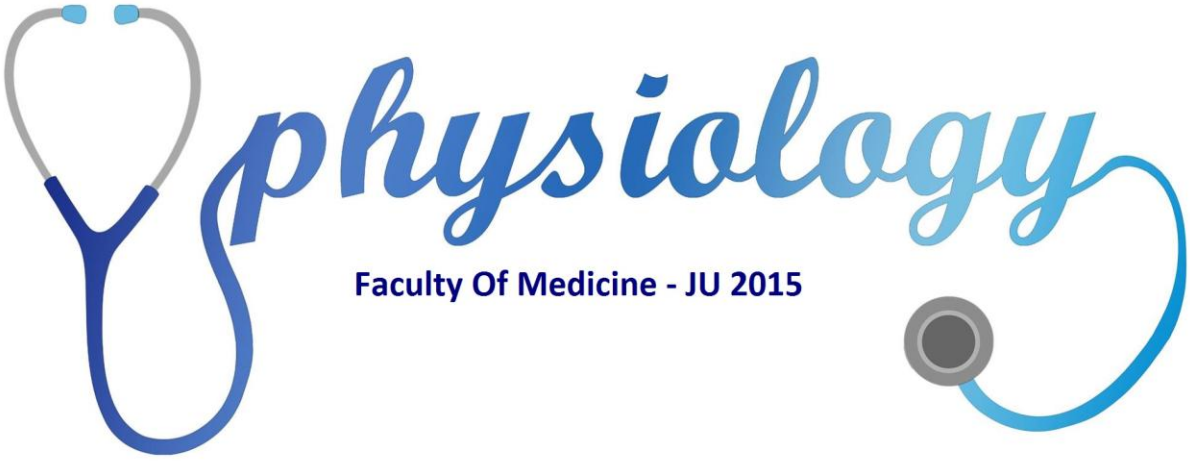
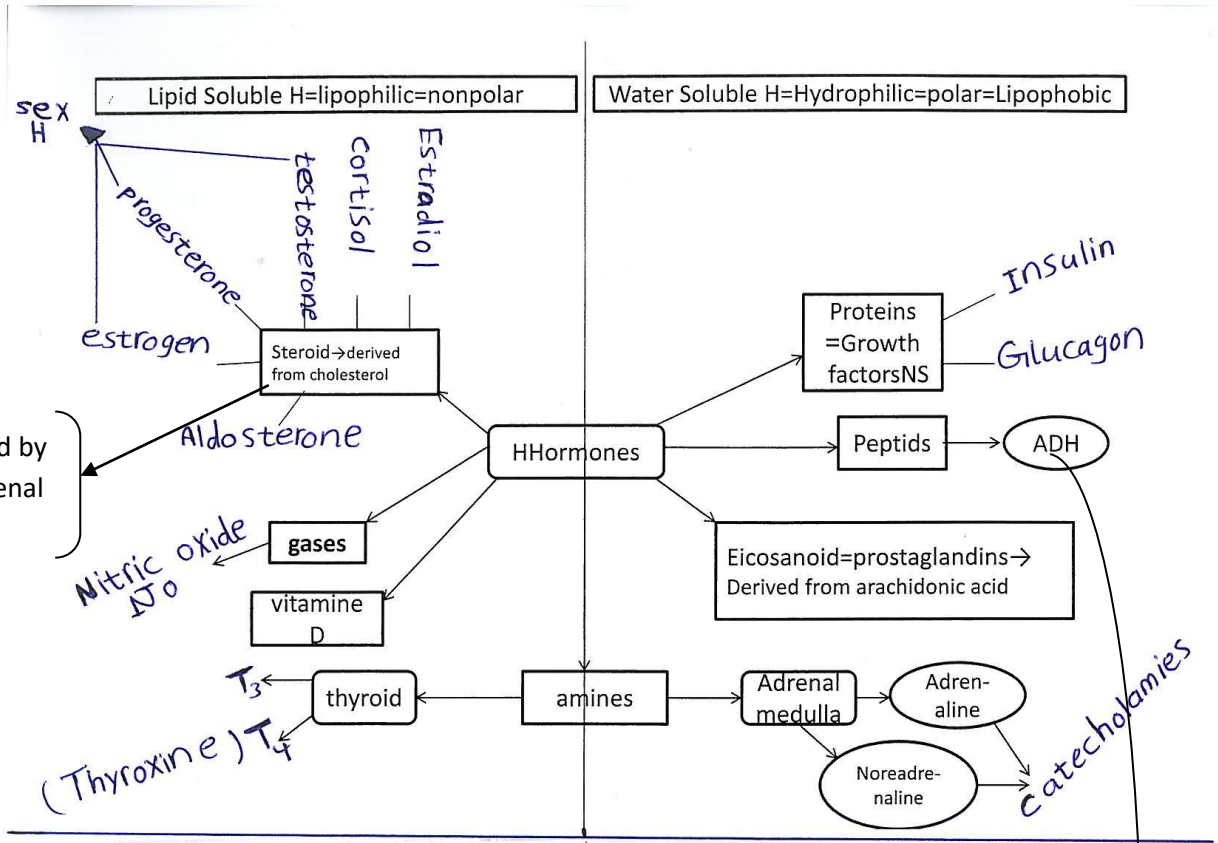


Signal Transduction

Dr. Ebaa's Lectures

Written by: Dawood Alatefi



1. **Its receptors are found inside the cell in the cytosol or in nuclear membrane or in the nucleus because they(lipid soluble H) can pass through the cell membrane easily; so they do NOT act by 2nd messenger.**
2. **Bind to intracellular receptor(same point 1)**
3. **Bound to a protein in the plasma of blood, because if they are not bound and the filtration happened in the kidney, they will be lost very easily with urine.**
4. **Since its receptors are inside the cell, they affect the DNA(change the genetic expression)**

1. **Its receptors are found on the cell surface, so that they act by mean of 2nd messenger.**
2. **Circulate in <u>free</u> form in the plasma of blood.**

- **Peptide: amino acid chain less than 100 amino acid**
- **Protein: amino acid chain more than 100 amino acid**
- **Glycoprotein: long polypeptides(>100) bound to one or more carbohydrate (CHO)groups. e.g: FSH,LH,TSH,hCG. They have α and β subunits(α is common and β is specific)**
- **<u>Exception:</u> Lipophilic hormones may also bind to cell surface receptors, but Lipophobic hormones just bind to cell surface receptors and do NOT bind to intracellular receptors, since they cannot pass through cell membrane actually.**

سؤال من الميد

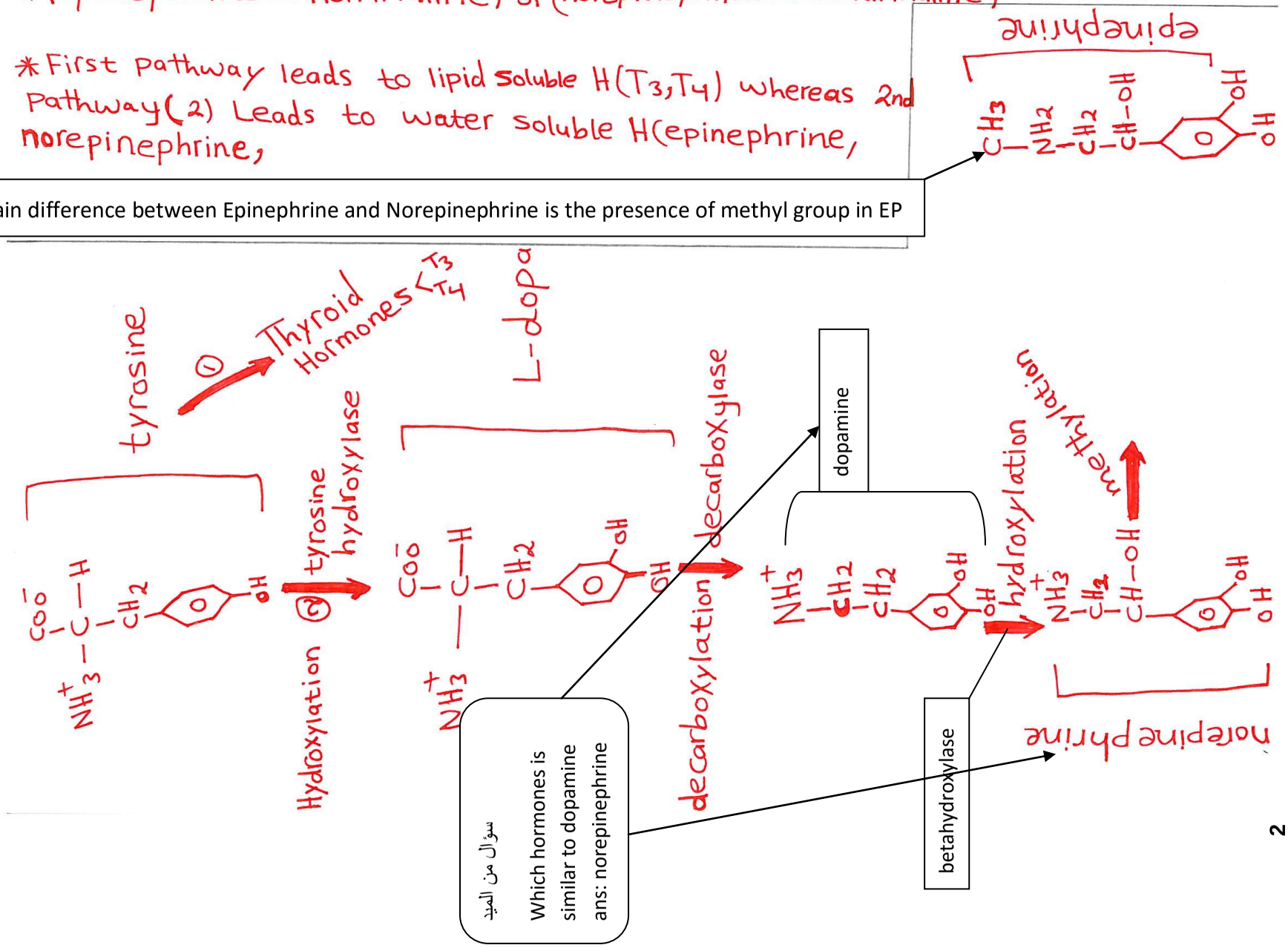
A hormone is mainly unbound to plasma proteins ans: ADH

