CONCEPT: MIXED INHIBITION

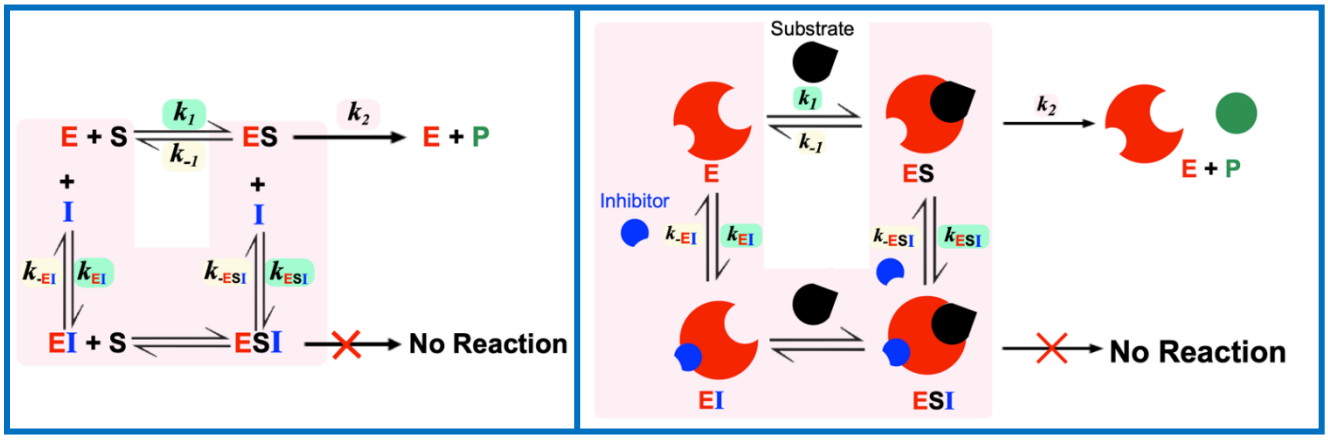
●*Mixed inhibitors*: have _____ binding since they bind either _____ enzyme ***OR*** _____-complex to *decrease* V_0.

□ Binding of a *mixed inhibitor* to E or ES-complex ultimately _____ conversion of **S** → **P**.

□ No competition with **S** since mixed-**I** bind to _____ sites: *alternative* sites on **E** other than active site.

□ Mixed inhibitors can bind with _____ *affinities* to the *free enzyme* & to the *ES-complex* ($K_I \neq K'_I$).

EXAMPLE: Mixed inhibition.



