# *Understanding Enzymes: Gateway Concepts of Reactions (and Interactions) from Chemistry*

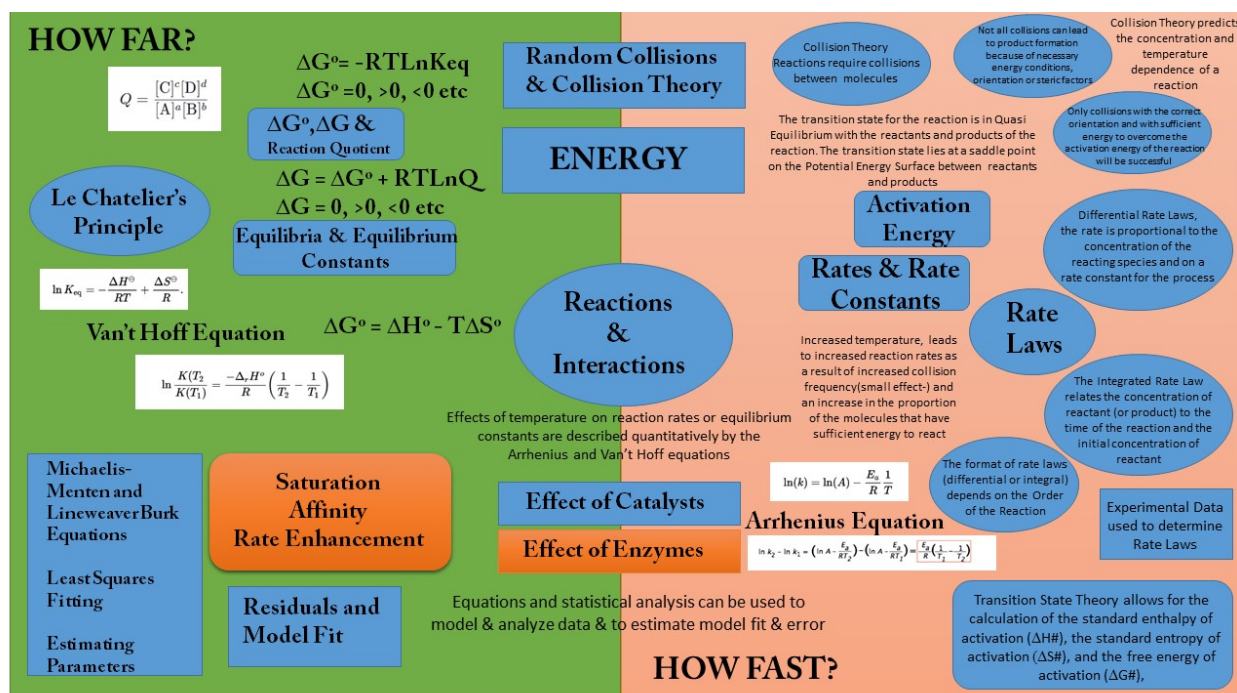*Ellis Bell*

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## Synopsis

Enzymes are biological catalysts that enable cells to control the wide variety of chemical reactions that continuously occur in a cell. Enzymes enable these processes to occur at ambient temperatures, with the requisite specificity, and unlike chemical catalysts, exhibit the phenomenon of saturability. The reactions catalyzed by enzymes often have mechanisms for regulation of the rates of the reactions. While many enzymes are proteins, some RNA molecules also exhibit enzymatic activity and are termed "catalytic RNA". In all cases the interactions of enzymes with their substrates, products and if appropriate, regulatory molecules, are governed by

the same foundational concepts that govern chemical reactions in general. Presented here are 5 "gateway" concepts necessary to understand the action of enzymes from a chemical perspective.
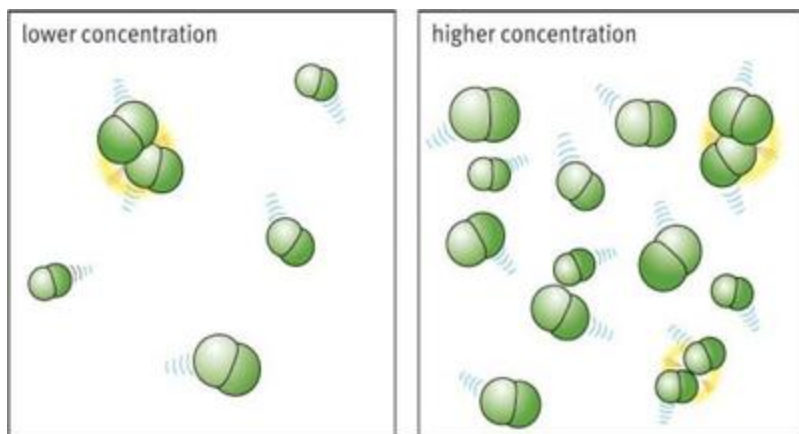
### 1: Collision theory