* Catecholamines = (dopamine + norepinephrine + epinephrine)

*(Epinephrine = Adrinaline) & (norepinephrine = noradrinaline)

*First pathway leads to lipid soluble H(T_3,T_4) whereas 2nd Pathway(2) Leads to water soluble H(epinephrine, norepinephrine,

The main difference between Epinephrine and Norepinephrine is the presence of methyl group in EP



Signal Transduction

- Receptors: we have two types of receptors in our body (1)Receptors for hormones (dr eeba) (2)Receptor for sensation (dr Faisal). And receptor of hormones is our Interest here. From now on, when I say receptors, I mean receptors of hormones. Receptors can be found either on the cell surface or inside the cell depending on the chemical nature of the hormone they bend to. Water soluble hormones (hydrophilic H) can't pass through the cell membrane so that its receptors are found on the cell surface and lipid soluble (lipophilic H) can pass the cell membrane so that their receptors are found inside the cell.
- Messengers: 1) First messenger: which is the hormone (ligand) and it is the substance that reacts and binds to the receptor.2) Second messenger: which is the substance that mediates the action of the first messenger (the hormone). Second messengers which we will meet are(cAMP,cGMP,Ca^{2+}, IP3,DAG, Calcium-calmodulin).

Which hormone is an example of neuroendocrine secretion ans: ADH

- Signaling: Cell-cell communication via signals. Signal transduction: Process of converting extracellular signals into intra-cellular responses. Ligand: The signaling molecule (like hormone).

Types of Ligands

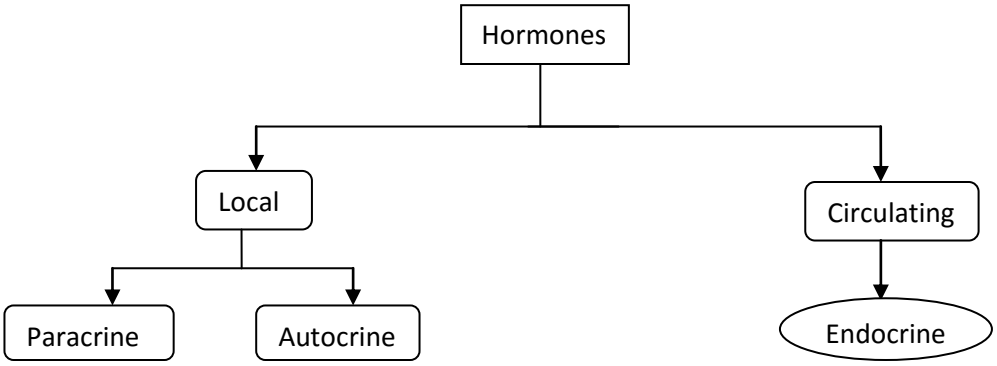
1. **Neurotransmitters**: are released by axon terminals of neurons into the synaptic junctions and act locally to control nerve cell functions. **2**. **Endocrine hormones:** are released by glands or specialized cells into the circulating blood and influence the function of target cells at another location in the body. **3**. **Neuroendocrine:(EX: ADH, oxytocin)** hormones are secreted by neurons into the circulating blood and influence the function of target cells at another location in the body. **4**. **Paracrines**: are secreted by cells into the extracellular fluid and affect neighboring target cells of a different type. **5**. **Autocrines**: are secreted by cells into the extracellular fluid and affect the function of the same cells that produced them. Autocrines are usually for feedback mechanism.**6**. **Cytokines:** are peptides secreted by cells into the extracellular fluid and can function as autocrines, paracrines, or endocrine hormones(e.g., leptin). This hormone is produced by adipocytes.

Types of signaling

Depending on the type of the ligand we can divide types of signaling into:

1) Autocrine: when the signaling molecule is autocrine hormone
2) Paracrine: when the signaling molecule is paracrine hormone
3) Synaptic: when the signaling molecule is neurotransmitter
4) Endocrine: when the signaling molecule is endocrine hormone.

Endocrine	Paracrine	Autocrine	Synaptic
Ligand is secreted into blood	Ligand is secreted into interstitial fluid	Ligand is secreted into interstitial fluid	Ligand is secreted into interstitial fluid
Receptors are on distant target cells	Receptors are on nearby cells	Receptors are on the same cell	Receptors are on nearby cells and action on postsynaptic cell in response to electrical stimuli



Cell signaling starts with ligand secretion.

Now when ligand(hormone) binds to its receptor it should affect something in the receptor which could be **conformational change** of the receptor and this change will be the **first step of transduction** and this change will be translated from the receptor into another protein to propagate the signal into the target that we want. The changes may be very fast or slow. If we need to change the genetic expression, it will take long time. If we changed the function of a protein or membrane potential, it might be very fast.

**ADH→ is synthesized from hypothalamus gland and then transmitted to posterior pituitary gland and then might be secreted according to a stimulus, this is a type of neurocrine signaling which induce water reabsorption in the tubules of the kidney *by increasing their permeability to water*.

- **Synthesis of Amine Hormones** (shown in the appendix 2→pathway is not important)
  One amino acid can be a precursor for the synthesis of several hormones, for example tyrosine. In the presence of tyrosine hydroxylase, tyrosine is converted to L-Dopa. L-Dopa is later converted to Dopamine which is converted to norepinephrine if the enzyme betahydroxylase is present in the cell. (Norepinephrine is a neurotransmitter released from neurons in the sympethatic nervous system). Lastly, norepinephrine is converted to epinephrine in the adrenal gland because it expresses the enzyme that is able to convert NE to EP. Production of these hormones depend on the presence of the specific producing enzymes. Thyroid hormones are synthesized from the same precursor (tyrosine) through a different pathway in the thyroid gland.
- **Chemical classes of hormones**(shown in appendix 1→SO so important.study it well)

  Endocrine hormones are classified into:

- Amino acid derivatives: epinephrine, norepinephrine, and thyroid hormones (T3,T4). These are all derived from the amino acid tyrosine.
- Peptides: antidiuretic hormone (vasopressin) and hypothalamus hormones. The hypothalamus hormones are called releasing factors since they stimulate other endocrine glands to release their hormones.
- Proteins or (glycoproteins): anterior pituitary hormones.
- Steroids: sex hormones, estrogen, progesterone, cortisol and corticosteroids,estradiol.

Paracrine hormones are classified into:

- Amino acid derivative: histamine. Histamine can be a neurotransmitter or a paracrine hormone depending on the secreting cell and the affected cell.
- Arachidonic acid derivatives: prostaglandins (inflammatory mediators).

