Regulation of Enzyme Activity

Manickam Sugumaran

Professor of Biology

U.Mass - Boston

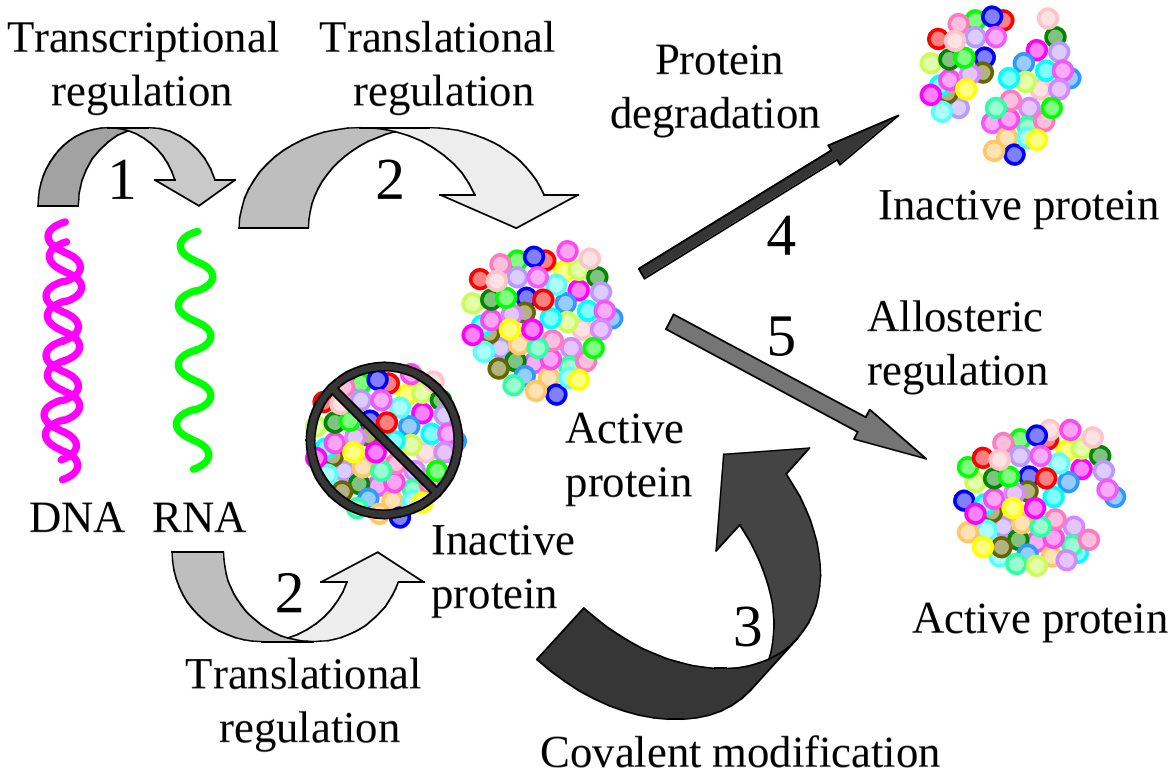
Boston, MA 02125

The Theme of This Lecture

Regulation of Enzyme Activity at Protein Level.

1. **Covalent modification.**
2. **Noncovalent (allosteric) regulation**
3. **Protein degradation (will not be considered).**