Reactions require collisions between molecules

Not all collisions can lead to product formation because of necessary energy conditions, orientation or steric factors
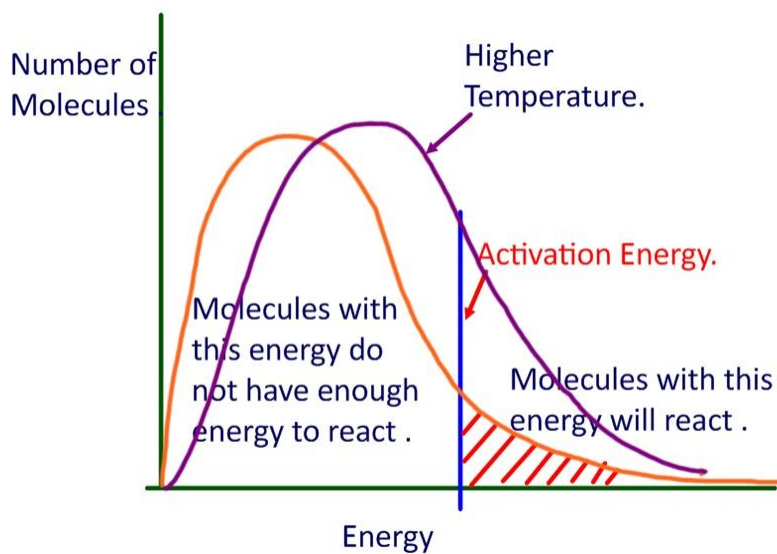
Only collisions with the correct orientation and with sufficient energy to overcome the activation energy of the reaction will be successful



collision 1

collision 2

collision 3
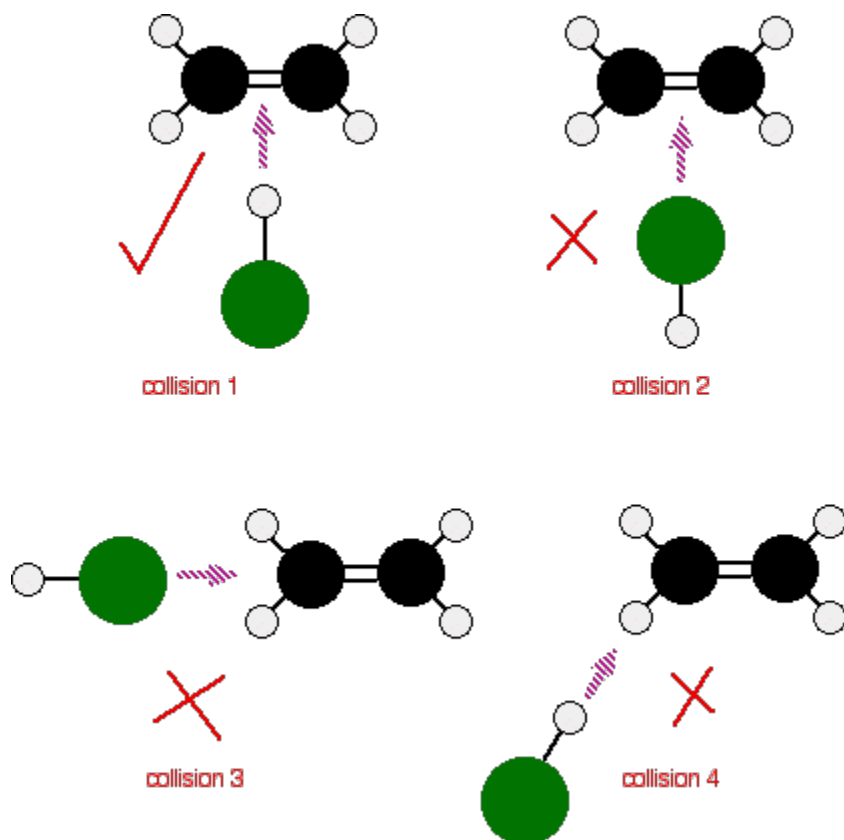
collision 4

Collision Theory predicts the concentration dependence of a reaction

Collision Theory predicts the temperature dependence of a reaction



**2: Transition state theory**

Reactions proceed through formation of an "activated complex" that lies at a saddle point on the Potential Energy Surface between reactants and products. This activated complex is the Transition State of the reaction.