➢ Prohormones & Prehormones

➢ • Preprohormone: Prohormone derived from larger precursor molecule. EX; Preproinsulin

• Prohormone: Precursor is a longer chained polypeptide that is cut and spliced together to make the hormone. EX:Proinsulin – gives insulin……. Preprohormone→ Prohormone→Hormone

➢ Prehormone: Molecules secreted by endocrine glands that are inactive until changed into hormones by target cells. EX:T4 converted to T3 (tri-iodothyronin)…. Prehormone→Hormone

•Notes: Polypeptide and protein hormones are synthesized in RER and they are stored in Secretory vesicles until needed. Steroid hormones are usually synthesized from cholesterol and are not stored. Amine hormones are derived from tyrosine.

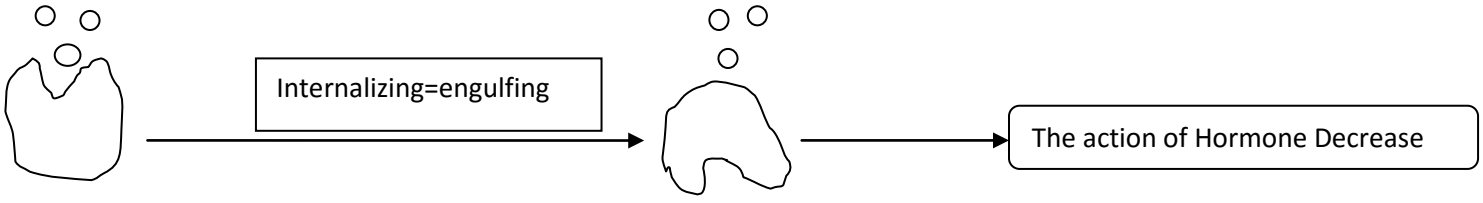
Hormone Activity: Hormones affect only specific target tissue with specific receptors. Receptors are dynamic and constantly synthesized and broken down.

→**Up-regulation** (sensitization): • Increase in receptors number by increase in synthesis or by increase in insertion into the plasma membrane to be exposed to the ligands. Usually occurs to increase response of hormone to the stimulus(bigger effect) • Also called "priming effect"

• Cells become more sensitive to stimulus

*RULE: More stimuli→ stimulates negative feedback in receptor production. Thus, there will be something called Down Regulation.

→**Down Regulation**(Desensitization): • Decrease in number of receptors • Could occur through (1) Inactivation of some receptor molecules(intracellular molecules) (2) Temporary engulfing of the receptor molecules, away from the receptor hormone interaction is



(Receptor is exposed to H) **(Receptor is not exposed to H anymore)**

(3) Destruction of the receptors (4) Decreased production of receptors

• Cells become less sensitive to hormone as a result of→ Long term or continuous stimuli.

• One way to reduce Down Regulation → Having a pulse-like-stimulus- ON and OFF .this will make sure anytime there is a stimulus the number of receptors will not decrease.

- Clinical Correlation: Diabetes II- Insulin Insensitivity • Normally, Insulin stimulates sugar uptake in cells after each meal. In patients with Insulin Insensitivity, Insulin is highly stimulated due to high sugar intake, which will induce down regulation of receptors. Consequently, the cells become desensitized to Insulin and no response to it occurs. Thus, sugar stays in the blood in very high amounts which can be deleterious. • This is prevented by a pulsatile stimulus. • Diabetes I → insulin is not enough

❖ **Hormonal Half Life**: the time that is required to reduce the normal physiological amount of a hormone by half (50% of it).Ex: if the half live of a hormone is 4 hours, then after 8 hours you will find ¼ of it.

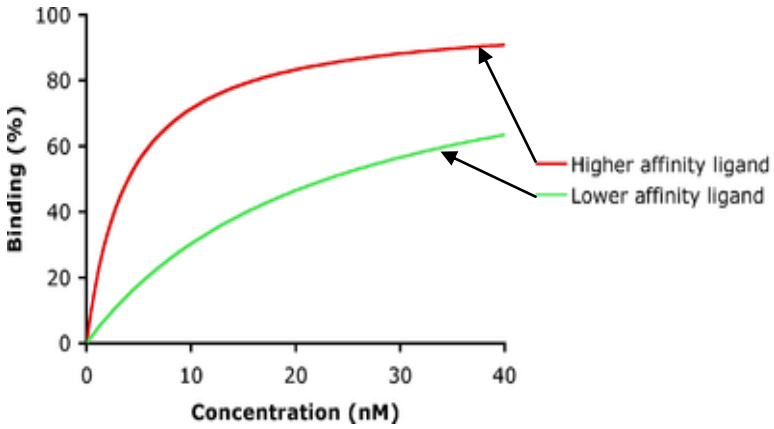
❖ **Affinity of receptors to ligand (judged by K association*Ka* or K dissociation*Kd* or *ΔGo*)**
Affinity describes how strong the hormone binds to its receptors.

*Low Kd →high affinity →low concentration of hormone is needed → Less hormones are unbound→very strong binding→large negative ΔGo

*High Kd →low affinity →more concentration of hormone is needed →more hormones are unbound→weak binding→large positive ΔGo

سؤال من الميد

Something about the affinity of ligand to a receptor except ans: the higher the affinity, the more ligand concentration is needed



- Responsiveness of target cell depends on: Hormone's concentration & Abundance of target cell receptors.
- Hormones exhibit: affinity & saturation (when all the receptors are busy and we have low capacity of receptor).

Hormones of same chemical class have similar mechanisms of action (as shown in appendix 1). These similarities include: •**Location of cellular receptor proteins** depends on the chemical nature of the hormone. •**Events that occur in the target cells**(water soluble hormones have 2nd messenger whereas lipid soluble hormones have no 2nd messenger).

Normal tissue responses are produced only when hormone is present within physiological range. That means that the hormone should be in a physiological reference range that is compatible with its affinity.

- **Specificity :** specificity means that each hormone *must* have specific receptor in the **target cell** ,and it is a description of how favorable the binding of the ligand for the receptor

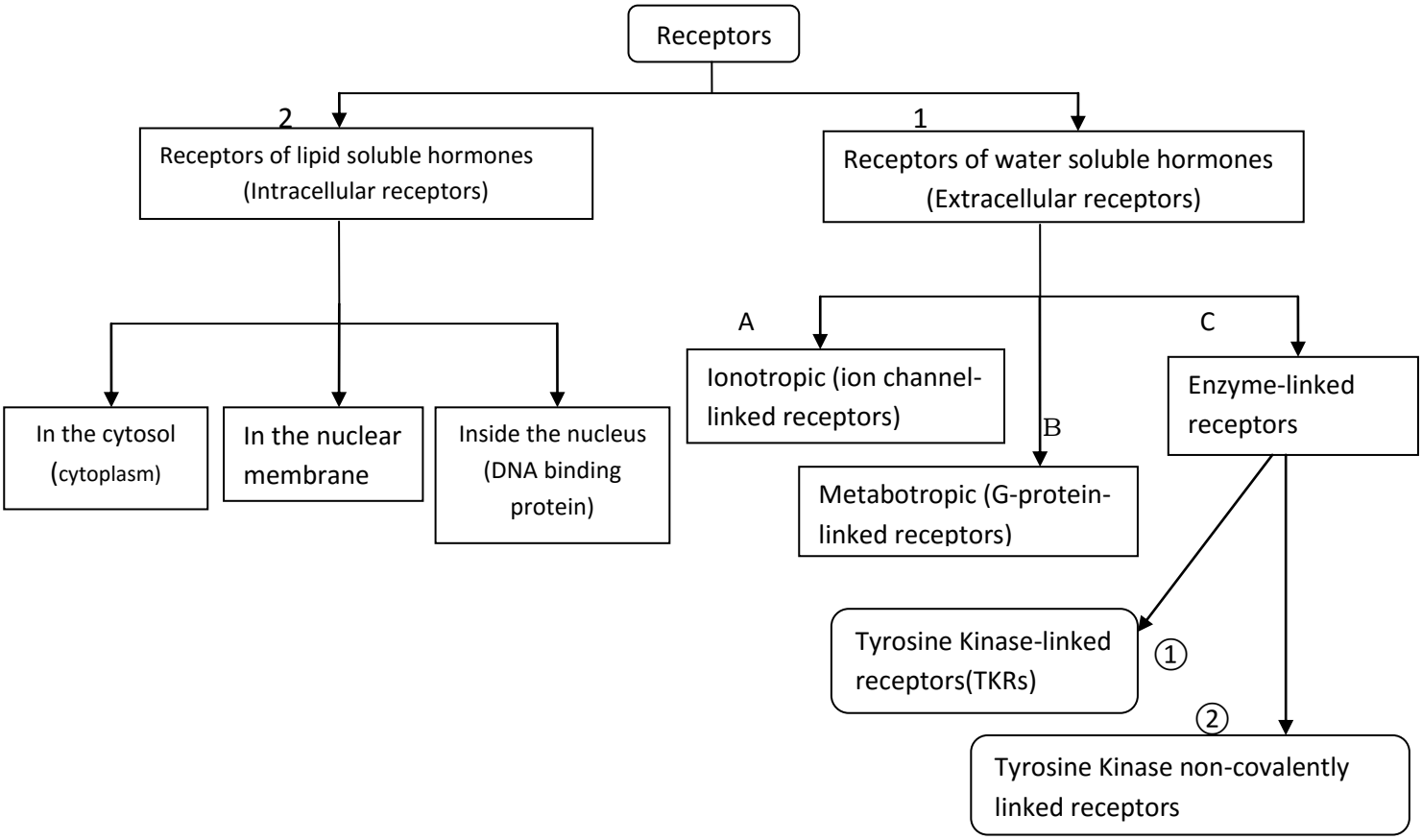
Mechanisms of Hormone Action:

