where E_a is the activation energy (kcal/mol) and [E] is the active enzyme concentration. A plot of ln υ versus $1/T$ results in a line of slope $-E_a/R$.

The descending part of Fig. 3.15 is known as *temperature inactivation* or *thermal denaturation*. The kinetics of thermal denaturation can be expressed as

$$-\frac{d[\mathrm{E}]}{dt} = k_d[\mathrm{E}] \tag{3.47}$$

or

$$[\mathrm{E}] = [\mathrm{E}_0]e^{-k_d t} \tag{3.48}$$

where $[\mathrm{E}_0]$ is the initial enzyme concentration and k_d is the denaturation constant. k_d also varies with temperature according to the Arrhenius equation.

$$k_d = A_d e^{-E_a/RT} \tag{3.49}$$

where E_d is the deactivation energy (kcal/mol). Consequently,

$$v = Ae^{-E_a/RT}\mathrm{E}_0 e^{-k_d t} \tag{3.50}$$

The activation energies of enzyme-catalyzed reactions are within the 4 to 20 kcal/g mol range (mostly about 11 kcal/g mol). Deactivation energies E_d vary between 40 and 130 kcal/g mol (mostly about 70 kcal/g mol). That is, enzyme denaturation by temperature is much faster than enzyme activation. A rise in temperature from 30° to 40°C results in a 1.8-fold increase in enzyme activity, but a 41-fold increase in enzyme denaturation. Variations in temperature may affect both V_m and K_m values of enzymes.

3.3.6. Insoluble Substrates

Enzymes are often used to attack large, insoluble substrates such as wood chips (in biopulping for paper manufacture) or cellulosic residues from agriculture (e.g., cornstalks). In these cases access to the reaction site on these biopolymers by enzymes is often limited by enzyme diffusion. The number of potential reactive sites exceeds the number of enzyme molecules. This situation is opposite that of the typical situation with soluble substrates, where access to the enzyme's active site limits reaction. If we consider initial reaction rates and if the reaction is first order with respect to the concentration of enzyme bound to substrate (i.e., [ES]), then we can derive a rate expression:

$$v = \frac{V_{\max,\mathrm{S}}[\mathrm{E}]}{K_{\mathrm{eq}} + [\mathrm{E}]} \tag{3.51a}$$

where

$$V_{\max,\mathrm{S}} = k_2[\mathrm{S}_0] \tag{3.51b}$$

and

$$K'_{\mathrm{eq}} = k_{\mathrm{des}}/k_{\mathrm{ads}} \tag{3.51c}$$