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**(a)**



**(b)**
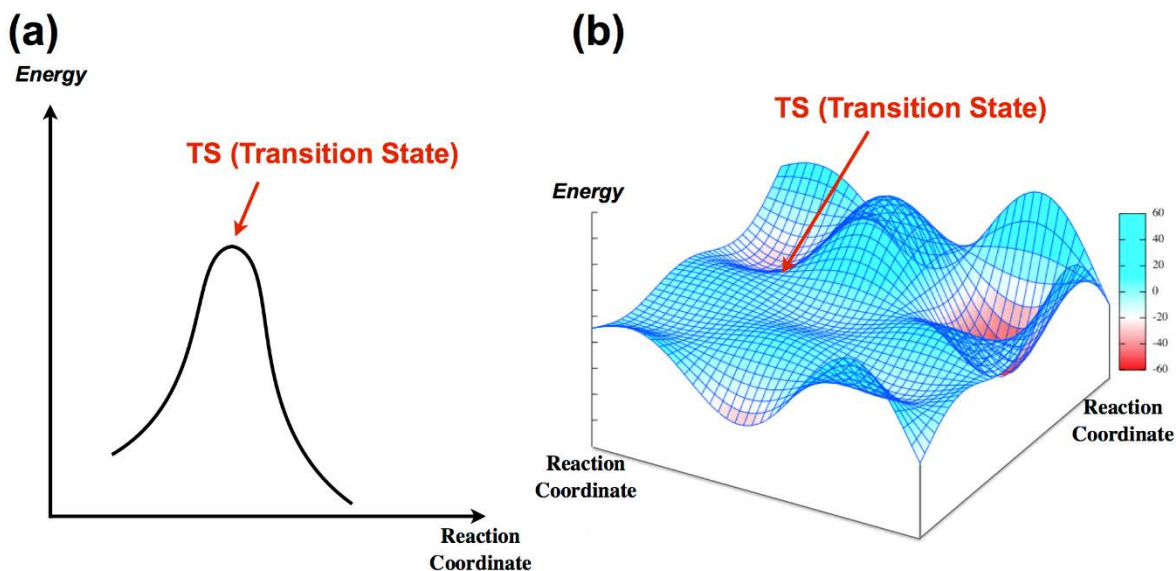


The transition state for the reaction is in Quasi-Equilibrium with the reactants and products of the reaction

The transition state can convert to products and kinetic theory allows calculation of the rate of this process.

Transition State Theory allows for the calculation of the standard enthalpy of activation ($\Delta H^{\#}$), the standard entropy of activation ($\Delta S^{\#}$), and the free energy of activation ($\Delta G^{\#}$), from experimental data using the Eyring Equation (which resembles the Arrhenius Equation) relating the reaction rate to the temperature.

### 3: Rate laws, Steady States and Equilibria

Rate laws for a reaction (which are usually experimentally determined) describe the dependence of the rate of the reaction on either concentration of the reactant(s) (Differential Rate Law) or the time of the reaction with a fixed starting concentration (Integrated Rate Law)

| Order | Rate | Integrated Rate Law |
|-------|------|---------------------|
| 0 | $r = k[A]^0 = k$ | $[A]_t = -kt + [A]_o$ |
| 1 | $r = k[A]$ | $\ln[A]_t = -kt + \ln[A]_o$ |
| 2 | $r = k[A]^2$ | $\dfrac{1}{[A]_t} = kt + \dfrac{1}{[A]_o}$ |

In Differential Rate Laws, depending upon the order of the reaction, the rate is proportional to the concentration of the reacting species and on a rate constant for the process.

The Integrated Rate Law relates the concentration of reactant (or product) to the time of the reaction and the initial concentration of reactant.

The format of rate laws (differential or integral) depends on the Order of the Reaction

In a Steady State, the concentrations( [x]) of the components of a system do not change over a period of time (t) and can be represented by $\delta[x]/\delta t = 0$.

[Int]

Steady State Zone

time
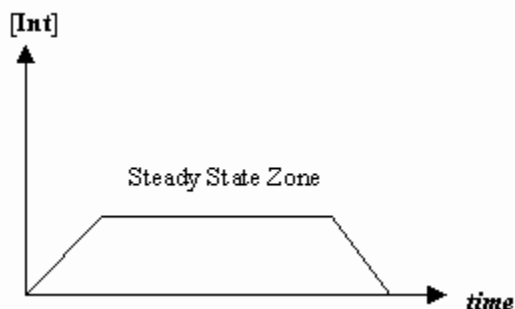
While a steady state is not necessarily at Equilibrium, an equilibrium, by definition, is a steady state and defined by an equilibrium constant, $K_{EQ}$ = [Products]/[Reactants]

As a result of rate laws, the Equilibrium Constant for a reversible reaction is related to the individual rate constants for the forward and reverse reactions ($K_{EQ} = k_f/k_r$ and the energy difference between reactants and products( $\Delta G^o = -RT\ln K_{EQ}$ )