- Response depends on both **hormone** and **target cell**
- Lipid-soluble hormones bind to receptors inside target cells
- Water-soluble hormones bind to receptors on the plasma membrane
  - Activates second messenger system
  - Amplification of original small signal

Hormone receptors:

Receptors are specific membrane proteins, which are able to recognize and bind to corresponding ligand molecules, become activated, and transduce signal to next signaling molecules. Usually they are Glycoproteins or Lipoproteins.

- Bind specific ligands.
- Transmit signals to intracellular targets.
- Different receptors can respond differently to the same ligand.
- Signals get translated into cellular responses or changes in gene expression
- Responses can be fast or slow



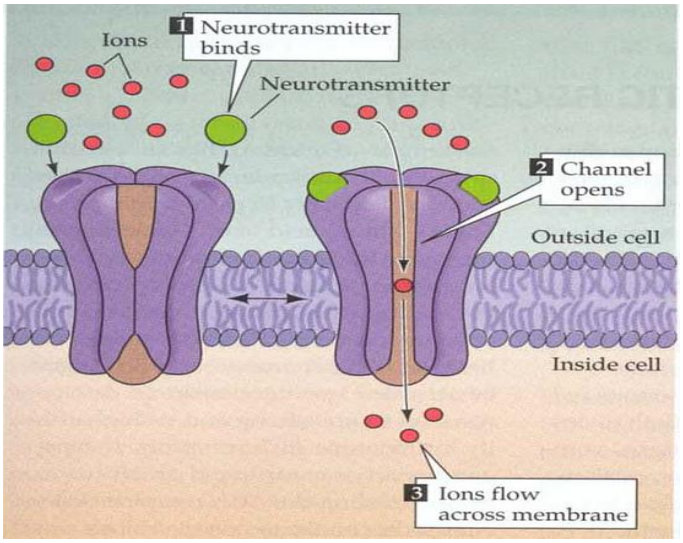
Now we are going to discus each of these receptors in details and we will see the mechanism for each which by they work by introducing examples for each with explaining different2nd messengers of G-protein-linked receptors . So let's start

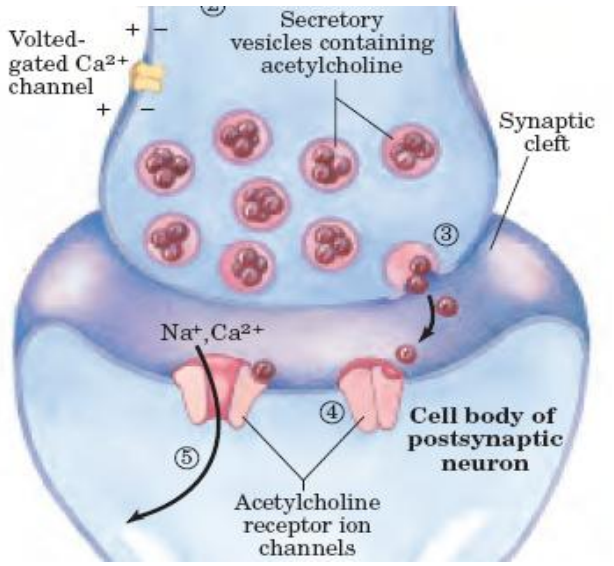
1)<u>Extracellular receptors</u>(membrane receptors)

<u>A) Ion channel-linked receptors</u>(ionotropic receptors)

Ligand → receptor →ion channel open or close

Let's take Acetylcholine as an example for ligand of ion channel-linked receptor. Ach receptors are on the post-synaptic membrane. When Ach bind to the channel, it induce depolarization. Na^+ flows in and the action potential is transmitted.

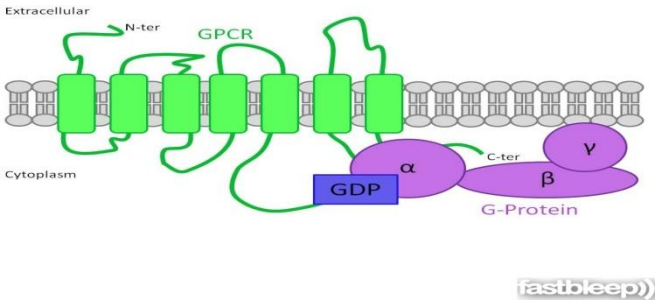




B) G-protein-linked receptors(Metabotropic receptors)→So So important

largest family of cell surface receptors. First we will discuss the structure of these receptors and structure of G-Protein they are bound to.

G-Protein-linked receptors are seven transmembrane segments that loop in and out of the cell membrane. Some parts of the receptor that protrude into the cell cytoplasm (especially the cytoplasmic tail of the receptor) are coupled to G proteins that include three (i.e., trimeric) parts—the α, β, and γ subunits.



When the ligand (hormone) binds to the extracellular part of the receptor, a conformational change occurs in the receptor that activates the G proteins and induces intracellular signals that either (1) open or close cell membrane ion channels, (2) change the activity of an enzyme in the cytoplasm of the cell, or (3) activate gene transcription. In the inactive state of the G-protein, the α, β, and γ subunits of G proteins form a complex that binds guanosine diphosphate (GDP) on the α subunit. When the receptor is activated, it undergoes a conformational change that causes the GDP-bound trimeric G-protein to associate with the cytoplasmic part of the receptor (the part of the receptor that protrude into the cell cytoplasm) and to exchange GDP for GTP. Displacement of GDP by GTP causes the α subunit to dissociate from the trimeric complex(we get two subunits, α subunit and βγ subunit) and to associate with other intracellular signaling proteins; these proteins, in turn, alter the activity of ion channels or intracellular enzymes such as adenylyl cyclase or phospholipase C, which alter cell function.

Let's now see how exchange between GDP & GTP in the G-Protein occurs. First as you know that G-Proteins are named so because that they are GTP-binding proteins. So G-proteins=GTP-binding proteins. So how exchange between GDP and GTP in G-proteins occurs? We have something called GEF and something called GAP. These two are the responsible for exchange between GDP & GTP in the G-

protein. GEF is responsible for giving the G-protein GTP and removing of GDP (it make sense since GEF→Give GTP to G-Protein and make it active so it is generous). GAP is responsible for the opposite action of GEF, it hydrolysis GTP to GDP). So when GAP hydrolysis GTP into GDP this means that GAP has GTPase activity which is the ability to hydrolysis GTP into GDP. One additional information you should know which is that activated receptor(GPCR) normally serves as GEF, but what does serve as GAP???

Its α subunit itself which have GTPase activity. So when the active α subunit

Hydrolysis GTP into GDP it become inactive and now the inactive α subunit bind

the βγ subunit and the cycle is terminated. So we have talked about structure

of G-Protein-coupled receptors(GPCR) and the structure of G-Proteins then we

discussed what happens when the ligand bind to these receptors then we

saw how these G-proteins get activated or inactivated and the cycle they undergo, then we knew GEF and GAP and now we will discuss some example of these G-Protein-coupled receptors

-Rodhopsin: is a light sensitive G-Protein-coupled receptor in the eye retin(does not need a ligand, it is stimulated by light). it was the first of these receptors to be known by its 7-helix structure confirmed by X-ray crystallography. It is responsible for low light vision. Other examples for GPCR : β-adrenergic receptor that is activated by epinephrine and norepinephrine, opioid receptor, glucagon receptor. G-protein coupled receptors mechanism is important in pharmacology, some drugs are agonists (stimulate the receptor's activity and reinforce it), other drugs are antagonists (inhibit receptor's activity).

