COCH_2CH_2COO^-Na^+$	2.287±0.849	–0.34	2.29

8.2.5. Diminishing Gastrointestinal Absorption

Many prodrugs have been evaluated in this context for colon-specific drug delivery. Colon targeting is of value for the topical treatment of diseases of the colon such as Crohn's disease, ulcerative colitis, and colorectal cancer. Sustained colonic release of drugs can be useful in the treatment of nocturnal asthma, angina, and arthritis. Prodrugs have been designed to pass intact and unabsorbed from the upper gastrointestinal tract and undergo biotransformation in the colon, releasing the active drug

molecule. Prodrug activation can be carried out by microflora and distinct enzymes present in the colon (such as azoreductase, glucuronidase, glycosidase, dextranase, esterase, nitroreductase, and cyclodextranase).[20,21] Balsalazide (**23**), ipsalazide (**24**), olsalazine (**25**), and sulfasalazine (**26**) are azo-containing prodrugs developed for colon-specific delivery of an anti-inflammatory agent in the treatment of inflammatory bowel disease. As shown in Scheme 1, they can undergo azoreduction in the colon to release the active 5-aminosalicylic acid (5-ASA or mesalazine, **27**). Other prodrugs evaluated for colon-specific delivery include conjugates of amino acids, glucuronide, glycoside, dextran, and cyclodextrin.[22]



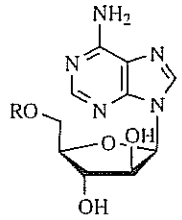
Scheme 1

8.2.6. Increasing Water Solubility

Poorly water-soluble, lipophilic drugs also have difficulty getting absorbed, as discussed earlier. The prodrug approach has been applied to circumvent solubility problems by introduction of an ionizable functional group such as phosphate esters, amino acid esters, and hemiesters of dicarboxylic acids, allowing various salts of such prodrugs to be formed. Prodrugs can also be used to increase water solubility in order to increase the amount of drug that will reach the systemic circulation through parenteral administration. Examples include chloramphenicol sodium succinate (**22**), hydrocortisone sodium succinate, methylprednisolone sodium succinate, betamethasone sodium phosphate, clindamycin phosphate, and prednisolone phosphate.

In addition to the use of ionizable groups, disruption of the crystal lattice can also result in a significant increase in aqueous solubility, as illustrated by the antiviral agent vidarabine (**28**). The 5′-formate ester derivative (**29**) of vidarabine is 67-fold more soluble in water than vidarabine itself and has been attributed to

disruption of the strong intermolecular interactions in the crystal, as indicated by the 85°C drop in the melting point.[23]



		ACD/Log*P*	m.p.	solubility$_{H_2O}$ (25 °C)
(**28**) Vidarabine	R = H	−1.458±0.470	260 °C	0.0018 M
(**29**) 5′-formate	R = C-H (=O)	−0.364±0.601	175 °C	0.12 M

8.2.7. Tissue Targeting and Activation at the Site of Action

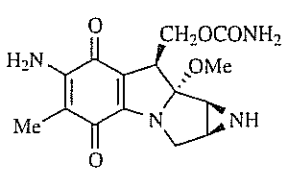
Prodrugs can be designed to target specific tissues. This is especially useful in improving the therapeutic effectiveness and decreasing the systemic toxicity of anticancer agents in the treatment of cancer. Anticancer agents are usually highly toxic, with a very small therapeutic index, and their therapeutic effectiveness is often limited by their dose-limiting side effects. Here, several strategies for targeting chemotherapeutic agents to cancers will be briefly discussed to illustrate the applications of prodrugs. For details, refer to Chapter 11 on metabolic activation and drug targeting.