Regulation at Different Levels



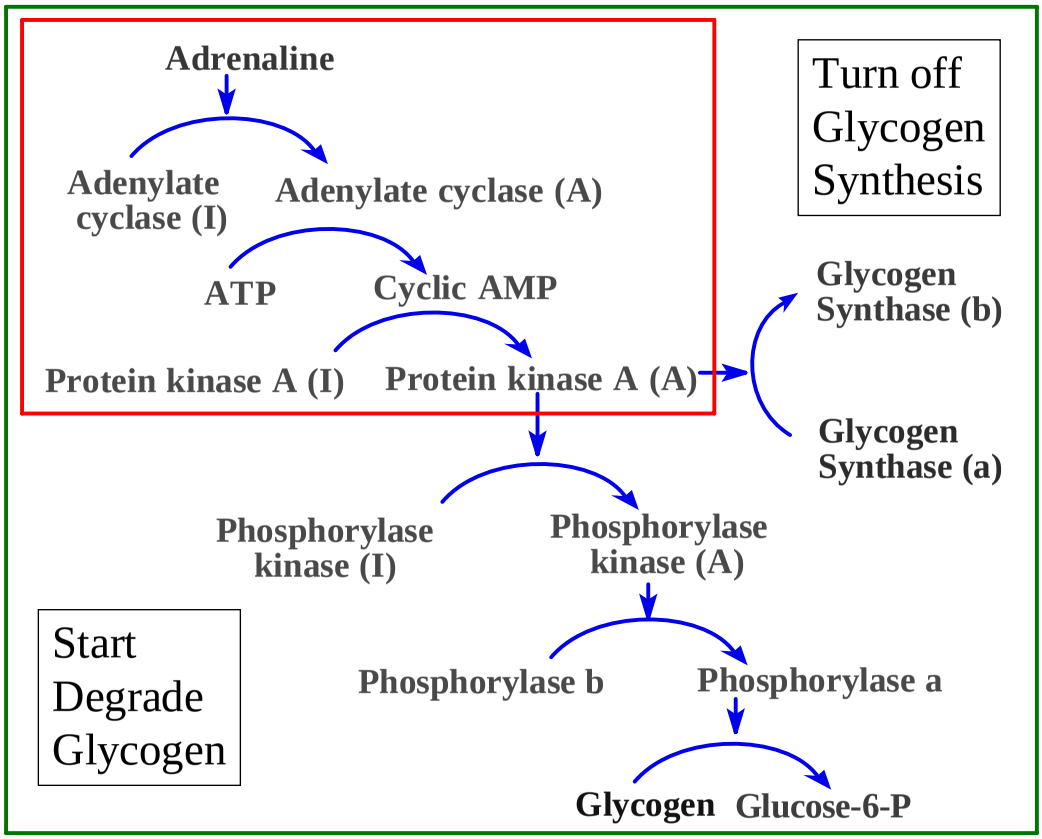
Why regulate?

- **In the cell, enzymes do not work alone but often work together in groups. These sets of reactions are called metabolic pathways. Given the fact the enormous amount of energy and resources are dedicated for each pathway to carry out different metabolic functions, the cells have to regulate the activities of the enzymes very precisely.**

- **Regulation will allow the changing needs of the cell to meet its energy and resource demands. If a product is available in excess, it could then divert the resources to other needy reactions. If a product is in demand, it could activate the pathway to produce more of the biomolecule that is needed.**

- **Thus regulation is the process by which cells can turn on, turn off, or modulate the activities of various metabolic pathways.**