### 4: The effects of temperature

Increased temperature, in general, leads to increased reaction rates as a result of increased collision frequency(small effect-frequency of collisions is proportional to square root of $^oK$) and an increase in the proportion of the molecules that have sufficient energy to react(large effect due to shift in Maxwell Boltzmann distribution etc) when they collide correctly.
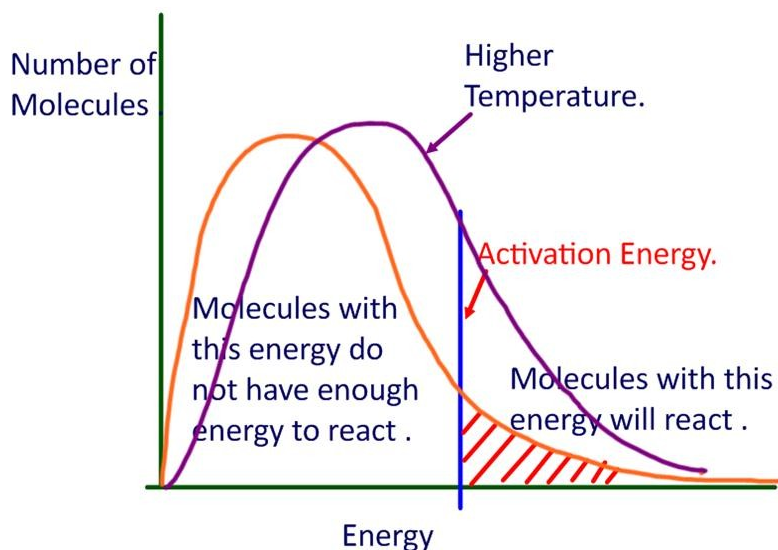
Increased reaction rates, reflected in the rate constants for the reaction may affect forward and reverse reactions differently resulting in equilibrium constant effects of temperature.

In the energy diagram for a reversible reaction, increased temperature in effect "raises" the energy of the reactants while decreased temperature lowers the "energy" of the reactants. Temperature changes have little effect on the energy of the transitation state.
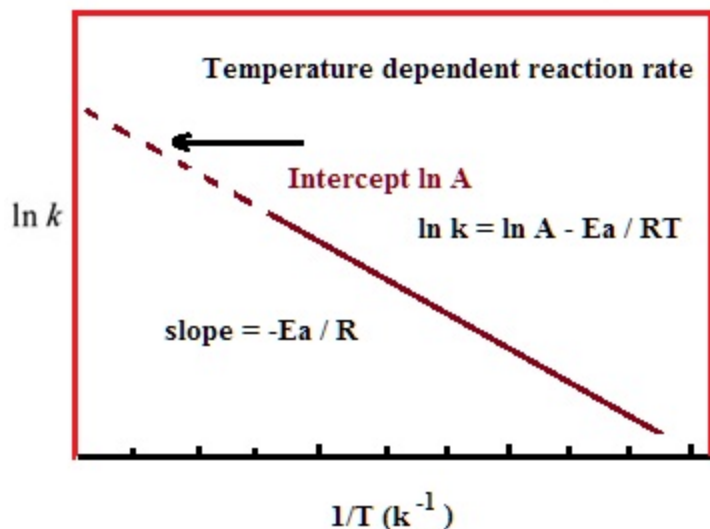
The effects of temperature on reaction rates or equilibrium constants are described quantitatively by the Arrhenius and Van't Hoff equations respectively and allow the appropriate thermodynamic data to be calculated from the temperature dependence of either rate or equilibrium constants.

Arrhenius Equation

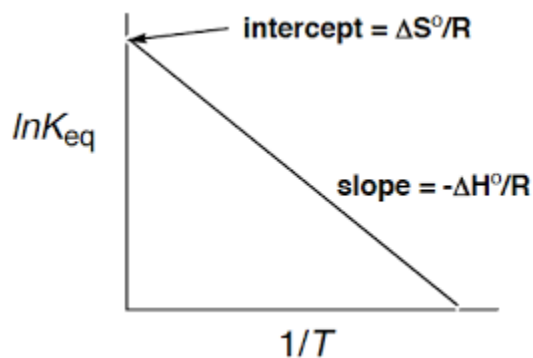$$k = Ae^{-\frac{E_a}{RT}} \quad or \quad ln\ k = -\frac{E_a}{RT} + ln\ A$$

Where:
k = Chemical Reaction Rate
A = Pre-exponential Factor
$E_a$ = Activation Energy
R = Gas Constant
T = Temperature in Kelvin

**Temperature dependent reaction rate**

$\ln k$

**Intercept ln A**

$\ln k = \ln A - Ea / RT$

slope = -Ea / R

$1/T\ (k^{-1})$

Vant Hoff Equation

$$\ln K_{eq} = -\frac{\Delta H^{\ominus}}{RT} + \frac{\Delta S^{\ominus}}{R}.$$

intercept = $\Delta S^{o}/R$

$lnK_{eq}$

slope = $-\Delta H^{o}/R$

$1/T$

As a general rule of thumb in biological systems a $10^{o}K$ rise in temperature approximately doubles the rate of a reaction.

