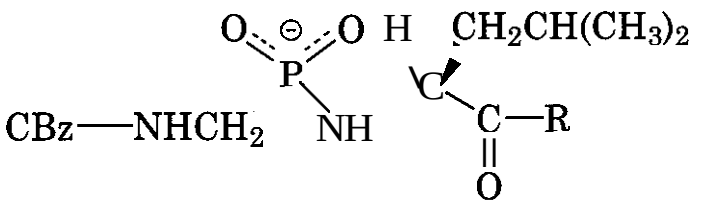
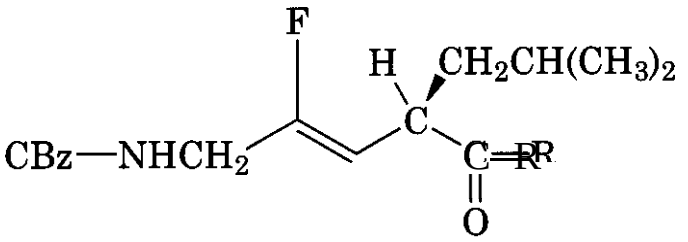
Table 17.6 Correlation of K_i Values for Inhibitors of Thermolysin with K_M and K_M/k_{cat} Values for the Corresponding Substrates[a]

	Inhibitor Data	Corresponding Substrate Data	
	K_i (nM)	K_M (mM)	K_M/k_2 (μM s^{-1})
R = D-Ala	1700	16.6	3200
R = NH_2	760	20.6	196
R = Gly	70	10.8	165
R = L-Phe	78	2.4	20
R = L-Ala	16.5	10.6	13.6
R = L-Leu	9.1	2.6	7.0
	K_i (mM)		
R = Gly	1.80	10.8	165
R = L-Ala	1.48	10.6	13.6
R = L-Leu	0.32	2.6	7.0
R = L-Phe	0.19	2.4	20

[a] Data are from Ref. 127.

tion state (45), and with their long P—O bonds, the phosphonamidate compounds (46, Table 17.6) were expected to a d as transition-state analog inhibitors. It was found that the K_i values for the putative transition-state analog inhibitors correlated linearly with the K_M/k_{cat} values of the corresponding substrates, although no correlation was found between K_i and K_M (Table 17.6). The fact that substrate binding (K_M) was relatively unaffected by a change at a remote site was not unexpected, but the observation that the binding of the phosphonamide inhibitors was greatly affected suggests that these inhibitors were, indeed, transition-state rather than ground-state analogs. Conversely, the K_i values for a series of fluoroalkene isosteres of the same substrates (47) (Fig. 17.22) correlated strongly with K_M but not K_M/k_{cat} (Table 17.6), indicating that the latter inhibitors were ground-state analogs (134). This approach has also been used to confirm that phosphonic acid peptides were transition-state analog inhibitors of pepsin (135).

One of the most popular targets for design of transition-state analogs is adenosine deaminase. Inhibitors of this enzyme have been used as immunosuppressants and are also potential antitumor agents, whereas lack of adenosine deaminase results in severe combined immunodeficiency disease (SCID).

Adenosine deaminase (ADA), which catalyzes the conversion of adenosine to inosine (Equation 17.42), is an extremely proficient enzyme, providing a rate enhancement of more than 12 orders of magnitude (123). The enzyme-catalyzed reaction is thought to pass through an unstable hydrated intermediate (48) (Fig. 17.23), with a K_T (Equation 17.41) in the region of 10^{-17} M (123). Clearly, even a crude analog of (48) would have the potential to be an extremely powerful inhibitor of ADA.

The structures of several inhibitors of ADA are shown in Fig. 17.23. Of these, the antibiotics coformycin (49) and (*R*)-deoxycoformycin (pentostatin, 50) were found to be potent ADA inhibitors, with K_i values of 1×10^{-11} M (136) and 2.5×10^{-12} M (137), respectively. The K_M for adenosine is around 30 μM (138, 139), whereas the K_i of the product, inosine, is 10^{-4} M. Thus, the two antibiotics show at least 10^6-fold greater affinity for ADA than for