You see the lion. You are afraid!! Start running

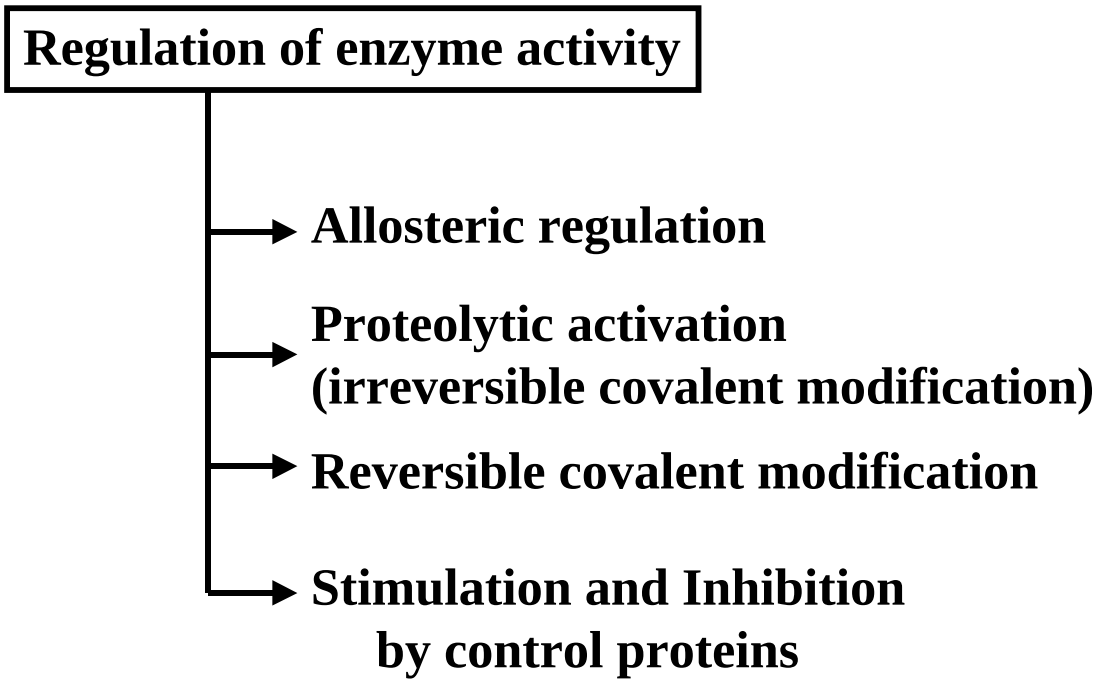


And get the ATP from glucose-6-P for running !!

Amplification of enzyme activity

- Approximately 0.1 nmoles of adrenaline per gram of muscle will trigger the synthesis of 25 μmoles of glucose -1- phosphate per minute per gram of muscle.
- **This represents an amplification of 250,000 fold.**

Four kinds of regulation



Proteolytic activation

This kind of activation is irreversible.

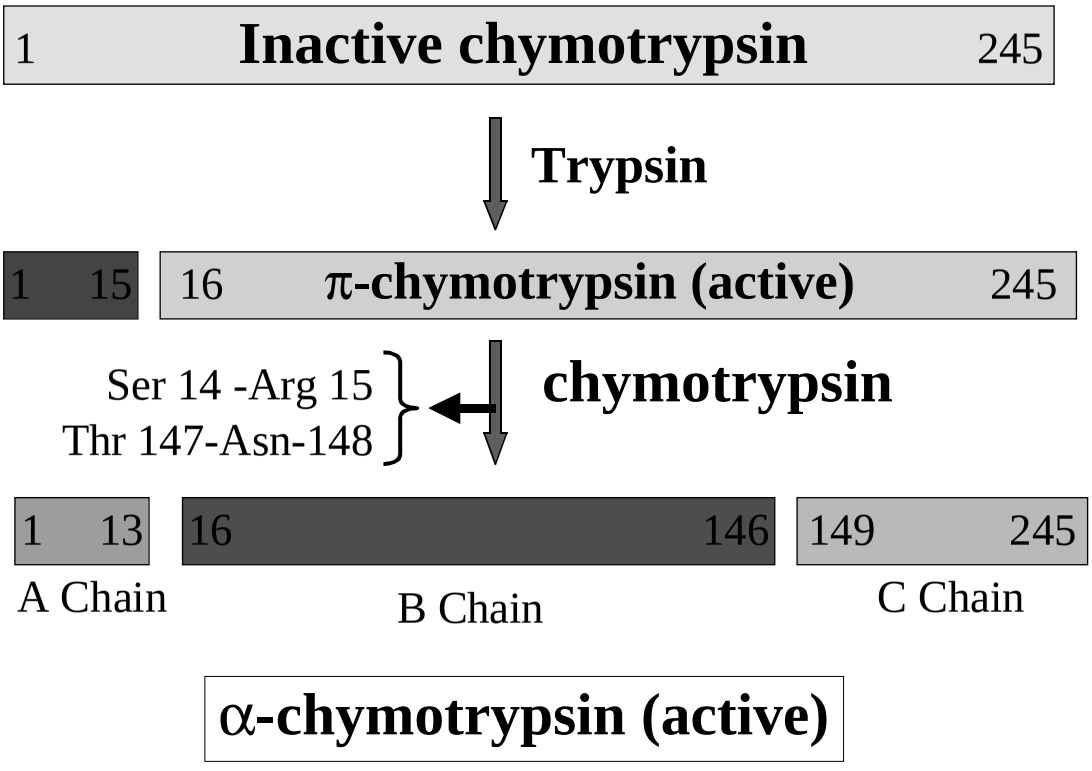
**Once the protein is activated,
the process cannot be reversed.**