### 5: Structure and reactivity

Chemical Reactions involve bond breaking or bond making events and involve changes in the electron sharing within or between molecules and are usually discussed in terms of three fundamental concepts: i) nucleophiles and electrophiles, ii) acid-base chemistry, and iii) redox reactions.

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Nucleophiles and electrophiles: the electron density of a given molecule determines its reactivity which can be influenced by the electron density of whatever molecule it is reacting with. Nucleophiles are regions of a molecule that are electron rich, while electrophiles are regions that are electron deficient.
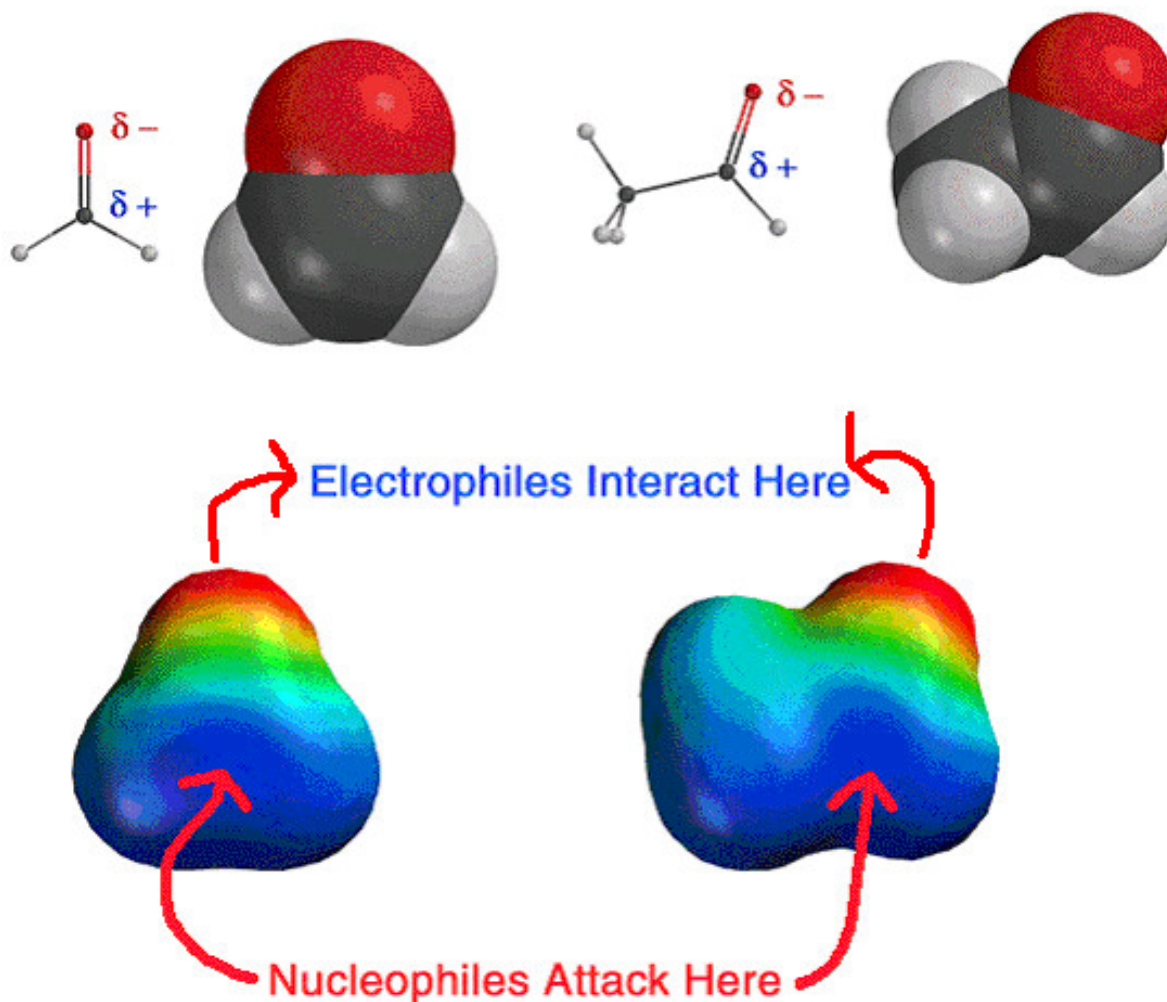


Nucleophilicity increases as you increase basicity- the conjugate base of a compound is always a stronger nucleophile. The most common nucleophiles contain lone pairs or pi bonds(especially with electron donating groups attached), although sigma bonds can be involved in some cases. Substitution reactions often include water as the nucleophile, leaving groups including role of ATP to make good leaving group (weak bases-weaker the conjugate base, the better the leaving group). The strength of an electrophile is governed not only by its electron deficiency but also by steric considerations and the stability of a potential carbocationic intermediate in $s_N2$ reactions.

