بسم الله الرحمن الرحيم

Synthesis of Neurotransmitters

MID | Lecture 2

﴿إِنِّي تَوَكَّلْتُ عَلَى اللَّهِ رَبِّي وَرَبِّكُمْ ۚ مَّا مِن دَابَّةٍ إِلَّا هُوَ آخِذٌ بِنَاصِيَتِهَا ۚ إِنَّ رَبِّي عَلَىٰ صِرَاطٍ مُّسْتَقِيمٍ﴾

Written by: Nour Elzogheir
Alhsna' Alhusban

Reviewed by: Sara Masadeh

BIOCHEMISTRY

رحلة اليقين مع سورة يس

بِسْمِ ٱللَّهِ ٱلرَّحْمَٰنِ ٱلرَّحِيمِ

إِنِّيٓ ءَامَنتُ بِرَبِّكُمْ فَٱسْمَعُونِ (٢٥) قِيلَ ٱدْخُلِ ٱلْجَنَّةَ ۖ قَالَ يَٰلَيْتَ قَوْمِي يَعْلَمُونَ (٢٦) بِمَا غَفَرَ لِي رَبِّي وَجَعَلَنِي مِنَ ٱلْمُكْرَمِينَ (٢٧)

{إِنِّيٓ ءَامَنتُ بِرَبِّكُمْ فَٱسْمَعُونِ} أأعبد من دون الله آلهة أخرى لا تملك من الأمر شيئًا، إن يردني الرحمن بسوء فهذه الآلهة لا تملك دفع ذلك ولا منعه، ولا تستطيع إنقاذي مما أنا فيه؟ إني إن فعلت ذلك لفي خطأ واضح ظاهر. إني آمنت بربكم فاستمعوا إلى ما قُلْته لكم، وأطيعوني بالإيمان. فلما قال ذلك وثب إليه قومه وقتلوه، فأدخله الله الجنة.

فـ { قِيلَ } له في الحال: { ادْخُلِ الْجَنَّةَ } فقال مخبرا بما وصل إليه من الكرامة على توحيده وإخلاصه، وناصحا لقومه بعد وفاته، كما نصح لهم في حياته: { يَا لَيْتَ قَوْمِي يَعْلَمُونَ بِمَا غَفَرَ لِي رَبِّي } أي: بأي: شيء غفر لي، فأزال عني أنواع العقوبات، { وَجَعَلَنِي مِنَ الْمُكْرَمِينَ } بأنواع المثوبات والمسرات، أي: لو وصل علم ذلك إلى قلوبهم، لم يقيموا على شركهم.

Biochemistry of neurotransmitters

Prof. Mamoun Ahram
Neuroscience

Resources

- This lecture
- Mark's Basic Medical Biochemistry, 6th ed, pp. 1027-1037
- http://what-when-how.com/neuroscience/neurotransmitters-the-neuron-part-1/

Definition and characteristics of a neurotransmitter

The functional and anatomical **criteria** needed for a chemical to be **classified as a neurotransmitter**

- **A chemical substance that:**
  - is synthesized and stored in a presynaptic neuron (the enzymes needed for its synthesis must be present in the neuron).
  - is released at a synapse following depolarization of the nerve terminal (usually dependent on an influx of calcium ions).