**Active protein can only be controlled
by other kinds of regulation,
such as inhibition or inactivation.**

Examples of Proteolytic Activation

- **Zymogen activation.**
- **Blood clotting.**
- **Complement activation.**
- **Prophenoloxidase activation.**
- **Inactive hormones to active hormones.**

Activation mechanism of Chymotrypsin



Reversible Covalent Modification

A single trigger rapidly switches a whole pathway on or off

Examples of reversible covalent modifications

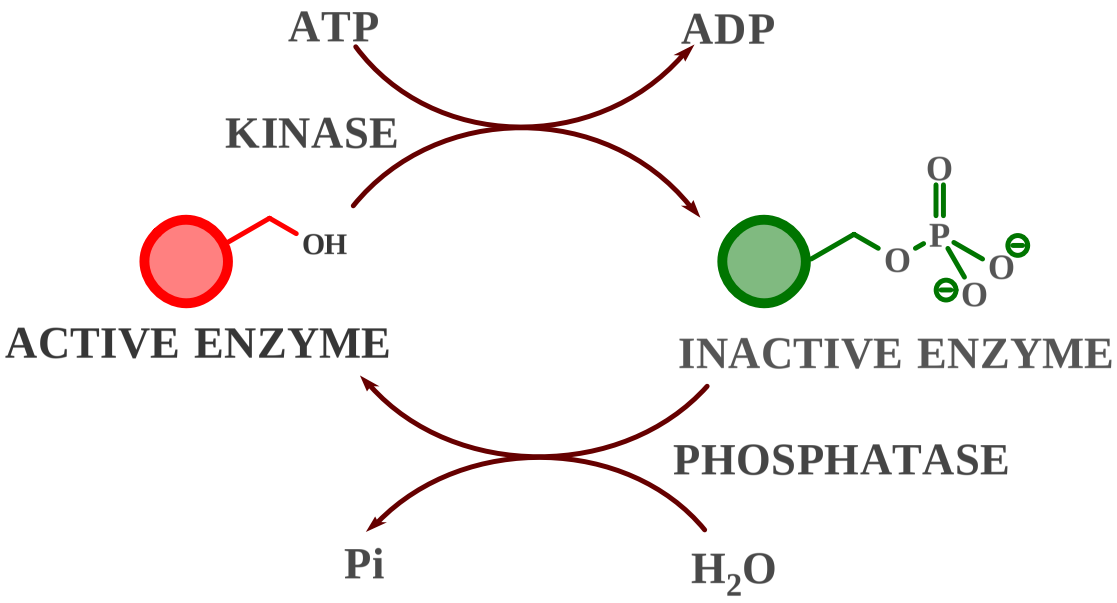
- **Phosphorylation - dephosphorylation**
- **Adenylation - deadenylation.**

Reversible covalent modification - Phosphorylation

While running, glycogen phosphorylase activity is enhanced by phosphorylation.

At the same time glycogen synthase activity is shut off by phosphorylation.