Modern concepts of acids and bases are derived from the Arrhenius theory (acids produce hydrogen ions, bases hydroxide ions), and include the Bronsted-Lowery theory (an acid is a proton donor while a base is a proton acceptor) & Lewis theory (an acid is an electron pair acceptor while a base is an electron pair donor).

## Acid/Base Venn Diagram
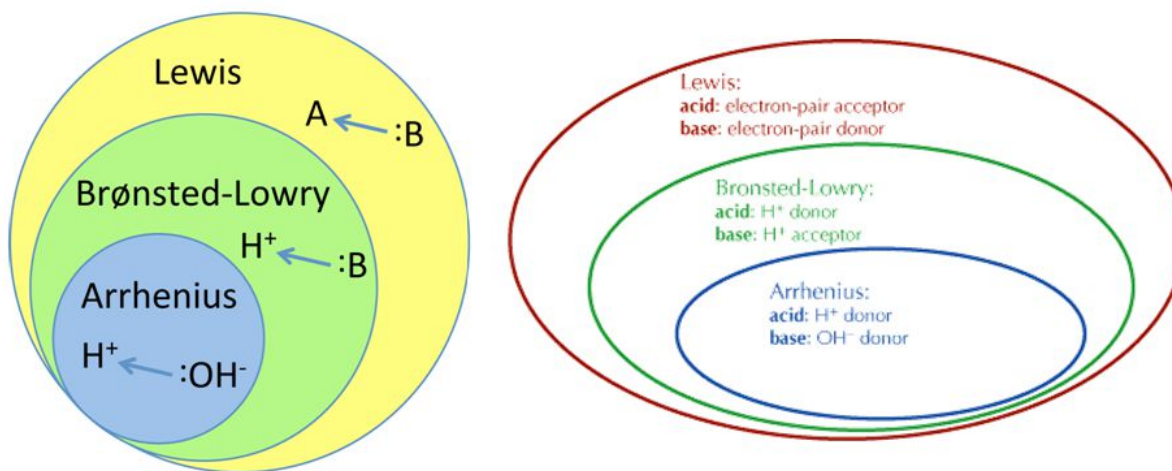


All Brønsted-Lowry acids are Arrhenius acids.

Not all Arrhenius acids are Brønsted-Lowry acids.