As we mentioned early that G-Proten-linked receptors act by activation of second messenger system, so before we start with G-protein-linked receptors I'm going to Paves the way by introducing this table containing different 2^{nd} messengers pathways we will meet. Remember: a second messenger is any molecule that transfers a signal from the plasma membrane to the cytosol

Hormone	Receptor	G-protein	Effecter Molecule	Action	2^{nd} messenger/s included
Norepinephrine Or Epinephrine	Adrenergic α1	Gq	Phospholipase C (stimulates)	$PIP_2 \rightarrow IP_3$+DAG	IP_3+DAG
	Adrenergic α2	Gi	Adenylyl Cyclase (inhibits)	ATP $\not\rightarrow$ PPi+cAMP	cAMP
	Adrenergic β1	Gs	Adenylyl Cyclase (stimulates)	ATP $\rightarrow$ PPi+cAMP	cAMP
	…….	…….	Guanylyl Cyclase	GTP $\rightarrow$ PPi+cGMP	cGMP

-**Gs** stands for **stimulatory G-Protein,** and **Gi** stand for **inhibitory G-Protein.**

Adenylyl Cyclase is <u>activated by Gs</u> and <u>turned of by Gi</u>

a. protein kinase A: is activated by cAMP.
b Calmodulin-dependent protein kinase.: is activated by calcium-calmodulin. → SO important
c. protein kinase C: is activated by DAG(Diacylglycerol) and calcium .

Note : Protein Kinase A = (cAMP-dependent protein kinase) & CAM Kinase = (Calmodulin-dependent protein kinase)

we will explain each pathway, but before that we will discuss the general Mechanism of activation of a G-protein–coupled receptor.

When the hormone activates the receptor, the inactive α, β, and γ G-protein complex associates with the receptor and is activated, with an exchange of guanosine triphosphate (GTP) for guanosine diphosphate(GDP). This process causes the α subunit (to which the GTP is bound) to dissociate from the β and γ subunits of the G-protein and to interact with membrane-bound target proteins (enzymes) that initiate intracellular signals.

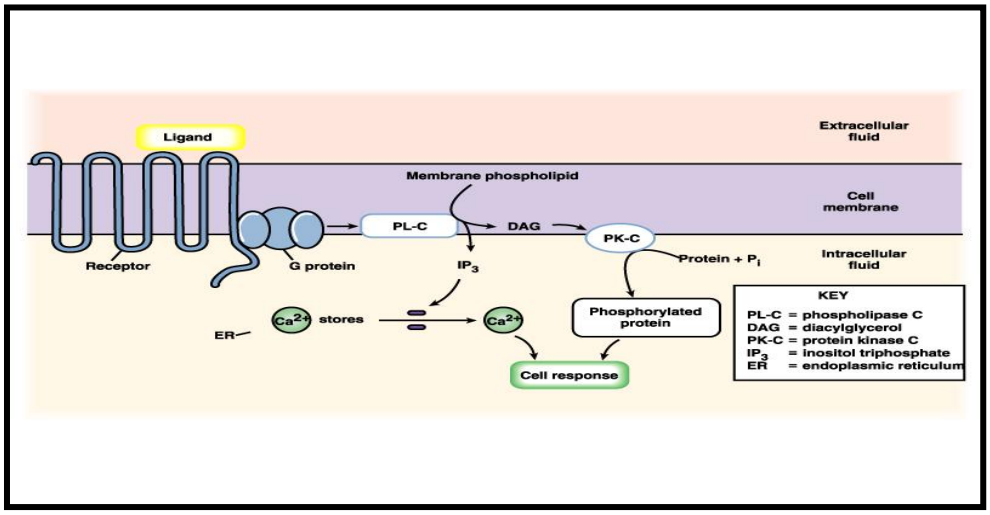
So let's now explain each pathway individually. BUT keep in your mind that the mechanism of activation of G-protein–coupled receptor is the same for all pathways:

- **(1) First pathway**

-Binding of Epinephrine to adrenergic α1 receptor in plasma membrane activates Gq (G-protein) and Gq activates phospholipase C (enzyme).
- This enzyme catalyzes the breakdown of phosphatidyinositol biphosphate(PIP2) into two different second messengers:

a. inositol triphosphate (IP3).
b. diacylglycerol (DAG).

- IP3 diffuses through cytoplasm to endoplasmic reticulum (ER), and binding of IP3 to receptor protein in ER causes Ca++ channels to open.
- Ca++ diffuses into cytoplasm,and Ca++ binds to calmodulin, then calmodulin activates CAM kinase(Calmodulin-dependent protein kinase) which alters the metabolism of the cell, producing the hormone's effects (cellular response).
Note:calcium is a very important trigger for vesicle formation and exocytosis.
This means that anything that requires



secretion of vesicles for neurotransmitters or hormones it can use this signaling pathway we talked about in order to get calcium

Note: DAG, the other second messenger, activates the enzyme protein kinase C (PKC), which then phosphorylates a large number of proteins, leading to cell's response.
Calcium itself is a second messenger so let's talk about it as second messenger. Calcium binds to a certain calcium binding protein called calmodulin
When sudden increase of calcium in the cytosol happens calmodulin will be activated and now can bind to different types of proteins.
One of these proteins is called calmodulin dependent kinase (ca-M-kinase), once this kinase is activated, it autophosphorylates itself and becomes fully active and can activate or deactivate many other proteins.
Note: whenever you see the word kinase always does phosphorylation
-calmodulin can also bind to adenylate cyclase or phosphodiesterase to activate them, so calmodulins target isn't just ca-M-kinase. So calcium can be a second messenger to multiple kinase systems.
- adenylate cyclase can also be activated by ca-M-kinase

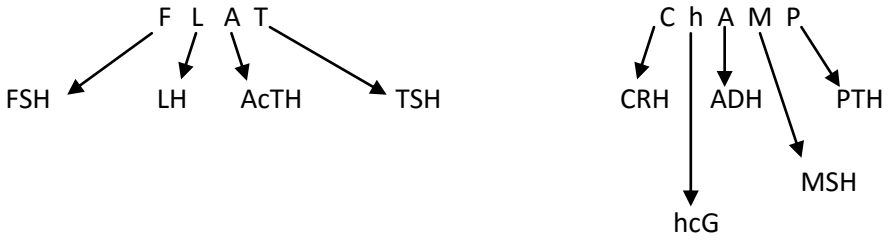
- **(2) Second pathway**

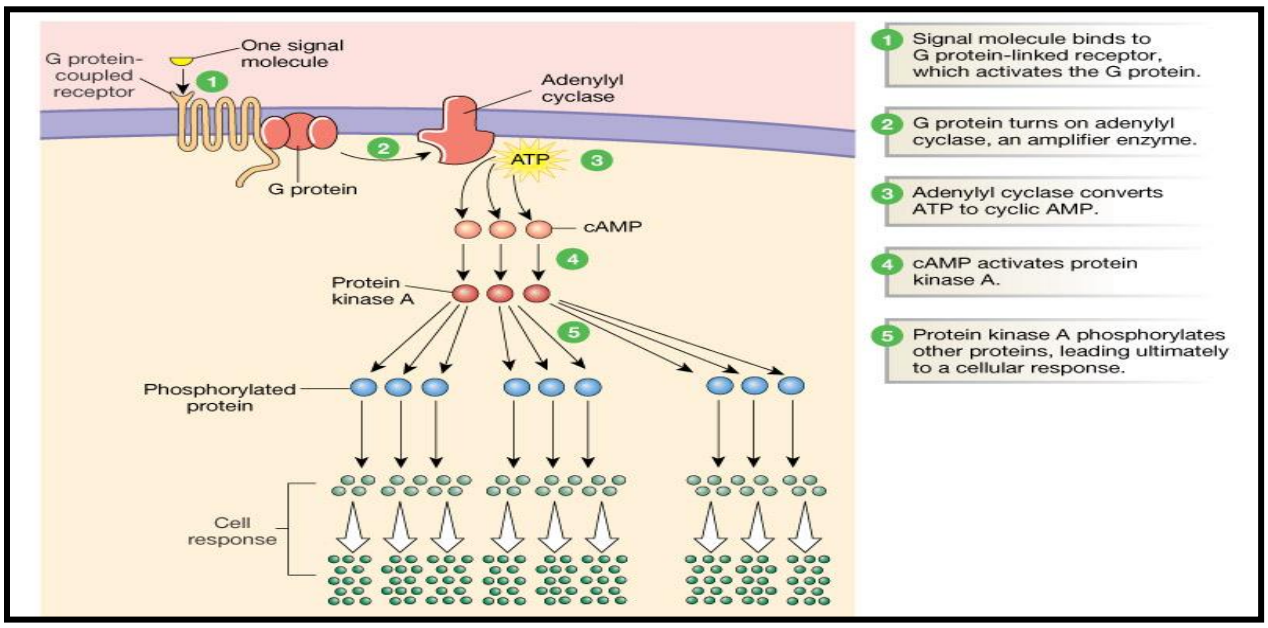
The second pathway is like the third pathway(we will discuss it below), but instead of activation of Adenylyl Cyclase, the Gi will inhibit it and the pathway is over. cAMP level will be decreased

- **(3) Third pathway**

-Binding of Epinephrine to adrenergic β1 receptor in plasma membrane activates Gs (stimulatory G-protein) and Gs activates Adenylyl Cyclase (enzyme).Once Adenylyl Cyclase get activated, it will convert ATP into (PPi) and second messenger which is (cAMP). This 2nd messenger will activate protein kinase A(PKA).
Protein kinase A(cAMP-dependent protein kinase)phosphorylates other proteins, leading ultimately to a cellular response.

