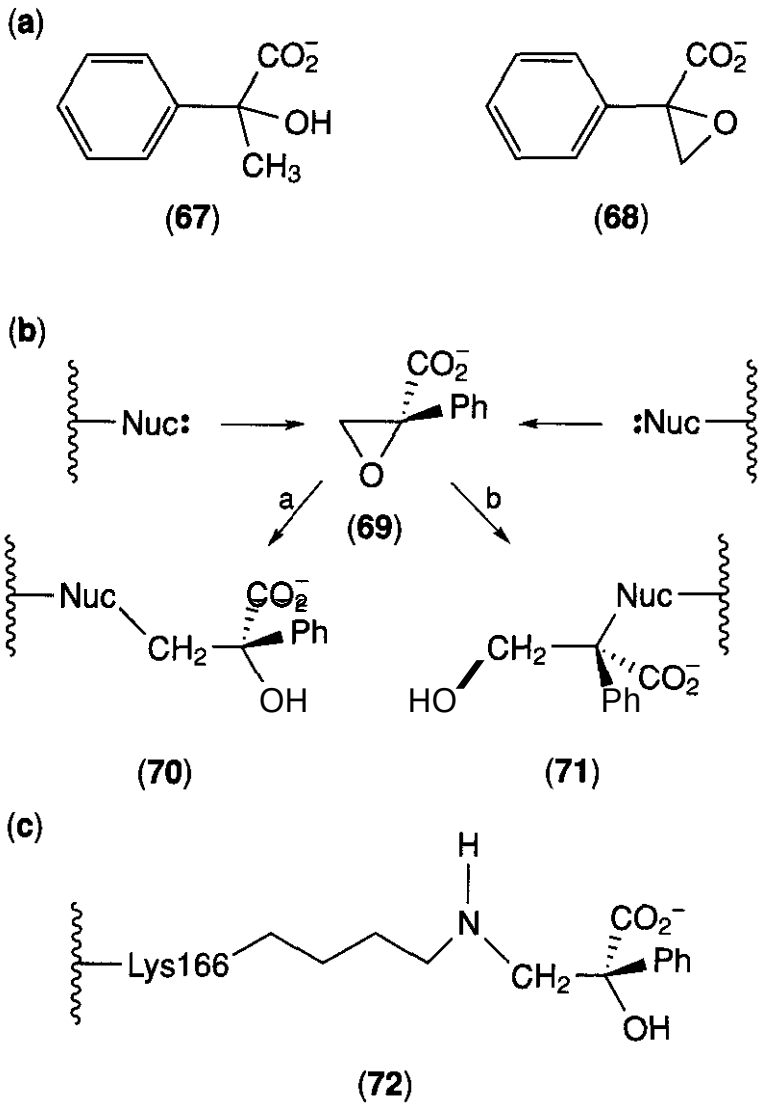
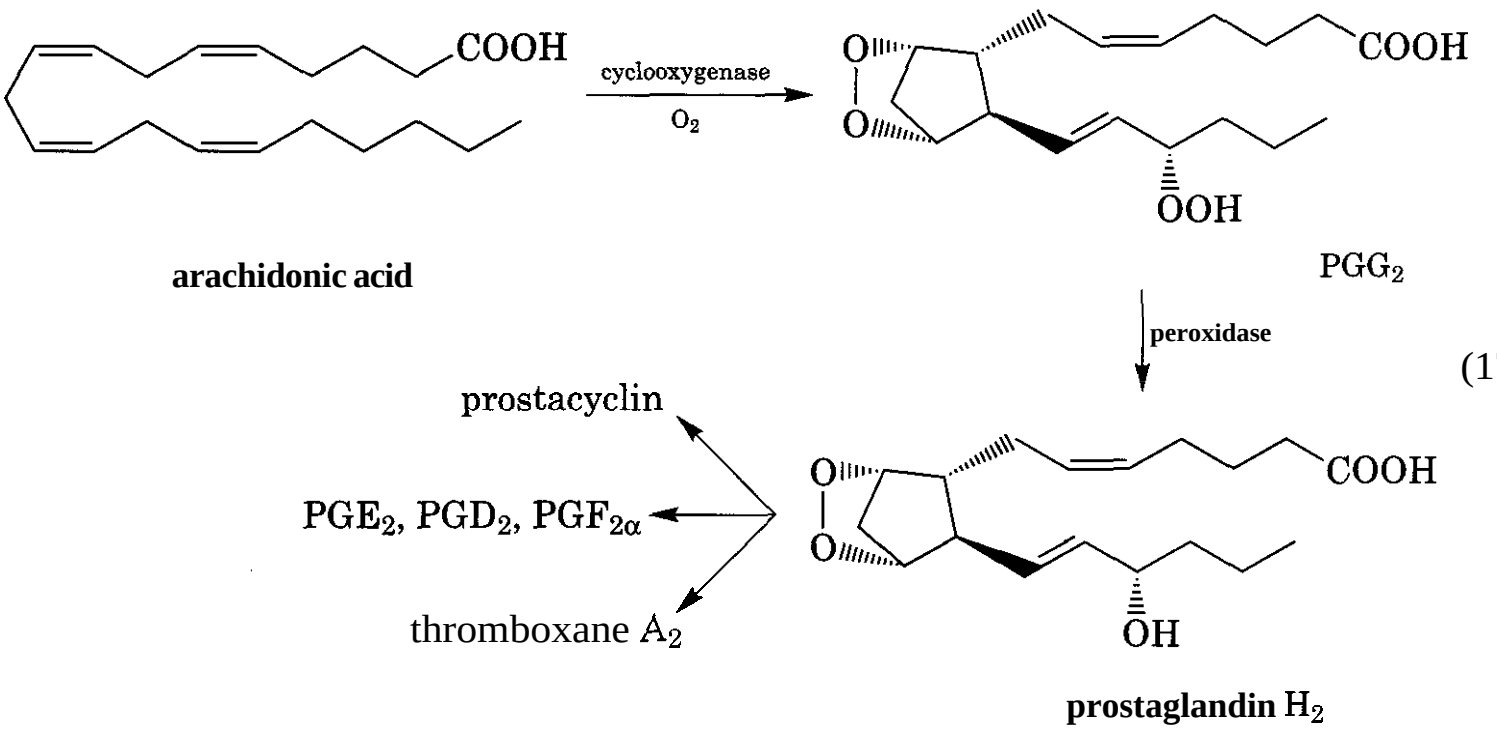