8.2.7.1. Tumor Hypoxia and Bioreductive Activation of Anticancer Prodrugs

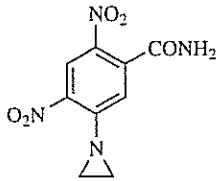
Solid tumors often contain regions which are subject to chronic or transient deficiencies of blood flow and, therefore, to the development of chronic or acute hypoxia owing to the primitive state of tumor vasculature.[24] Hypoxic cells in a solid tumor frequently constitute 10–20% and occasionally over half of the total viable tumor cell population. Agents that are active against proliferating cells are relatively ineffective against these hypoxic tumor cells, which are not actively replicating at the time of treatment but are capable of commencing proliferation at a later time and causing the tumor to regrow. Hypoxic cells also may be resistant to conventional chemotherapy due to pharmacodynamic considerations.[24] To produce a therapeutic response, appropriate drug concentrations must be reached. Drugs that have physicochemical properties not conducive to diffusion into tumor tissue, or that are unstable or metabolized rapidly, may not reach chronically hypoxic tumor cells located in regions of severe vascular insufficiency. Therefore, the presence of hypoxic cells in solid tumors is an obstacle to effecting a cure.

Since hypoxic cells located remotely from the vascular supply of a tumor mass may have a greater capacity for reductive reactions than their normal, well-oxygenated counterparts, hypoxia could provide an opportunity for the design of selective cancer chemotherapeutic agents that could be reductively activated in these hypoxic cells.[24] Several classes of agents are presently known which exhibit preferential cytotoxicity toward hypoxic cells through reductive activation. They

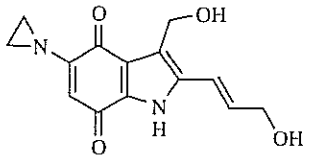
ıclude nitro compounds, quinones, and aromatic *N*-oxides.[25,26] Examples include ıitomycin C (**30**), CB1954 (**31**), EO9 (**32**), and AQ4N (**33**).



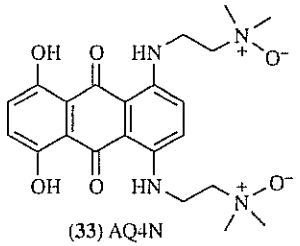
(**30**) Mitomycin C



(**31**) CB1954



(**32**) EO9

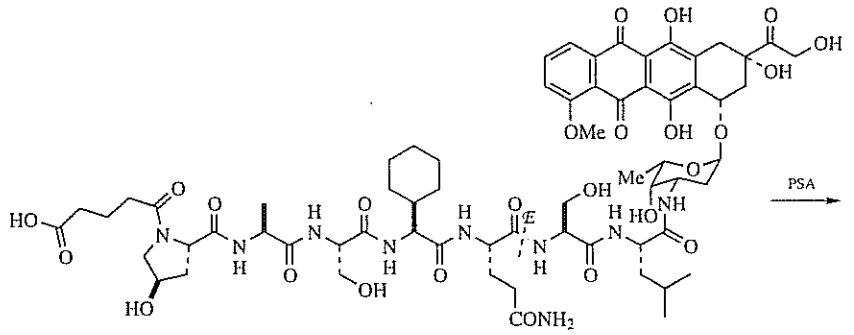


(**33**) AQ4N

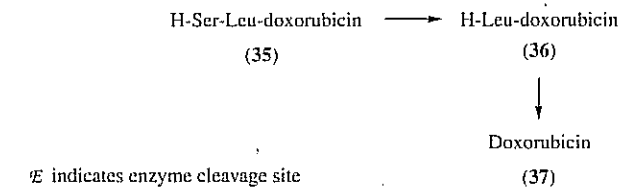
.2.7.2. Activation of Prodrugs by Tissue- or Tumor-Specific Enzymes Investiations of the biochemistry and molecular biology of cancer have also identified ɔveral reductive or proteolytic enzymes that are unique to tumors or tissues and ɔuld be used as potential therapeutic targets or prodrug-converting enzymes for ovel cancer therapy. These include DT-diaphorase,[27] prostate specific antigen ?SA),[28] plasminogen activator,[29] and members of matrix metalloproteinases.[30]

One such example is the peptide doxorubicin conjugate, glutaryl-Hyp-Ala-Ser-hg-Gln-Ser-Leu-Dox, L-377202 (**34**), which was reported to have the profile of hysical and biological properties needed for further clinical development.[31] Conıgate **34** was found to have a greater than 20-fold selectivity against PSA-secreting NCaP cells relative to non-PSA-secreting DuPRO cells. In nude mouse xenograft ıudies, it reduced PSA levels by 95% and tumor weight by 87% at 21 μmol/kg, a ose below its maximal tolerated dose (MTD). On the basis of these results, this ɔnjugate was selected for further studies in clinical trials to assess its ability to ıhibit human prostate cancer cell growth and tumorigenesis. It was believed that SA cleavage in and around prostate cancer cells would release, as shown in cheme 2, dipeptide-doxorubicin conjugate (**35**), which would be further cleaved y aminopeptidases to the cytotoxic Leu-doxorubicin (**36**) and doxorubicin (**37**).