etc…

Acid-base chemistry in the molecular life sciences mostly involves weak acids and bases (only partially ionized in water) where $pKa = -\log Ka$ ($Ka = ([A^-][H^+])/[HA]$. The Henderson-Hasselbach equation ($pH = pKa + \log[A^-]/[HA]$) can be used to assess the state of ionization of a group at any pH. There is a relationship between weak acidity to structure (the stability of the conjugate base etc- eg serine vs cysteine). Changing the local environment (charge environment, polarity of environment etc) has an effect on the pKa of a weak acid. TheHenderson Hasselbach equation is also the foundation for understanding buffers and titration curves.
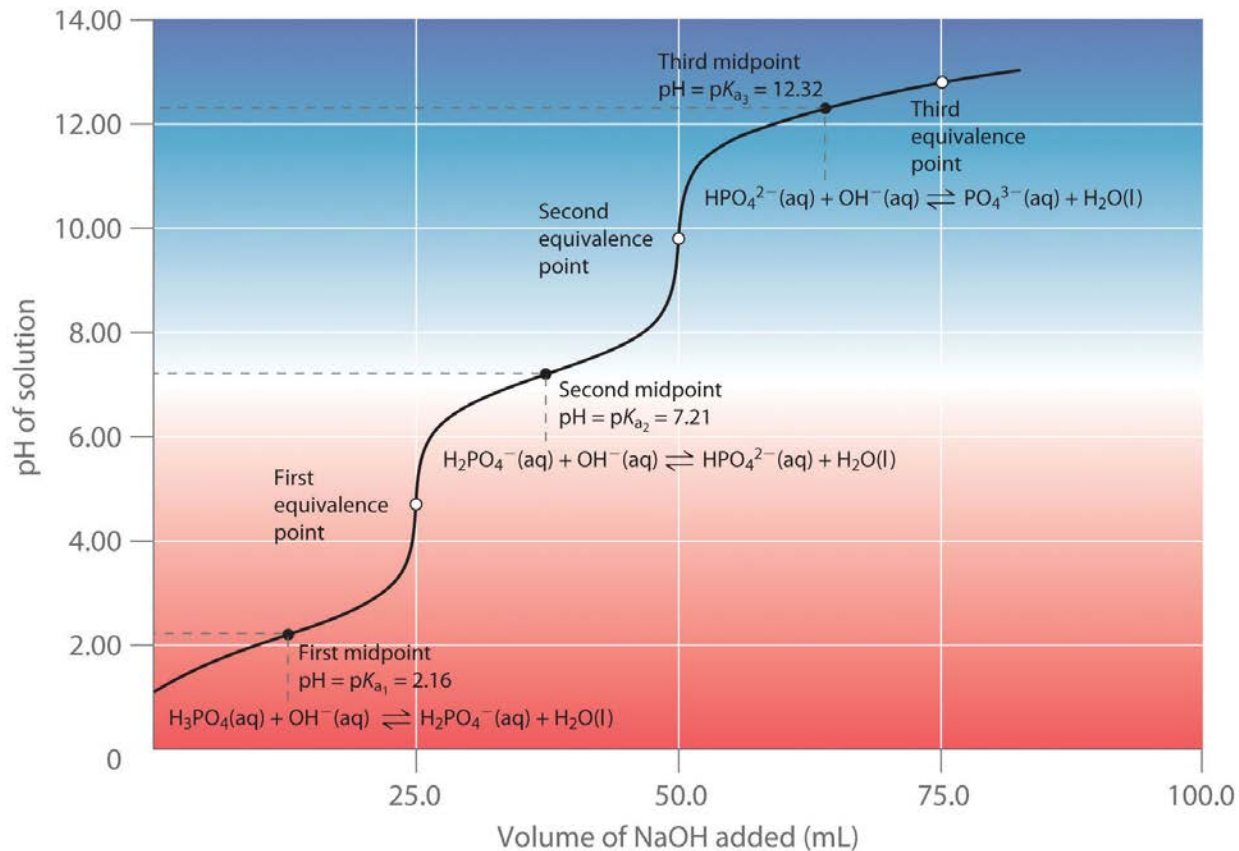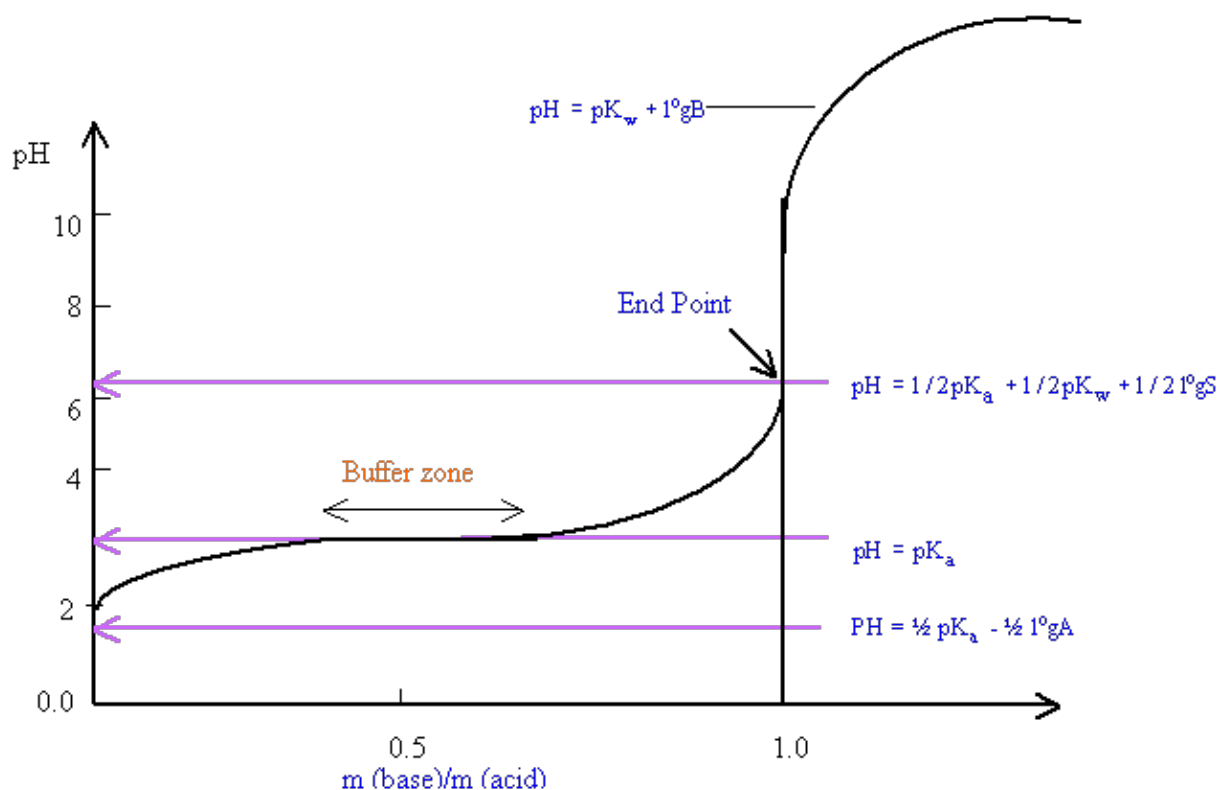
In Redox reactions, oxidation is loss of electrons, while reduction is gain of electrons. In such a reaction you can consider two "half reactions" but you cannot have one half reaction without the other-half reactions. An oxidizing agent gains $e^-$ during reaction and is therefore reduced during reaction while a reducing agent loses $e^-$ during reaction and is therefore oxidized during reaction. ( The oxidized form of a molecule is the form of molecule lacking $e^-$ and the reduced form of a molecule is the form of the molecule having $e^-$.
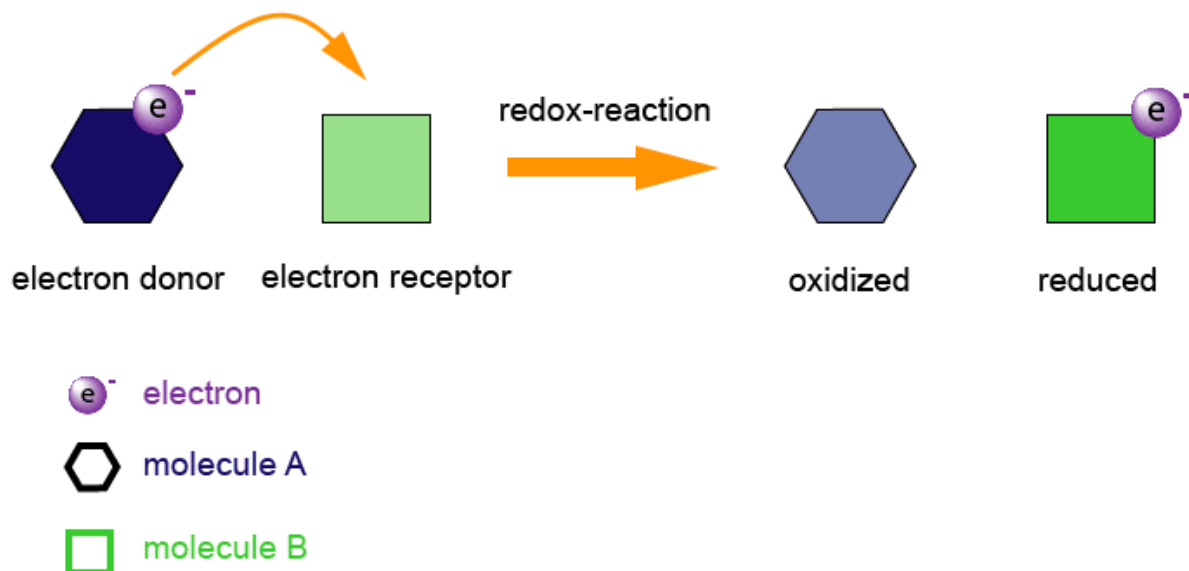
Oxidation numbers are often used to consider molecules undergoing redox reactions. For atoms in their elemental form, the oxidation number is 0. For ions, the oxidation number is equal to their charge. For single hydrogen atoms, the number is usually +1 (but in some cases it is -1). For oxygen, the number is usually -2. The sum of the oxidation number (ONs) of all the atoms in the molecule or ion is equal to its total charge.

