Ramaprilat was also shown to be a slow-tight-binding inhibitor of ACE, operating by mechanism B, with K_i^* (Equation 17.30) of 7 pM (122). A more detailed discussion of the development of the ACE inhibitors is available (121).

2.5.3 Transition-State Analogs. As a chemical reaction proceeds from substrates to products, it will pass through one or more transition states. The energy barrier imposed by the highest energy transition state controls the overall rate of the reaction. Enzymes bring about rate enhancements of 10^{10}–10^{15} (123) by lowering this energy barrier. They do this by having a greater affinity to the structure of the transition state than to the structures of either substrates or products. Although an enzyme may have good *affinity* for its substrate, as evidenced by a low dissociation constant (K_S, Equation 17.411, for the Michaelis ($E \cdot S$) complex, the enzyme can further stabilize the inherently unstable transition state, for example, by forming extra electrostatic or hydrogen bonds, by providing more effective hydrophobic interactions, or by using structural rearrangements to exclude solvent, thereby strengthening existing electrostatic contacts.

$$\begin{array}{ccccc} E + S & \overset{K_N^{\ddagger}}{\rightleftharpoons} & E + S^{\ddagger} & \xrightarrow{k_N} & E + P \\ \Big\uparrow\downarrow K_S & & \Big\uparrow\downarrow K_T & & \Big\uparrow\downarrow \\ E \cdot S & \overset{K_E^{\ddagger}}{\rightleftharpoons} & E \cdot S^{\ddagger} & \overset{k_E}{\rightleftharpoons} & E \cdot P \end{array} \qquad (17.41)$$

$$\frac{K_T}{K_S} = \frac{K_E^{\ddagger}}{K_N^{\ddagger}} = \frac{k_E}{k_N}$$

Simple transition-state theory states that the rate of an enzyme-catalyzed reaction is correlated with the rate of a noncatalyzed reaction by the same factor as the affinity of an enzyme for the transition state to the affinity of an enzyme for a substrate (Equation 17.41) (99).

Therefore, the magnitude of enzymatic catalysis (k_E/k_N) is related to the enhanced binding of the transition state to the enzyme (K_T/K_S). Compounds that can take advantage of this enhanced binding to the transition state can prove to be potent and selective enzyme inhibitors. Such compounds, referred to as transition-state analogs, can theoretically have ratios of the binding constants of inhibitor to substrate (K_i/K_S) on the order of 10^{-8} to 10^{-14}. In addition, transition-state analogs may have the further advantage of reduced molecularity, as outlined earlier (Section 2.5.2) for multisubstrate analog inhibitors. Several reviews on the theory and general aspects of transition-state analog inhibitors are available and are recommended for a more complete understanding of this topic (37, 96, 99, 100, 124, 125).

The design of a good transition-state mimic is quite challenging. It requires, at the least, sufficient knowledge of the mechanism of the target enzyme to predict transition-state structure(s). This is why transition-state analogs are sometimes (but not in this review) referred to as mechanism-based inhibitors. A detailed knowledge of the true energy profile, including details such as the existence of distinct chemical steps, high-energy intermediates, and their associated transition states, is also useful (126). Further, by definition, the transition state is unstable, often highly charged, and possesses partially broken/formed covalent bonds. Designing a ***stable compound*** that will closely mimic a transition state is impossible. However, the Hammond postulate states that the transition state between a reactant and a high-energy reaction intermediate will resemble the intermediate rather than the reactant. It is possible to design/synthesize an analog of a high-energy intermediate. Indeed, the majority of so-called transition-state analogs are actually analogs of high-energy reaction intermediates. Although a clear distinction exists, the design process is, for all practical purposes, the same.

It should also be noted that an enzyme is designed to initially recognize the features of its substrates. Often substrate binding brings about a conformational change in the enzyme that will then maximize the'attractive forces between the enzyme and transition state. The transition-state analog may not possess those features of the substrate that facilitate rapid binding, even though its affinity for the enzyme is extremely high. Although some transition-state analogs bind rapidly to enzymes,