.2.7.3. Antibody- or Gene-Directed Enzyme Prodrug Therapy Besides targeting ypoxic tumor cells and using tumor- or tissue-specific enzymes like PSA to actiıte prodrugs, other specific enzymes can be delivered to tumor tissues using ıtibodies or expressed by tumor cells through gene therapy and can be used as rodrug converting enzymes. These strategies are called "antibody-directed

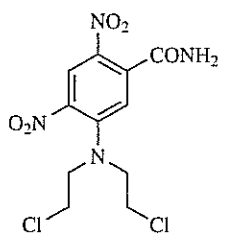


(**34**) L-377202

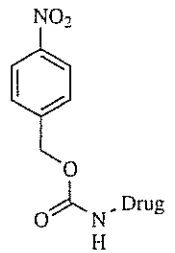


Scheme 2

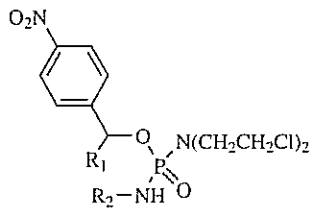
enzyme prodrug therapy" (ADEPT) or "gene-directed enzyme prodrug therapy" (GDEPT). In these approaches, an enzyme is delivered site specifically by chemical conjugation or genetic fusion to a tumor-specific antibody or by enzyme gene delivery systems into tumor cells. This is followed by the administration of a prodrug, which is selectively activated by the delivered enzyme at the tumor cells. A number of these systems are in development and have been reviewed.[32] Among the enzymes under evaluation is a bacterial nitroreductase from *Escherichia coli*. This is a flavin mononucleotide (FMN)–containing flavoprotein capable of reducing certain aromatic nitro groups to the corresponding amines or hydroxylamines in the presence of a cofactor NADH or NADPH. The nitroaromatics that were found to be good substrates of *E. coli* nitroreductase include dinitroaziridinylbenzamide CB1954 (**31**), dinitrobenzamide mustards SN 23862 (**38**), 4-nitrobenzylcarbamates (**39**), and nitrophenyl phosphoramides (**40** and **41**).[33]



(**38**) SN 23862



(**39**) 4-nitrobenzylcarbamates



nitrophenyl phosphoramides
(**40**) $R_1 = R_2 = H$ (LH7)
(**41**) $R_1, R_2 = CH_2CH_2$

8.2.7.4. Tumor-Specific Transporters Antibody-drug conjugates would have to overcome problems inherent in proteins such as susceptibility to proteolytic cleavage and high immunogenicity; the latter could lead to an antibody response against the conjugate, thereby precluding further use. To increase the selectivity of chemotherapeutic agents, considerable efforts have also been made to identify biochemical characteristics unique to malignant tumor cells that could be exploited in a therapeutic intervention. The small and nonimmunogenic, tumor-specific molecules like folic acid are among the promising alternatives to antibody molecules as targeting agents for drug delivery. Folate conjugates of radiopharmaceuticals, magnetic resonance imaging (MRI) contrast agents, antisense oligonucleotides and ribozymes, proteins and protein toxins, immunotherapeutic agents, liposomes with entrapped drugs, and plasmids have all been successfully delivered to folate receptor–expressing cells.[34] More details can be found in Chapter 9 on receptor-mediated endocytosis.

