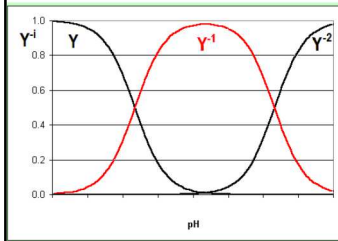


6. OTHER EFFECTS ON ENZYME ACTIVITY

- Ionic strength
- pH
- TEMPERATURE
- Shear
- Pressure (hydrostatic)
- Surface tension
- Chemicals (alcohol, urea, H₂O₂...)
- Light, sonication, ionising radiations

Reverzible changes
 Irreverzible changes

Effect of pH



$$Y^- = \frac{1}{1 + H^+ / K_1 + K_2 / H^+}$$

$$H^+_{\text{optimum}} = \sqrt{K_1 K_2}$$

$$(pH)_{\text{optimum}} = \frac{1}{2} (pK_1 + pK_2)$$

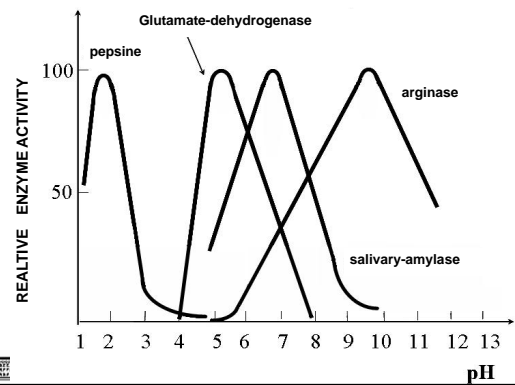
$$V_{\text{max}} = k_2 E_0 Y^- = k_2 E_0 \frac{1}{1 + H^+ / K_1 + K_2 / H^+}$$

Active side chains

Changes in activity of proteins are caused by changes of amino acid side chains.

- Acidic: -COOH: Asp, Glu Basic: -NH₂: Lys, Arg
 (and terminal -COOH and -NH₂)
 amide: -CO-NH₂: Asn, Gln
- Polar: -OH: Ser, Thr -SH: Cys, -S-CH₃: Met
- Imidazole: His Guanidin: Arg
- H-bonds: C=O H-O- C=O H-NH-

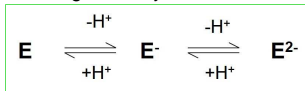
Effect of pH



Effect of pH

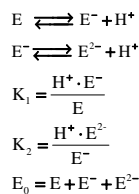
Proteins: + and - charged side chains ← their charge depends on dissociation ← determined by pH → it effects the active centre.

Recharge of enzyme:



Only E⁻ is active!

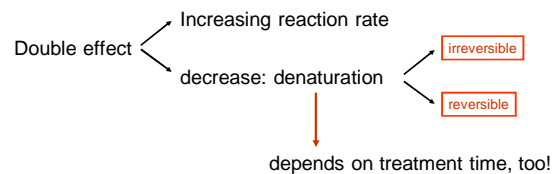
Ratio of active enzymes: $Y^- = E^- / E_0$



Michaelis-féle pH függvények:

$$Y^- = \frac{1}{1 + H^+ / K_1 + K_2 / H^+}$$

Effect of pH temperature



$$\frac{dE_a}{dt} = -kE_a \longrightarrow E_a(t) = E_{a0} e^{-kt}$$

Effect of pH temperature

$$E_a \xrightleftharpoons{K_d} E_i \quad \frac{E_i}{E_a} = K_d = \exp\left(\frac{-\Delta G_d}{RT}\right) = \exp\left(\frac{-\Delta H_d}{RT}\right) \exp\left(\frac{\Delta S_d}{R}\right)$$

$S_d = -900 \text{ KJ/mol.K}$
 $H_d = 280-310 \text{ KJ/mol}$

Large: sensitively reacts on small change (one H-bond: 12,5-29,3 kJ/mol)

If: $V_{max} = k_2(T)E_a$

$$E_0 = E_a + E_i \rightarrow E_a = \frac{E_0}{1 + K_d}$$

$$V_{max} = \frac{\alpha T e^{-E/RT}}{1 + e^{\Delta S^*/R} \cdot e^{-\Delta H_d/RT}}$$

$\alpha = \text{combination of } (\beta, k_2, h, E_0, \Delta S^*)$

and $k_2(T) = \beta \left(\frac{k_B T}{h}\right) e^{\Delta S^*/R} \cdot e^{-E/RT}$

K_a also depends on T!

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Temperature optimum and stability

Temperature °C 20 30 40 50 60 70 80 90 100 110

Microbial rennet ————

A. oryzae α - amylase ————

B. subtilis α - amylase ————

B. licheniformis α - amylase ————

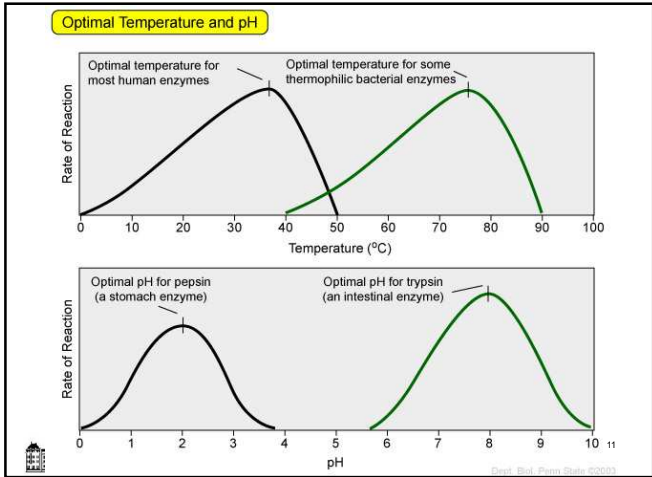
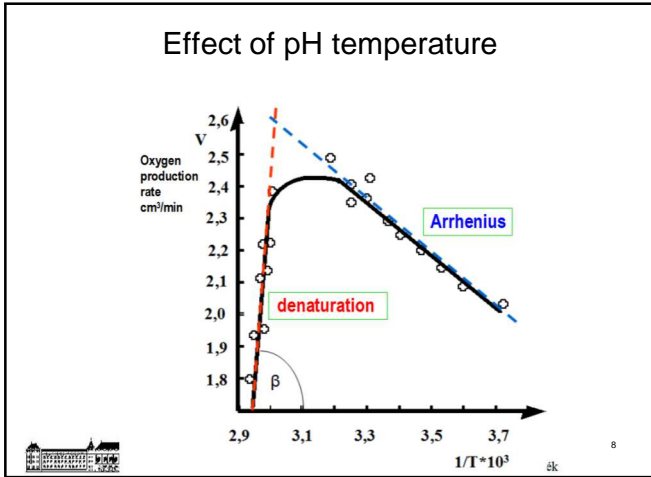
A. oryzae β - galactosidase ————

Stability optimum ————

Activity optimum ————

(optimum set at ≥ 80 % of maximum)

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pH optimum and stability

pH 2 3 4 5 6 7 8 9 10

Microbial rennet ————

A. oryzae α - amylase ————

B. subtilis α - amylase ————

B. licheniformis α - amylase ————

A. oryzae β - galactosidase ————

Stability optimum ————

Activity optimum ————

(optimum set at ≥ 80 % of maximum)

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