Figure 17.31. (a) Inhibitors of mandelate racemase, (b) potential products from nucleophilic attack on the epoxide of *R*-α-phenylglycidate, and (c) adduct formed by attack of Lys166.

corporated per mole of enzyme (185,186). Enzymatic digest of the labeled enzyme provided evidence that a serine residue, later identified as Ser530, was acetylated (186, 187), probably by the mechanism shown in Fig. 17.32. The X-ray structure of bromoaspirin (74) inactivated prostaglandin synthase has been solved, and the bromoacetylation of Ser530 confirmed (188). The structure was similar to that of flurbiprofen (**75**), complexed with prostaglandin synthase (189). Aspirin is the only NSAID known to inactivate prostaglandin synthase through covalent modification, and the brominated aspirin analog was also determined to be a potent irreversible inhibitor. Conversely, flurbiprofen (**75**), another NSAID, has been classified as a slow-tight-binding inhibitor (190) and was expected to induce a conformational change upon binding. However, there were no significant differences between the two X-ray structures, and it is yet to be determined whether the binding of aspirin also induces a conformational change in the enzyme.

Although affinity labels have played a major role in characterizing the active sites of a large number of proteases, they have also proved to be particularly useful in mapping nucleotide-binding sites (161, 163, 191). Numerous compounds, some of which are shown in Fig. 17.33, have been designed to be analogs of the various nucleosides and nucleotides. Perhaps the best known of these is 5'- *p*-fluorosulfonylbenzoyl adenosine (5'-FSBA) (**76**), which was designed to be an analog of ADP or ATP (77). It has both the adenosine and ribose moieties, as well as a carbonyl group adjacent to the 5' position. The latter mimics the first phosphoryl group of the purine nucleotides. If



(17.50)