8.3. PRODRUG DESIGN CONSIDERATIONS

Often medicinal chemists encounter a situation where a structure has adequate pharmacological activity but an inadequate pharmacokinetic profile (i.e., absorption, distribution, metabolism, and excretion). Prodrugs can be designed to improve physicochemical properties, resulting in improvement in pharmacokinetic as well as pharmaceutical properties. The pharmaceutical properties that could be improved, as discussed earlier, include drug product stability, taste and odor, pain on injection, and gastrointestinal irritation. These are great benefits that can be achieved through the design of prodrugs. However, regulatory issues should also be considered in the design process. In general, regulatory agencies are reluctant to register this type of product. Of particular concern is the fact that toxicological studies might not be relevant for human use of the drug because of differences in the rate and/or extent of formation of the active moiety—metabolic aspects. Experiments should thus be designed early to address these concerns. As examples of interspecies differences, the pivaloyloxyethyl ester of methyldopa was essentially hydrolyzed presystematically to pivalic acid and methyldopa at the same rate in human, dog, and rat, while the succinimidoethyl derivative was hydrolyzed faster in rat than in man and dog.[35] This suggests that the succinimidoethyl ester of methyldopa was more resistant to extrahepatic esterase action in man and dog but not in rat. For different ester prodrugs of dyphylline, the relative rates of release were 1.3 to 13 times faster in rabbit plasma than in human plasma.[36]

The bond between the active moiety (parent drug) and the promoiety plays a major role in determining the pharmacokinetic properties of a prodrug. Knowledge about the nature of the bond and the promoiety may help explain the nature of the biotransformation process and its location in specific tissues or cells. The study of the fate in the body of the promoiety is particularly important from the safety point of view and should be investigated just as thoroughly as that of the active moiety. In some cases, the fate of the released carrier moiety is well known, such as the esters of methanol or ethanol; no extra study is needed during drug development. In other

Rational design of a prodrug should begin with identification of the problem(s) encountered with the delivery of the parent compound/drug and the physicochemical properties needed to overcome the delivery problem(s). Only then can the appropriate promoiety be selected to construct a prodrug with the proper physicochemical properties that can be effectively transformed to the active drug in the desired biological compartment.

The most important requirement in prodrug design is naturally the adequate reconversion of the prodrug to the active drug *in vivo* at the intended compartment. This prodrug-drug conversion may take place before absorption (e.g., in the gastrointestinal system), during absorption (e.g., in the gastrointestinal wall or in the skin), after absorption, or at the specific site of drug action. It is important that the conversion be essentially complete because the intact prodrug, being usually inactive, represents unavailable drug. However, the rate of conversion would depend on the specific goal of the prodrug design. A prodrug designed to overcome poor solubility for an intravenous drug formulation should be converted very quickly to the active moiety after injection. If the objective of the prodrug is to produce sustained drug action through rate-limiting conversion, the rate of conversion should not be too fast.

Prodrugs can be designed to use a variety of chemical and enzymatic reactions to achieve cleavage to generate their active drug at the desired rate and place. The design is often limited by the availability of a suitable functional group in the active drug for the attachment of a promoiety. Table 8.1 lists some of the common reversible prodrug forms for various functional groups that are often present in biologically active substances.

The most common prodrugs are those that require hydrolytic cleavage, but reductive and oxidative reactions have also been used for the *in vivo* regeneration of the active drug. Besides using the various enzyme systems for the necessary activation of prodrugs, the buffered and relatively constant physiological pH may be used to trigger their release.

Enzymes considered important to orally administered prodrugs are found in gastrointestinal walls, liver, and blood. In addition, enzyme systems present in the gut microflora may be important in metabolizing prodrugs before they reach the intestinal cells. In addition, site-specific delivery can be accomplished by exploiting enzymes that are present specifically or at high concentrations in the targeted tissues relative to nontarget tissues. A number of enzymes can also be delivered to targeted tissues through antibodies or gene-delivery approaches for the activation of subsequently administered prodrugs, as discussed earlier in this chapter.

8.4. PRODRUGS OF VARIOUS FUNCTIONAL GROUPS

8.4.1. Ester Prodrugs of Compounds Containing —COOH or —OH

Due to the presence of a wide variety of esterases in various body tissues, it is not surprising that esters are the most common prodrugs used to improve gastrointestinal absorption. By appropriate esterification of molecules containing a carboxylic