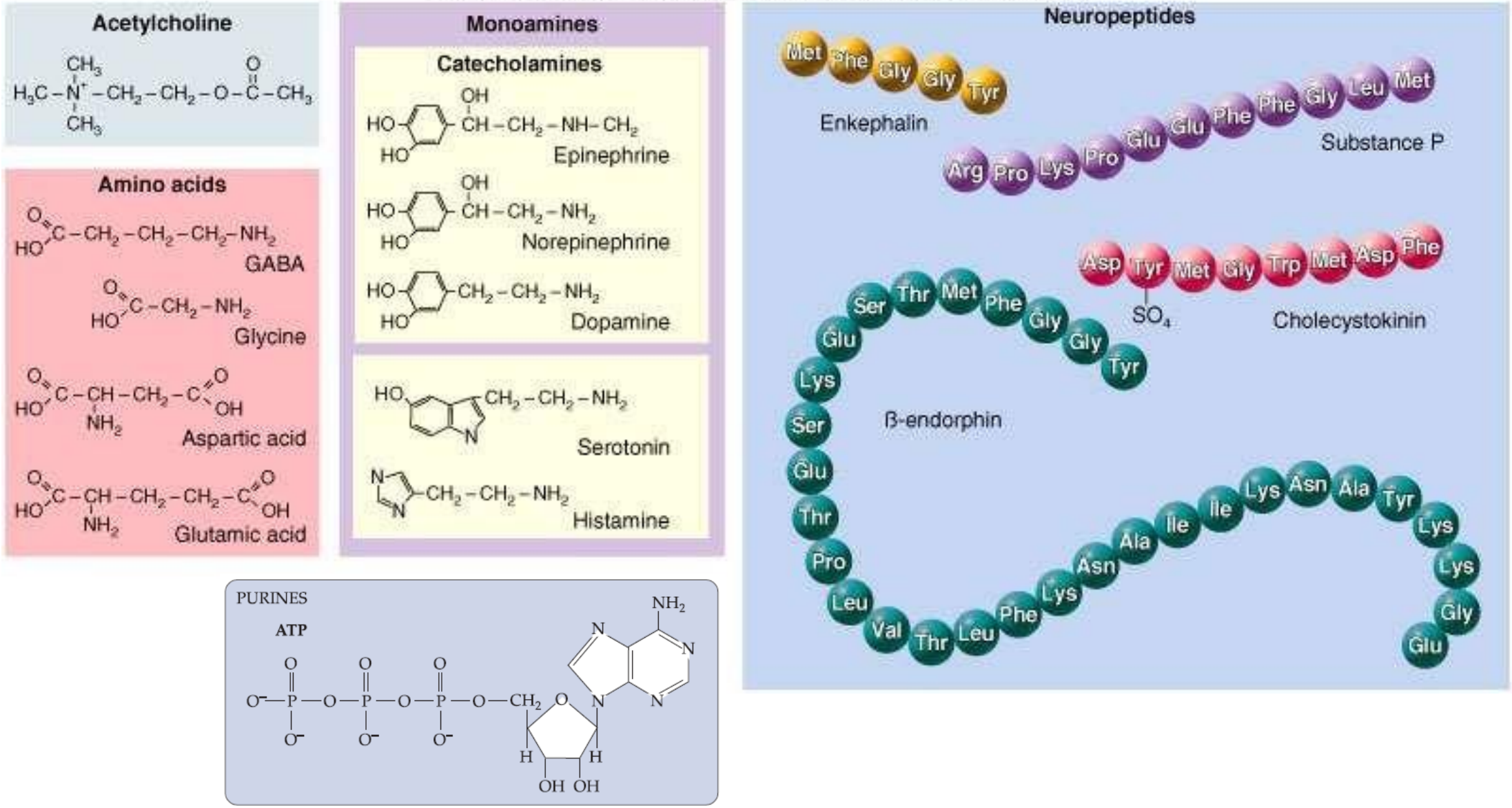
Its concentration will increase intracellularly

  - binds to receptors on the postsynaptic cell and/or presynaptic
  - terminal, once released, it can bind to either post- or presynaptic receptors. As a result, signal transduction occurs.
- elicits rapid-onset and rapidly reversible responses in the target cell, is removed or inactivated from the synaptic cleft. Causing the termination of the signal, and we go back to the resting stage.

Types of neurotransmitters (Classified depending on their chemical structure)

- Small-molecule neurotransmitters subclasses include:
  - Biogenic amines (epinephrine, dopamine, histamine, serotonin), synthesized from amino acids.
  - Amino acids (GABA, glutamate, aspartate, glycine), Keep in mind that GABA is a modified amino acid (metabolite of glutamate)
  - Acetylcholine
  - Purines (ATP)
- Neuropeptides Classified depending on their chemical structure
- Gases (nitric oxide, carbon monoxide)
- It was once thought that each neuron type released only one small molecule neurotransmitter and one neuropeptide, but it turns out that some neurons can release a combination of multiple small molecule neurotransmitters and multiple neuropeptides.
- Now we know that two or more transmitters (usually a small-molecule neurotransmitter and a neuropeptide) can coexist in neurons (e.g., most spinal motor neurons contain acetylcholine and calcitonin gene-related peptide).
- Which one gets released versus the other is still an area of research, but it appears to depend on the level and localization of calcium ion influx.