Regulation - covalent modification

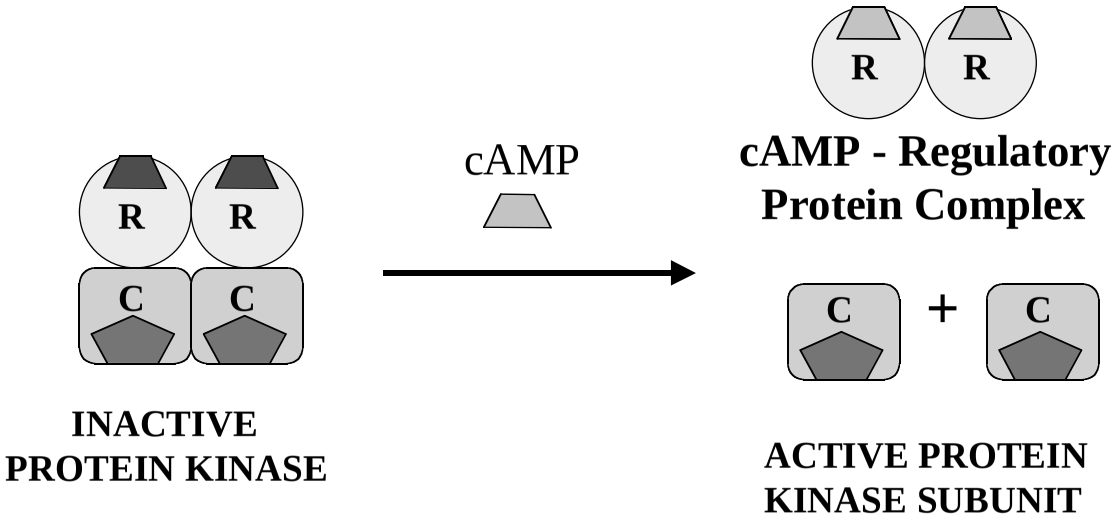


PHOSPHORYLATION AND DEPHOSPHORYLATION

OF SERINE (THREONINE AND TYROSINE)

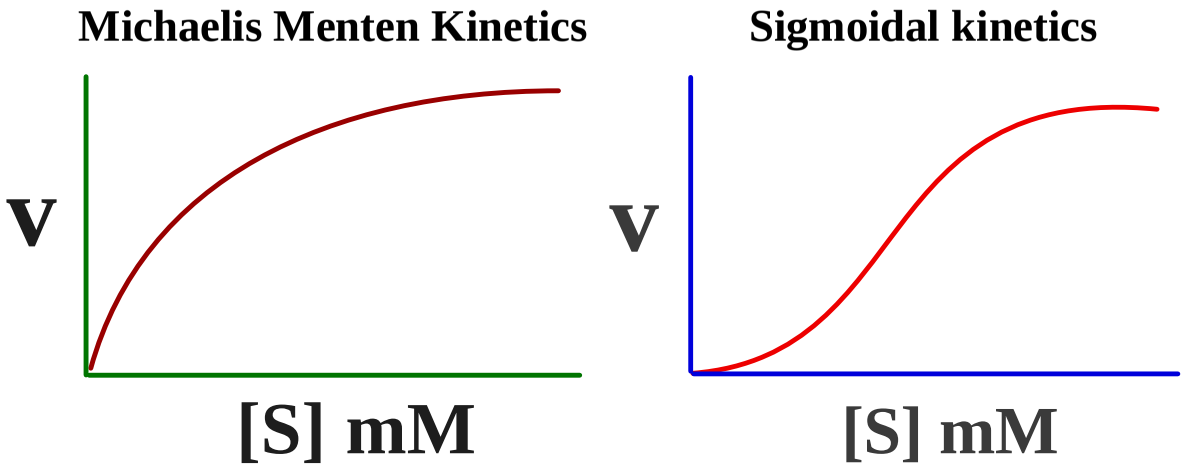
Stimulation and inhibition by control proteins

