A Quick Note on Rate/pH profiles.

In Das & Piccirilli, they plot the $\log_{10}$ of rates/rate constants vs. pH. This makes the interpretation of pK_a easy if you know what you are looking for. Consider a deprotonation to create the active site general base. The fraction of the enzyme that has been deprotonated to create the general base B is:

$$\frac{[\mathrm{B}]}{[\mathrm{Enz}]} = \frac{[\mathrm{B}]}{[\mathrm{B}]+[\mathrm{HB}]} = \frac{[\mathrm{HB}]\mathrm{K_a}/[\mathrm{H^+}]}{[\mathrm{HB}] + [\mathrm{HB}]\mathrm{K_a}/[\mathrm{H^+}]}$$

$$\frac{[\mathrm{B}]}{[\mathrm{Enz}]} = \frac{\mathrm{K_a}}{[\mathrm{H^+}]+\mathrm{K_a}}$$

Thus means that you can have two simplified situations.

(a) When pH is must lower than pK_a ($[H^+] >> K_a$) one has the following:

$$\frac{[\mathrm{B}]}{[\mathrm{Enz}]} = \frac{\mathrm{K_a}}{[\mathrm{H^+}]+\mathrm{K_a}} \approx \frac{\mathrm{K_a}}{[\mathrm{H^+}]}$$

$$\log_{10}\left(\frac{[\mathrm{B}]}{[\mathrm{Enz}]}\right) = \log_{10}\mathrm{K_a} - \log_{10}[\mathrm{H^+}]$$

$$\log_{10}\left(\frac{[\mathrm{B}]}{[\mathrm{Enz}]}\right) = \mathrm{pH} - \mathrm{pK_a}$$

When you plot $\log_{10}$ of the activity (which is proportional to [B]/[Enz]) vs. pH you get a line whose slope is 1 (remember pH is the x variable).

(b) When pH is much greater than pK_a ($[H^+] << K_a$) then the following happens.

$$\frac{[\mathrm{B}]}{[\mathrm{Enz}]} = \frac{\mathrm{K_a}}{[\mathrm{H^+}]+\mathrm{K_a}} \approx \frac{\mathrm{K_a}}{\mathrm{K_a}} = 1$$

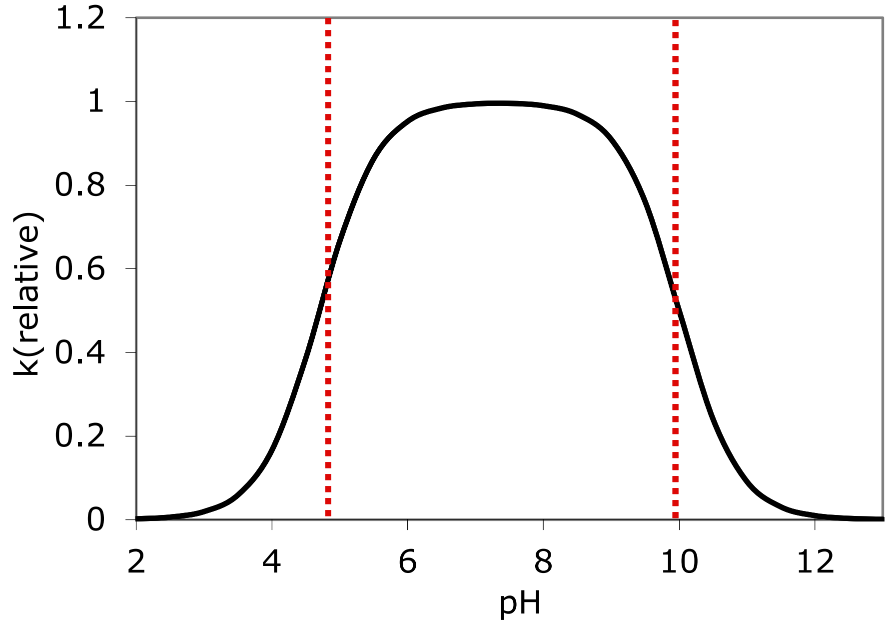
$$\log_{10}\left(\frac{[\mathrm{B}]}{[\mathrm{Enz}]}\right) = 0$$

What really matters here is that [B]/[Enz] reactions a constant value so that you get a horizontal line (slope of 0) at this point.

Thus a plot of $\log_{10}$(activity) vs. pH will yield two straight lines when pH is either much lower or much higher than pK_a. **Those lines will intersect at the point where pH = pK_a** (though the data in that region don't conveniently fall on either line).

See back for example.

Here is the plot of activity vs. pH. Note that the inflection points can be used to ID the pK_a's of the general base (4.7) and the general acid (10.0).



Here is the plot of log activity vs. pH. Note that one can draw straight lines at the three linear portions of the plot, and where they intersect gives pK_a's of the general base (4.7) and the general acid (10.0)