Structures of neurotransmitters



VERY IMPORTANT!

Neuropeptides	Small-molecule neurotransmitters
Short-chain peptides (3-60 aa's), Large MW	Endogenous chemicals, Low MW
Slow-acting, Interact with metabotropic receptors, which are GPCRs, so they take time to cause an effect.	Fast-acting, Interact mostly with ionotropic receptors (not always the case).
Slow response	Acute response
Prolonged action	Short-term response
Acts on several receptors	Acts on specific a receptor
Can change metabolism	Most do not change metabolism
Alter gene expression	Most do not change gene expression (some do actually change gene expression)
Synthesized in the ER and Golgi apparatus	Synthesized in the presynaptic nerve terminal (mainly in cytosol)(Exceptions in vesicles)
Synthesized in low concentrations	Synthesized in high concentrations
Found allover the neuron	Found in the axon terminals of presynaptic neurons
Stored in large dense-core vesicles	Stored in small secretory vesicles
released at low increase of cytosolic Ca^{2+} concentrations	released at high cytosolic Ca^{2+} concentrations
Have a different site of actions than their origin	Acts in direct apposition of to the releasing cell
Not re-taken up and not reused by the presynaptic neuron	Can be re-taken up and reused (Go back to the presynaptic neuron where it will get repackaged)
Relatively more potent	Relatively less potent
Terminated when proteolytically degraded or diffused	Terminated by reuptake, uptake by glial cells, diffused, or enzyme degradation