ACTIVATION OF PROTEIN KINASE BY CYCLIC AMP

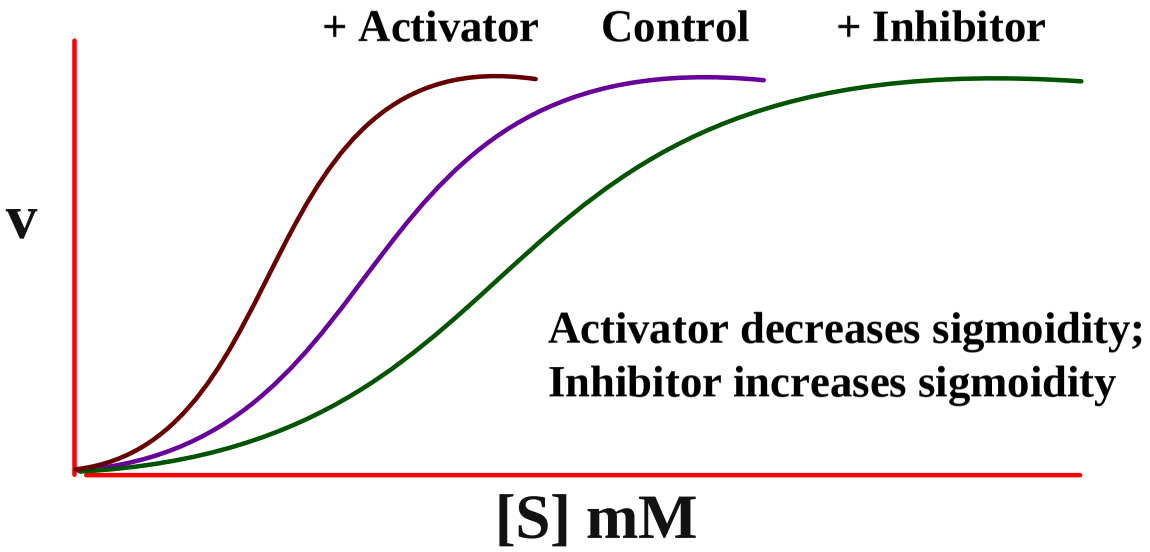


ALLOSTERIC PROTEINS SHOW SIGMOIDAL KINETIC BEHAVIOR

Normal Protein Allosteric Protein



Effect of activator and Inhibitor



For ATCase, ATP is an allosteric activator
CTP is an allosteric inhibitor

Allosteric enzymes

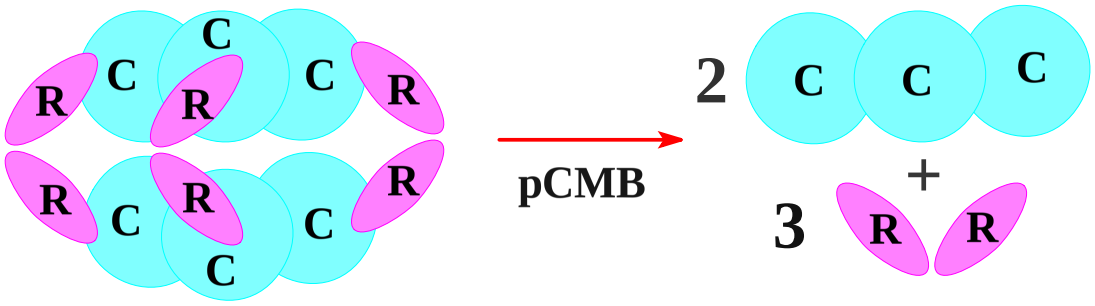
- Do not exhibit Michaelis Menten type kinetics.
- Show sigmoidal kinetic behavior.
- Must undergo drastic conformational changes upon binding of modulatory ligand.
- Usually possess catalytic and regulatory subunits.
- Therefore, they are larger and more complex than non-allosteric enzymes.

Allosteric regulation

- If the substrate itself is regulator, it is called homotropic interaction.
- If it is a different ligand, it is called heterotropic interaction (can be an activator or an inhibitor).
- The sigmoidal graph arises due to cooperativity or subunit interaction.
- Binding of a ligand causes conformational changes in a subunit that may or may not be transmitted to other subunits.

Desensitization

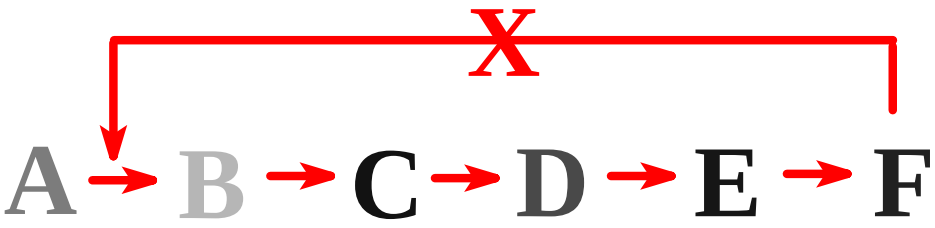
ATCase treated with mercurials do not exhibit allosteric kinetics. This desensitization is caused by the separation of catalytic and regulatory subunits by the reaction with mercurials.