<u>Hormones That Use the Adenylyl Cyclase–cAMP Second Messenger System:</u>(FLAT ChAMP)





Now let's see the structure of protein kinase A and the mechanism by which PKA get activated by cAMP.

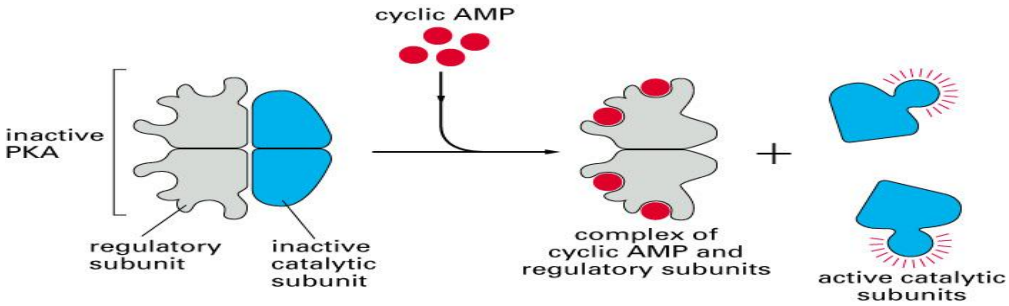


Figure 15–32. Molecular Biology of the Cell, 4th Edition.

cAMP binds to protein kinase A. PkA is compsed to 2 parts :(a). two units are regulatory (b). two units are catalytic ** When these two parts are bound to each other, PKA is inactive. When cAMP binds to the regulatory part of PKA, these 2 parts separate. PKA is now active and is works to phosphorylate. Kinases either phosphorylate the amino acid serine or threonine, or both of them together. Why serine and threonine specifically? They have hydroxyl groups and these are the parts that are phosphorylated. After being phosphorylated, proteins either become active or inactive. Just as we have Kinase, we have phosphatase action.

<u>Note: It's important to know that phosphorylation doesn't mean activation but it means changing (may be activation or deactivation)</u>

<u>Remember;</u> most effector enzymes controlled by G-Protein are involved in synthesis of second messenger. And remember that the 1st messenger is the

ligand(Hormone), and 2nd messenger is a molecule that transfer the signal to the cytosol.

Now that the signal caused a response, we have to find a **<u>way to turn off the signal</u>** because everything in the body needs to be balanced. We cannot depend on the hormonal life spane, so we need to act on every step along the pathway:

→1**) Inactivate α subunit of the G-Protein** (which is active when bound to GTP). We can have an exchange of GDP for GTP(releasing GTP). This is done by the GAP enzyme. This enzyme has a GTP-ase activity. This GTP-ase activity exists intrinsically in the G-protein α subunit itself. Now there is no free α subunit(they bind back to inhibitory beta and gamma), and adenylate cyclase(adenylyl cyclase)becomes inactive. cAMP production ceases.

→2) **Phosphodiesterase stimulation**: An enzyme that turns cAMP to AMP and thus there is no longer a second messenger. cAMP is what stimulates phosphodiesterase., so, cAMP is what stimulates its degradation. PKA can also phoshorylate Phosphodiesterase and stimulate its activity.
Thus, the more cAMP we have, the higher the rate at which it is degraded, and this is needed to turn off the signal or the message.

→3) **Desensitization(Down regulation)** : •Decreasing sensitivity •PKA can phosphorylate the receptor. The receptor now has a tag. The tag is identified by a regulatory protein called Beta-arrestin. B-arrestin binds to receptors and this induces invagination/calathrin coated endocytosis of the whole associated area. Beta-arrestin activation also stimulates the B-arresin to bind to phosphodiesterase. Phosphodiesterase becomes in close proximity with cAMP and stimulates its breakdown.

→4**) Phosphatase** can also remove the phosphate group added by PKA to cellular proteins.

- Beta-Gamma complex: When bound to the alpha subunit, it deactivates it(inhibitory) . They can bind to certain isoforms of adenylate cyclase and deactivate it.

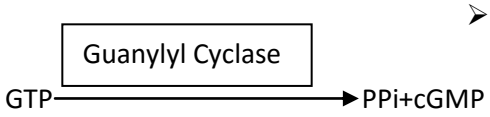
✓ Clinical Correlations:

- Cholera Toxin:
  - Causes ADP ribosylation, a chemical reaction that adds ADP to a molecule covalently
  - This acts on Galpha(activated form)
  - Gs no longer can exchange GDP for GTP (cant release GTP) and it stays permanently activated = Increased cAMP
  - Because it's a covalent bond, it's very hard to break and is irreversable
  - Affected receptors are G-protein linked receptors in the Gastro- Intestinal tract
  - In the GI tract, there are lots of electrolytes in the lumen. When these receptors are active, Cl and many other electrolytes flow out of the GI accompanied by water and dehydration occurs as diarrhea.
- Pertussis Toxin(whooping cough disease):

- Causes Gia(inhibitory) unable to release GDP. GDP remains bound to it, so it won't be ab;le to be activated.
- Thus we have an inactivation of the inhibitory G-protein, so excitation results from absence of the inhibition.
- This produces the same results as the cholera toxin = increased cAMP

→Now we have finished talking about G-Protein-coupled receptor. Remember the main topics we are discussing .We are talking about Extracellular receptors which are three types(A*.Ion-channel-linked receptors.*B*.G-Protein-linked receptors.*C*.Enzyme-linked receptors). We finished talking about G-Protein-linked receptors, so let's now talk about the third type which is Enzyme-linked receptors.

- **(4) forth pathway**
-



C) Enzyme-linked receptors

①Tyrosine Kinase-linked receptors(TKRs)→EX: Insulin receptors , Growth factors receptors(NGF, EGF , PDGF….)

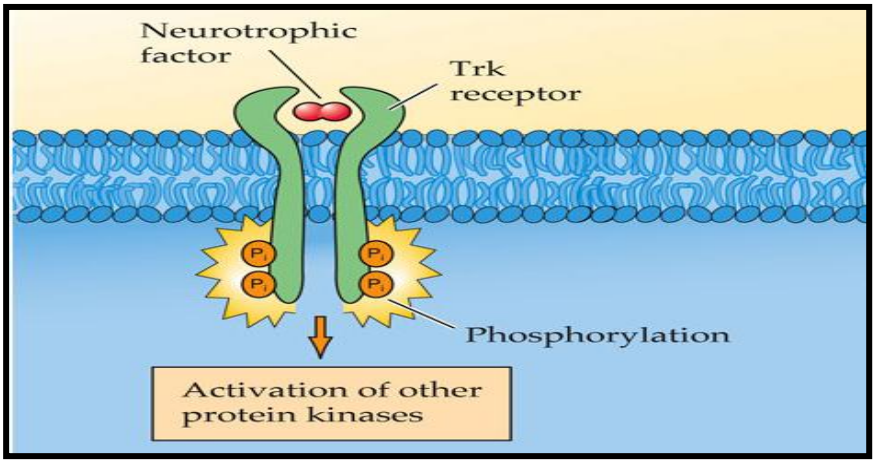
②Tyrosine Kinase non-covalently linked receptors→ Cytokine receptors

The third type of Extracellular receptors is Enzyme-linked receptors. Now the enzyme can be a main part of the receptor as in the case of Tyrosine Kinase(TrK) or it may be attached to it as in the case of Tyrosine Kinase non-covalently linked receptors.