NEUROPEPTIDES

These websites are not
required for exam purposes

LISTS OF NEUROPEPTIDES

NEUROPEPTIDES | HUGO GENE NOMENCLATURE COMMITTEE

NEUROPEPTIDE GENE FAMILIES

LIST OF NEUROTRANSMITTER INHIBITORS AND NEUROPEPTIDES

Some characteristics of neuropeptides

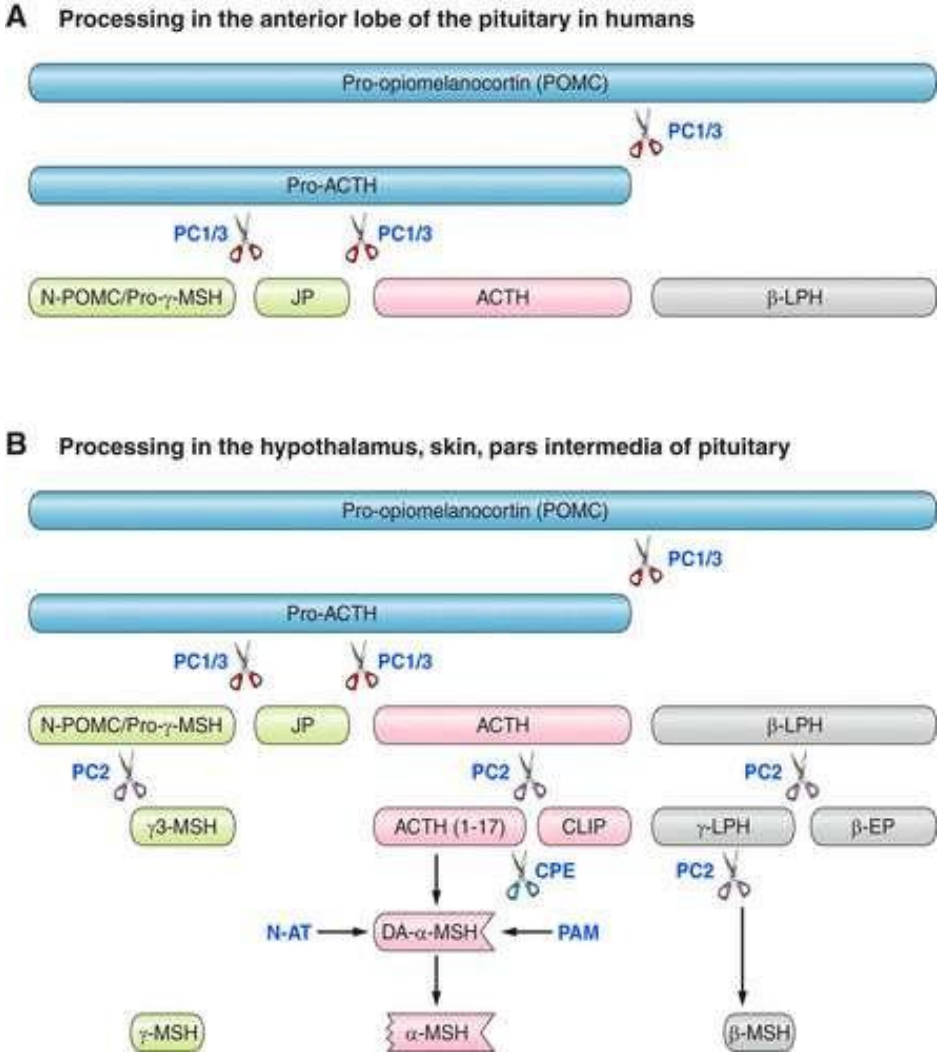
A huge group with variable functions

- More than 50 neuropeptides have been described affecting: Behavior, pain
  - perception, memory, appetite, thirst, temperature, homeostasis, and sleep
- They can be considered neurotransmitters or neurohormones. (Dual function)
- They are synthesized just like proteins are.
  - They are subject to alternative splicing and protein processing, post transcriptional modification at the mRNA level as well as protein expression regulation and post-translational modifications, like cleavage and glycosylation.
  - They can be tissue-specific.
  - Examples: substance P, neurokinin A and proopiomelanocortin