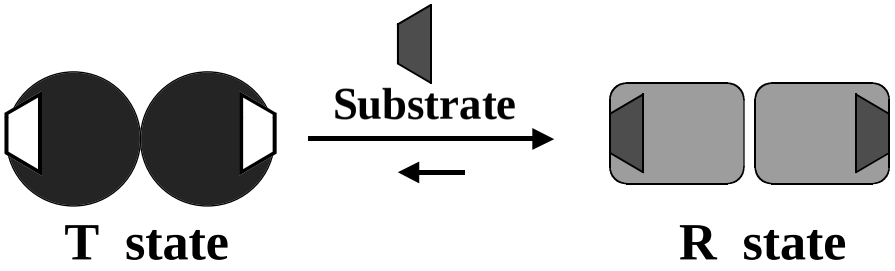
ATCase Subunits

End Product Inhibition

If F is available by some chance, the pathway should be shut off.
End product inhibition provides this regulation



Concerted model or symmetry model



The enzyme exists only in two states

The two states are T (taut or tensed) and R (relaxed)

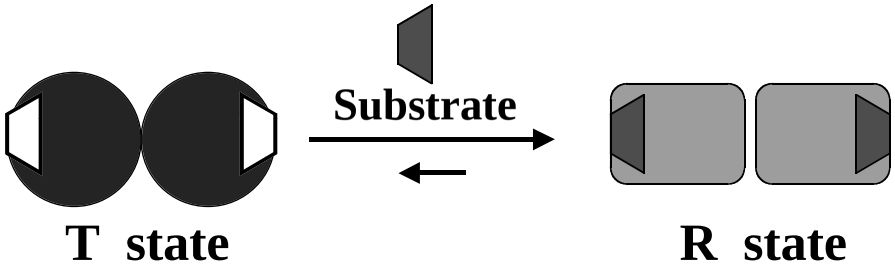
Substrates and activators have great affinity for R state

Inhibitors have higher affinity for T state

Ligands affect the equilibrium between T and R states

While going from one state to the other symmetry must be conserved.

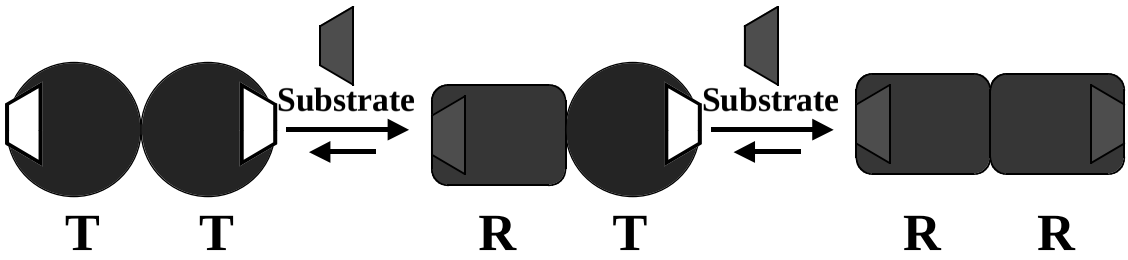
Concerted model or symmetry model



Binding of ligand to one subunit always assists the binding of the same ligand to the next subunit - This means that only positive cooperativity is possible.

Heterotropic interactions could be either positive or negative.

Sequential model



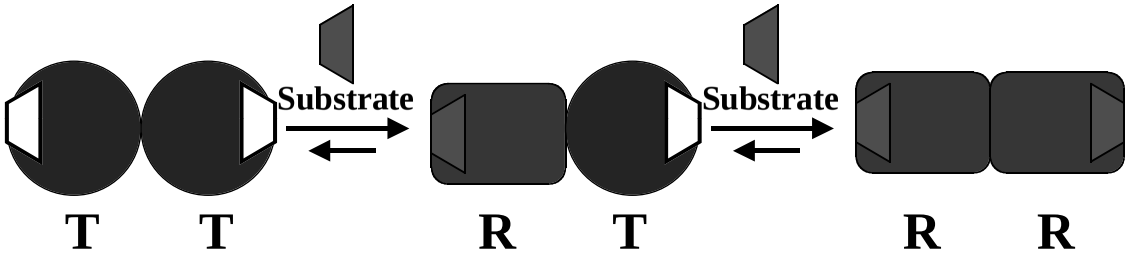
Ligand binding alters the conformation of the enzyme.