They are cell surface receptors(Extracellular receptors) that are directly linked to intracellular enzymes(Kinase). Examples for this kind include **insulin receptors** (the receptors of insulin hormone) and **Growth factors receptors** that can be found in all body parts in high numbers and have many types like NGF, EGF , PDGF.
As you know that any Enzyme-linked receptor has extracellular domain(part) that is responsible for binding with the ligand(hormone) and intracellular domain that has enzymatic activity(kinase activity). It could be present as a monomer or dimer, in the case of insulin there is a dimerization between two of the transmembrane receptor(this process is very important for activation of kinase)
So do you know actually what kinase activity means? →a kinase is an enzyme that catalyzes the transfer of phosphate groups from high-energy, phosphate-donating molecules to specific substrates.
NOW we have known what does kinase mean. Still we need to know what Tyrosine means or for what does tyrosine refer?

Actually tyrosine is an amino acid (you will see it when you study biochemistry).So kinase causes phosphorylation of substances in the cell (mostly proteins) and that phosphorylation is done on the tyrosine residues. Now keep in your mind that kinase activity is the ability to do phosphorylation. But as we said previously that phosphorylation does not necessarily mean activation. It could be activation or inactivation.

Let's now discuss the general mechanism of **Tyrosine Kinase-linked receptors(TKRs)** then we will discuss the mechanism of insulin receptors as an example for this group.



When a hormone binds to tyrosine kinase receptor(if it is insulin receptor, there will be dimerization) it activates tyrosine kinase , which itself **is a part of this receptor,** and tyrosine kinase is **autophosphorylated** then it phosphorylates the intracellular proteins, and finally we get cell response.

Q: what is the difference between tyrosine kinase and other protein kinase?

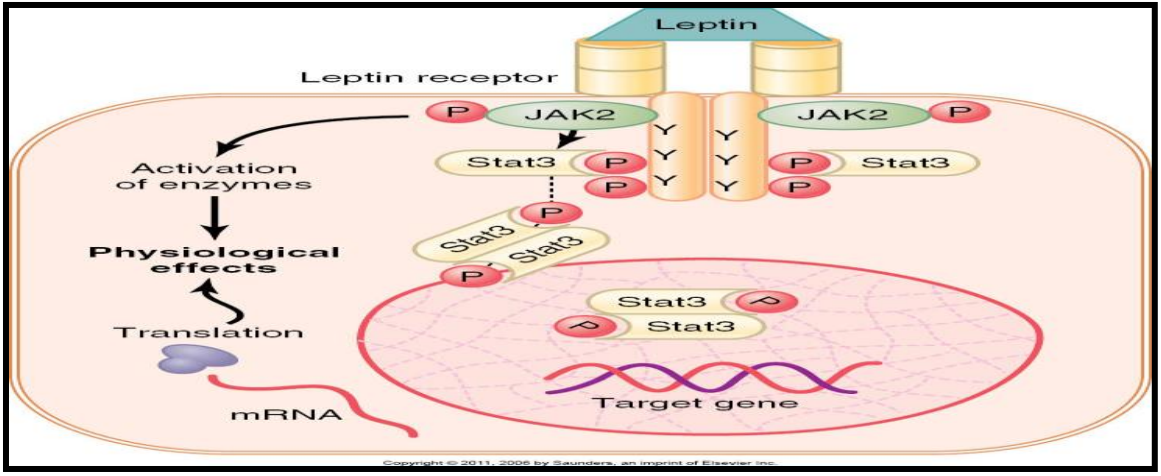
→Tyrosine kinase is attached to its receptor which is found in the cell membrane, but other protein kinase are found inside the cell and they are attached to their receptors non-covalently. Other difference is that tyrosine kinase is **autophosphorylated** whereas other protein kinase need second messenger to get them active.

Now we are going to introduce an example for this is kind of receptors which the leptin receptors

leptin receptors:

Leptin is a hormone secreted by fat cells and has many physiological effects, but it is especially important in regulating appetite and energy balance. The leptin receptor is a member of a large family of cytokine receptors that do not themselves contain enzymatic activity but signal through associated enzymes. In the case of the leptin receptor, one of the signaling pathways occurs through a tyrosine kinase of the janus kinase (JAK) family, JAK2. The leptin receptor exists as a dimer (i.e., in two parts), and binding of leptin to the extracellular part of the receptor alters its conformation, enabling phosphorylation and activation of the intracellular associated JAK2 molecules. The activated JAK2 molecules then phosphorylate other tyrosine residues within the leptin receptor–JAK2 complex to mediate intracellular signaling. The intracellular signals include phosphorylation of signal transducer and activator of transcription (STAT) proteins, which activates transcription by leptin target genes to initiate protein synthesis. Phosphorylation of JAK2 also leads to activation of other intracellular enzyme pathways such as mitogen-activated protein kinases (MAPK) and phosphatidylinositol 3-kinase (PI3K). Some of the effects of leptin occur rapidly as a result of activation of these intracellular enzymes, whereas other actions occur more slowly and require synthesis of new proteins.

Now we have finished talking about Extracellular receptors, so let's now talk about Intracellular receptors or Receptors of lipid soluble hormones.



2).Intracellular receptors

As you know that all the left side hormones mentioned in first appendix(lipid soluble hormones) all of them have its receptors inside the cell .steroid hormones and thyroid hormones(T3,T4) are lipid soluble hormones which means that its receptors are intracellular receptors. So we discuss the mechanism of action of these two hormones.

Note: lipid soluble hormones need transporter in the blood to travel to the target cell, and this transporter is globulin which is produced in the liver.

Because these receptors are lipid soluble hormones that means that they readily cross the cell membrane and interact with receptors in the cytoplasm or nucleus. The activated hormonereceptor complex then bind with a specific regulatory(promoter) sequence of the DNA called the hormone response element, and in this manner either activates or represses transcription of specific genes and formation of messenger RNA(mRNA). Therefore, minutes, hours, or even days after the hormone has entered the cell, newly formed proteins appear in the cell and become the controllers of new or altered cellular functions. Many different tissues have identical intracellular hormone receptors, but the genes that the receptors regulate are different in the various tissues. An intracellular receptor can activate a gene response only if the appropriate combination of gene regulatory proteins is present, and many of these regulatory proteins are tissue specific. Thus, the responses of different tissues to a hormone are determined not only by the specificity of the receptors but also by the expression of genes that the receptor regulates.

Mechanisms of steroid hormone action:

1.the steroid hormone diffuses across the cell membrane and enters the cytoplasm of the cell, where it binds with a specific receptor protein.

2.the combined receptor protein-hormone then diffuses into or is transported into the nucleus.

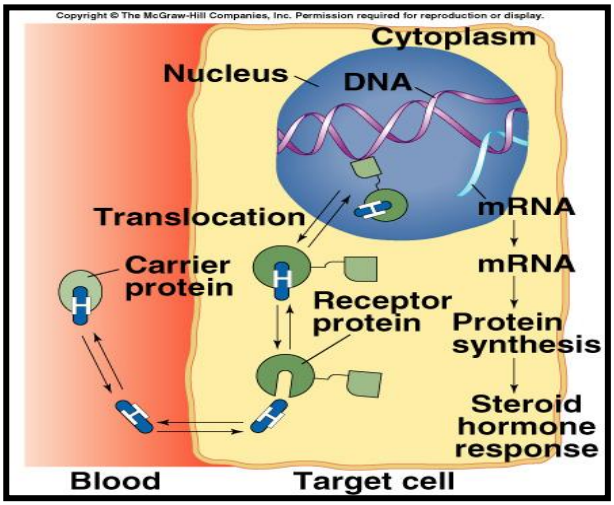
3.the combination binds at specific points on the DNA strands in chromosomes (Hormone response element=HRE).

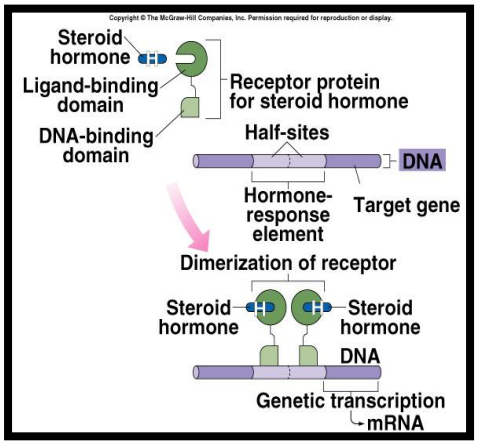
4.Dimerization occurs(process of 2 receptor-units coming together at the 2 half-sites)

Note: in this case, the two ligand-binding domains are binding with steroid hormones

5.this binding activates the transcription process of specific genes to form mRNA.