For the general redox reaction (written as a reduction) aA + n e-⟷ bB the Nernst equation takes the form:

$$E = E^o - (RT/nF) \log [(A_R)^b / (A_O)^a ]$$

Where E is the measured electrode potential, $E^o$ is the standard reduction potential, R is the gas constant (8.314 T is temperature in K, n is the stoichiometric number of electrons involved in the process, F is the Faraday constant (96,485 C/equivalent) and $A_R$ and $A_O$ are the activities of the reduced and oxidized members of the redox pair, respectively

In considering redox reactions you need to account for all of the electrons as they transfer from one species to another, and deal with each of the two half-reactions individually

**Overview of types of enzyme chemical mechanisms:**

Six basic types of enzyme activities exist based in large part on the type of chemical reactions they catalyze. As categorized by the Enzyme Commission, these are: 1)Oxidoreductases: involve oxidation/reduction reactions- movement of electrons between molecules, 2) Transferases- mostly substitution reactions (usually with alcohol, amine or thiol as nucleophile), 3) Hydrolases- mostly substitution with water as nucleophile, 4) Synthases/Lyases- reactions where two molecules are brought together (or broken apart) that is not already class 1 or 3- can be aldol reactions, eliminations, additions, etc., 5) isomerases- where you switch between isomers

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(constitutional or stereo). Can involve cofactors. Can involve acid/base chemistry or substitution etc. and 6) Ligases/Synthetases- usually substitution reactions, but need ATP (usually) to make a good leaving group.

Images from Google Image

Mindmap from Project Mindmap, https://www.molecularlifesciences.org/