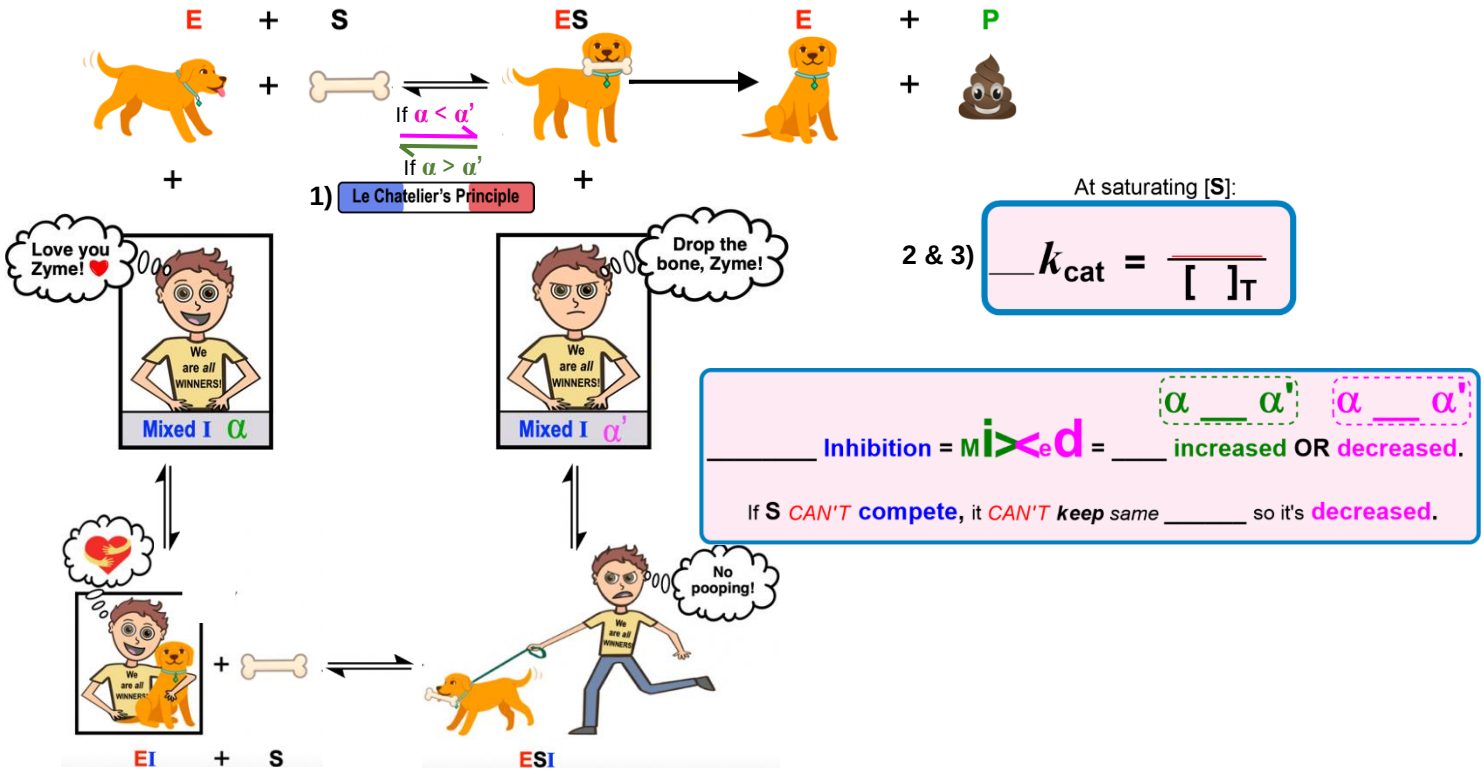
Mixed Inhibitor Effects

●With mixed inhibitors, the K_m^{app} may _____ increase *OR* decrease, but V_{max}^{app} is always _____.

1) By Le Chatelier's Principle, magnitude of ___ & ____ dictate reaction shift ($\alpha > \alpha'$ shift **left**; $\alpha < \alpha'$ shift _____).

2) Since **S** *can't* outcompete mixed inhibitors, effects are *NOT reversed* by ____ [**S**], so V_{max}^{app} is *decreased*.

3) Since mixed inhibitors *decrease* V_{max}^{app}, k_{cat} is also _____.



At saturating [**S**]:

2 & 3) $\_\_k_{cat} = \frac{\_\_\_\_}{[\ ]_T}$

α ___ α' α ___ α'

_____ Inhibition = Mixed = ____ increased OR decreased.

If **S** *CAN'T* **compete**, it *CAN'T* ***keep*** *same* _____ so it's **decreased**.

CONCEPT: MIXED INHIBITION

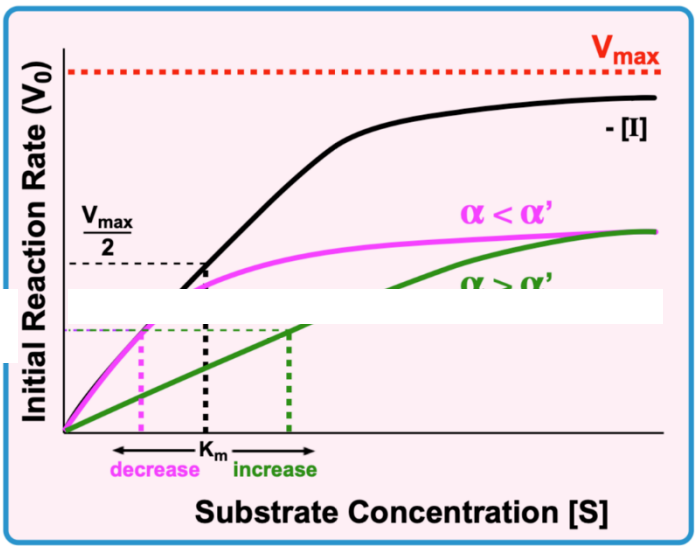
Mixed Inhibition & Michaelis-Menten-Plots

●Recall: mixed inhibitors bind to either *free enzyme* ***OR*** *ES-complex*, so ____ & ____ measures its *degree of inhibition*.

- □ α' always _____ V_{max}^{app} (V_{max}/α'), but *ratio* of α to α' can either increase or decrease K_m^{app} ($\alpha K_m/\alpha'$).
- □ *Greater* degree of inhibition to the free enzyme relative to ES-complex ($\alpha > \alpha'$) means K_m^{app} will be _____.
- □ If $\alpha = \alpha'$, then the K_m^{app} is not changed & the inhibitor is called a ____ncompetitive inhibitor.

