The most potent inhibitors of DHFR are folic acid analogs that differ from the natural ligand in that they bear a 2,4-diaminopyrimidine unit. The inhibitors in which the side chain ends in a glutamic acid residue, as in folic acid, are known as classical antifolates. Other inhibitors with lipophilic substituents are known as nonclassical antifolates.

5.1 CLASSICAL DHFR INHIBITORS

The two main classical DHFR inhibitors are aminopterin (AM) and methotrexate (MTX, amethopterin), which were designed by replacing an enol-type OH group at C-4 of the natural substrate (DHF) by an amino group. Another DHFR inhibitor bearing a glutamate side chain is pemetrexed, previously mentioned as a TS inhibitor (Figure 2.36).

In the design of methotrexate and aminopterin, the implicit assumption was made that the two ligands would bind similarly and that the 4-amino group of MTX would go to the position in the binding site normally occupied by the DHF carbonyl. However, X-ray diffraction structures of DHFR with dihydrofolate and methotrexate showed different binding modes, with the aminopteridine ring of methotrexate flipped 180 degrees about the C_2–NH_2 bond compared to that of dihydrofolate (Figure 2.37). Both ligands bind to the DHFR active site by hydrogen bonds and by additional interactions with

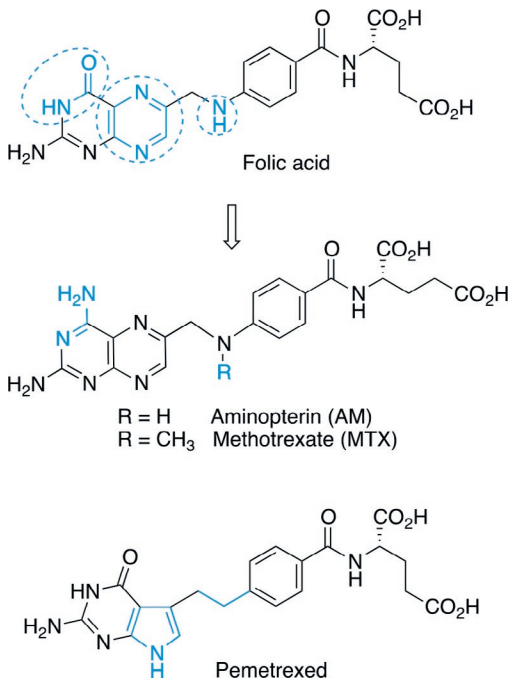

FIGURE 2.36

The design of DHFR inhibitors from folic acid.