6.the mRNA diffuses into the cytoplasm, where it promotes the translation process at the ribosomes to form new proteins

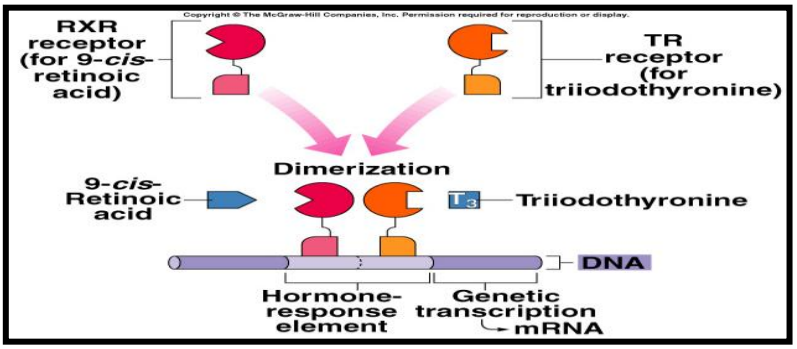




Mechanism of Thyroid hormone action:

1. T4 passes into cytoplasm and is converted to T3(receptors are located in nucleus).
2. T3 binds to ligand-binding domain and the other half-site is vitamine A derivative(9-cis-retinoic) acid.
3.DNA-binding domain can then bind to the half-site of the HRE(two partners can bind to DNA to activate HRE).
4. stimulating transcription of genes.

Note: the transcription takes place at axons of the DNA strand.



Actions of Thyroid hormones:

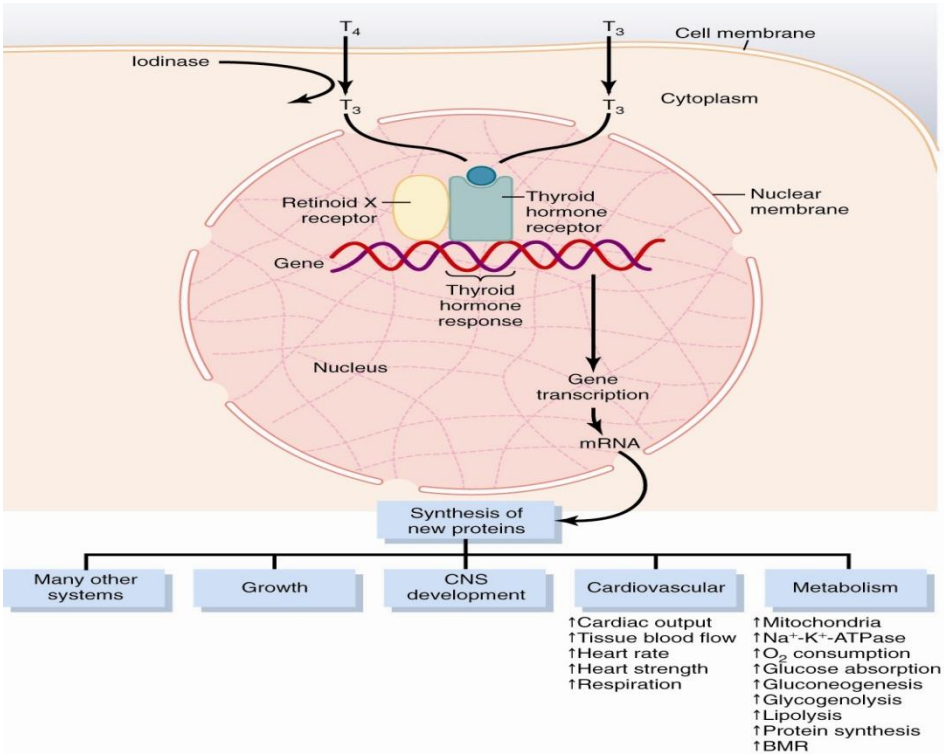
-synthesis of new proteins:

1. many other systems
2. Growth:
3.central nervous system development(CNS):
4.cardiovascular:

A .high cardiac output.
B .high tissue blood flow
c. high Heart rate
d. high Heart strength
e. high respiration.

5.Metabolism: increase basal metabolic rate

a.High glucose absorption.
b.high protein synthesis.



Study the following images:

Image 14:

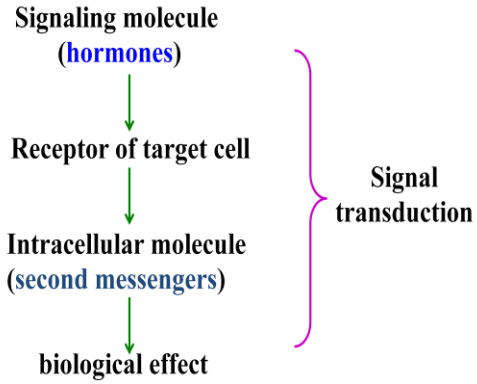
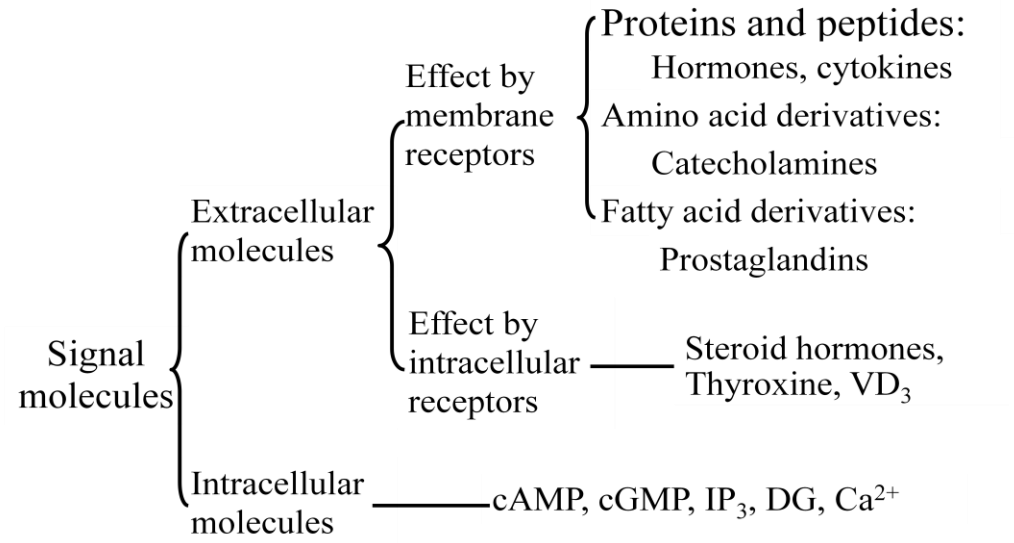


Image 15:

Third messengers:

Third messengers are the molecules which transmit message from outside to inside of nucleous or from inside to outside of nucleous, also called DNA binding protein.

Image 16:



- Vitamins D, E, A, and K are all lipid soluble.
- Prostaglandins are ecosanoids. They are a derivative of arachidonic acid that has 4 double bonds and they are water soluble hormones .

Questions:

1. When a cell releases a signal molecule into the environment and a number of cells in the immediate vicinity respond, this type of signaling is

A. Autocrine signaling.(*on the same cell)*

B. Endocrine signaling.*(on a distant cell)*

C. Paracrine signaling.

D. Synaptic signaling.

E. Typical of hormones.

2. Phosphorylation cascades involving a series of protein kinases are useful for cellular signal transduction because

A. They are species specific.

B. They always lead to the same cellular response.

C. They amplify the original signal manyfold.

D. They counter the harmful effects of phosphatases.

E. The number of molecules used is small and fixed.

3. Under which of the following situations would receptor down-regulation most likely occur?(down-regulation means that the receptors decrease)

A. The concentration of a neurotransmitter is too low.

B. The concentration of a hormone is too high.

C. The number of receptors in the plasma membrane is too low.

D. The number of G proteins is too high.

E. The cell is unable to manufacture cyclic AMP.

4. When a receptor binds to its G protein, which of the following happens next?

A. The signaling molecule binds to the receptor.

B. The G protein activates an enzyme.

C. The three G protein subunits come together.

D. GTP is replaced by GDP.

E. GDP is replaced by GTP.

5. Consider this pathway: epinephrine → G–protein–linked receptor → G protein → adenylyl cyclase → cAMP. Identify the second messenger.

A. cAMP

B. G protein

C. GTP

D. adenylyl cyclase

E. G–protein–linked receptor

Special thanks for:

Renad Al-Awamleh