Mixed Inhibitor Michaelis-Menten Equation:

$$V_0 = \frac{\left(\frac{V_{max}}{\alpha'}\right)[S]}{\left(\frac{K_m}{\alpha'}\right) + [S]}$$



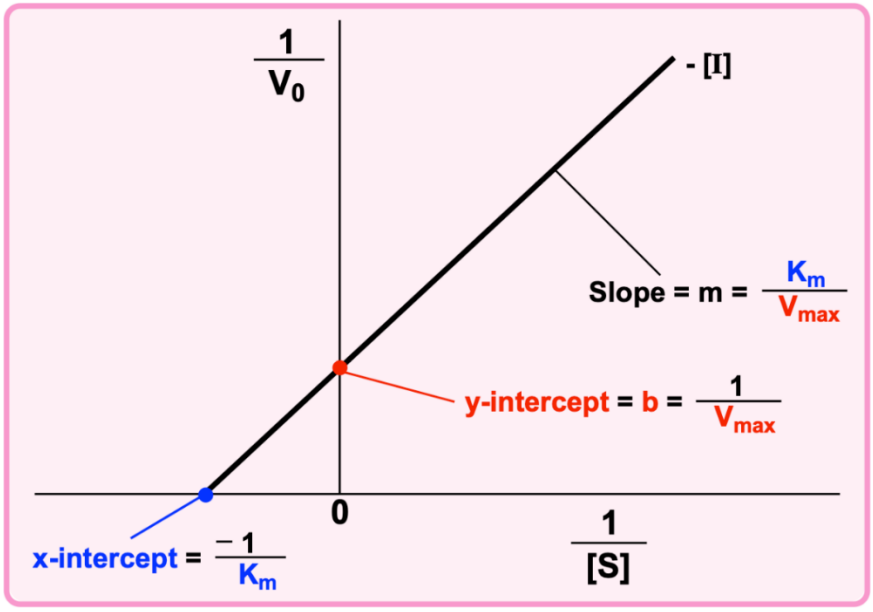
Mixed Inhibition & Lineweaver-Burk-Plots

●Recall: mixed inhibitors always *decrease* the V_{max}^{app} but can either increase ***OR*** decrease the K_m^{app}.

- □ *Mixed inhibitors* can change the slope of the line on a LW-Burk plot (slope = K_m/V_{max}) in _____ ways.
- □ ___-intercept ($1/V_{max}$) on a LW-Burk-Plot always *increases*, but ___-intercept ($-1/K_m$) can decrease or increase.

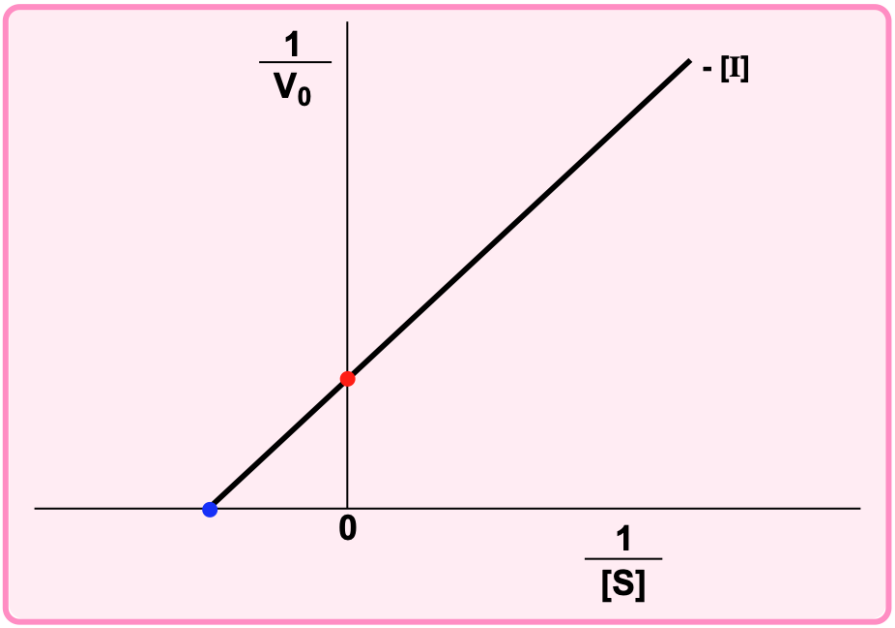
Mixed Inhibitor Lineweaver-Burk Equation:

$$\frac{1}{V_0} = \frac{\alpha K_m}{V_{max}}\left(\frac{1}{[S]}\right) + \frac{\alpha'}{V_{max}}$$



CONCEPT: MIXED INHIBITION

PRACTICE: Draw the representative lines for enzyme activity for an inhibitor with $\alpha > \alpha'$ and a separate line with $\alpha < \alpha'$.



PRACTICE: When a mixed inhibitor favors binding to the enzyme-substrate complex (ES) over the free enzyme (E), the apparent substrate affinity (apparent K_m) is:

a) Greater than the substrate affinity for E (K_m)

b) Less than the K_m

c) Equal to the K_m

PRACTICE: Complete the chart by indicating with an "x" which kinetic parameters are affected by each factor.

K_m	V_{max}	Both	Neither	Factor
				Competitive Inhibitor
				Mixed Inhibitor
				6M Urea
				Doubling [S]