Binding of ligand to a subunit alters the conformation of only that subunit.

This alteration is transmitted to other subunits by subunit interaction. Therefore, multiple states are possible.

While going from one state to the other, symmetry need not be conserved.

Sequential model



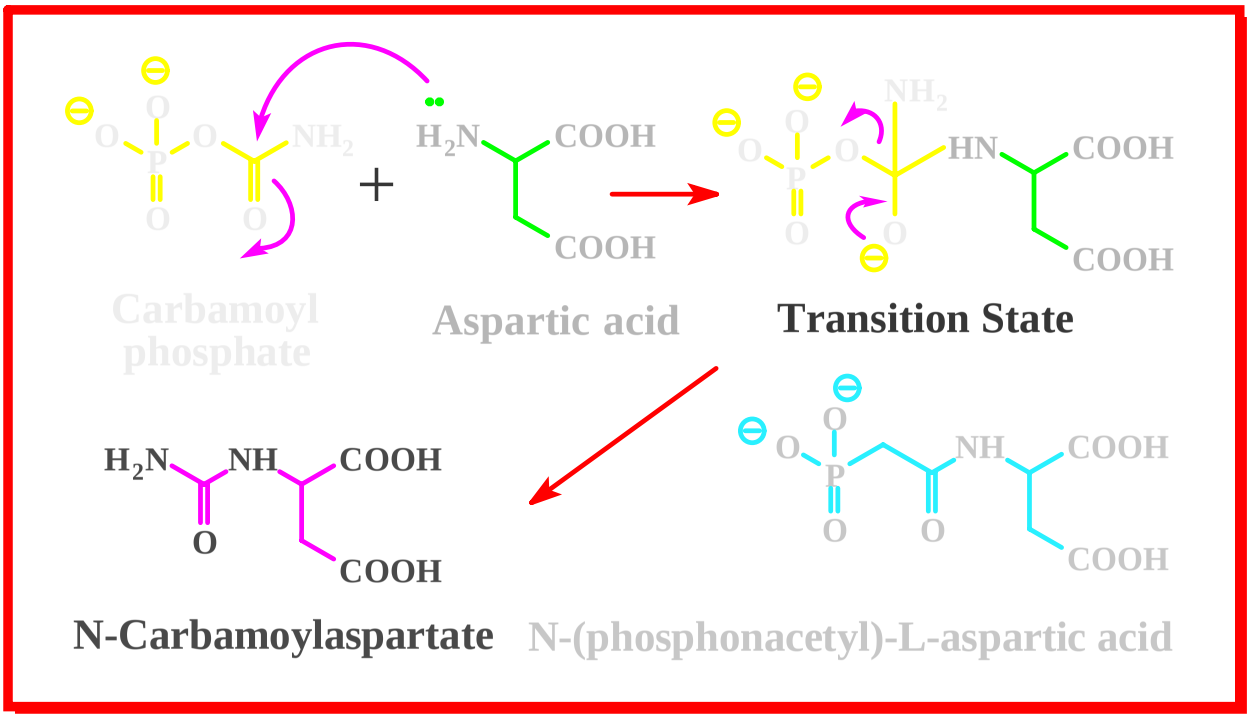
Binding of ligand to one subunit may help or hinder its binding to the other subunit - That is both positive and negative homotropic interaction are possible in this model.

Heterotropic interactions could be either positive or negative.

In the Symmetry model only positive homotropic interaction is allowed. [S binding always helps another S binding] Heterotropic interactions could be positive or negative. [S and I binding is negative heterotropic interaction and S and A binding is positive heterotropic interaction].

In the Sequential model, both homotropic and heterotropic interactions could be positive or negative. [That is even S binding to one subunit could inhibit the S binding to the next subunit (negative homotropic interaction is possible here)].

Aspartate transcarbamoylase



For ATCase, CTP is an allosteric inhibitor. ATP is an allosteric activator.