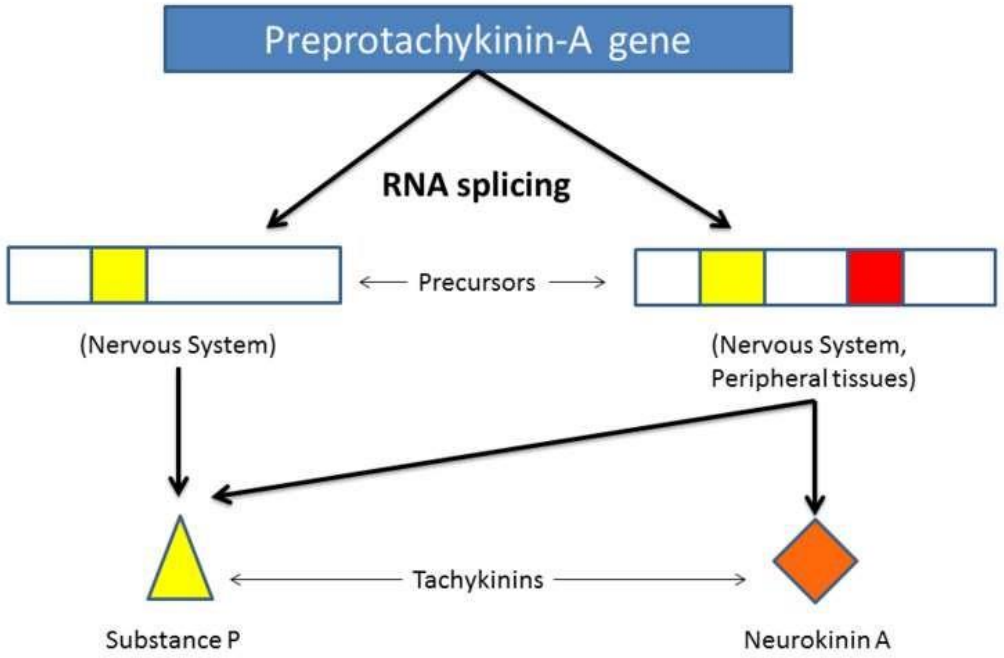
Just enjoy the colors

Examples of neuropeptides and how they get modified

Post-translational processing

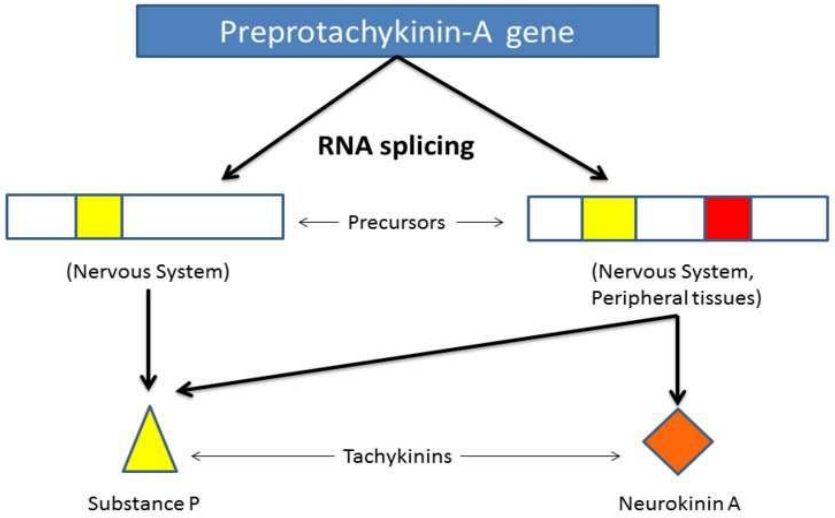


Post-transcriptional processing

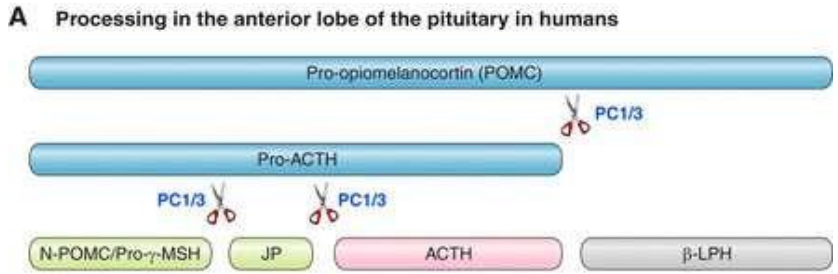


Processing of the pro-opiomelanocortin (*POMC)* precursor proceeds in an ordered, stepwise fashion. Some of the reactions are tissue specific. *ACTH,* adrenocorticotropic hormone; *CLIP,* corticotropin-like intermediate lobe peptide; *JP,* joining peptide; *LPH,* lipotropin; *MSH,* melanocyte-stimulating hormone; *PC,* prohormone convertase.

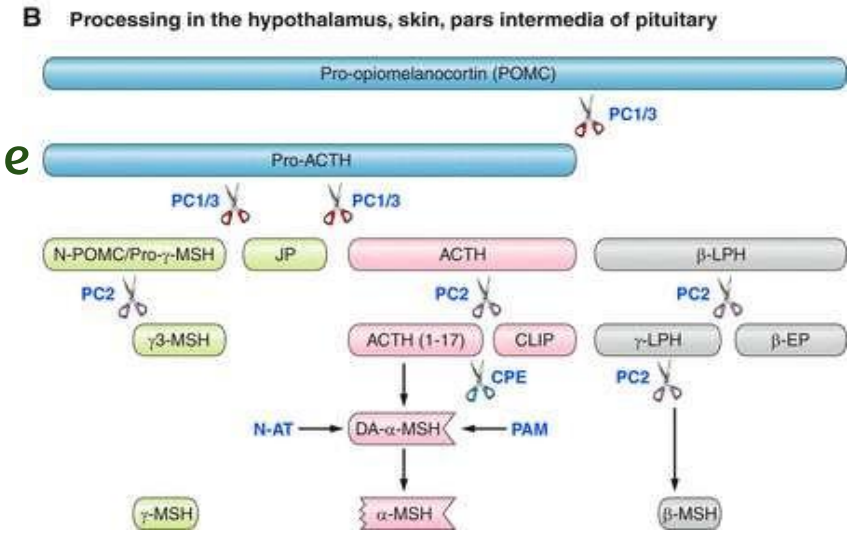
Just enjoy the colors



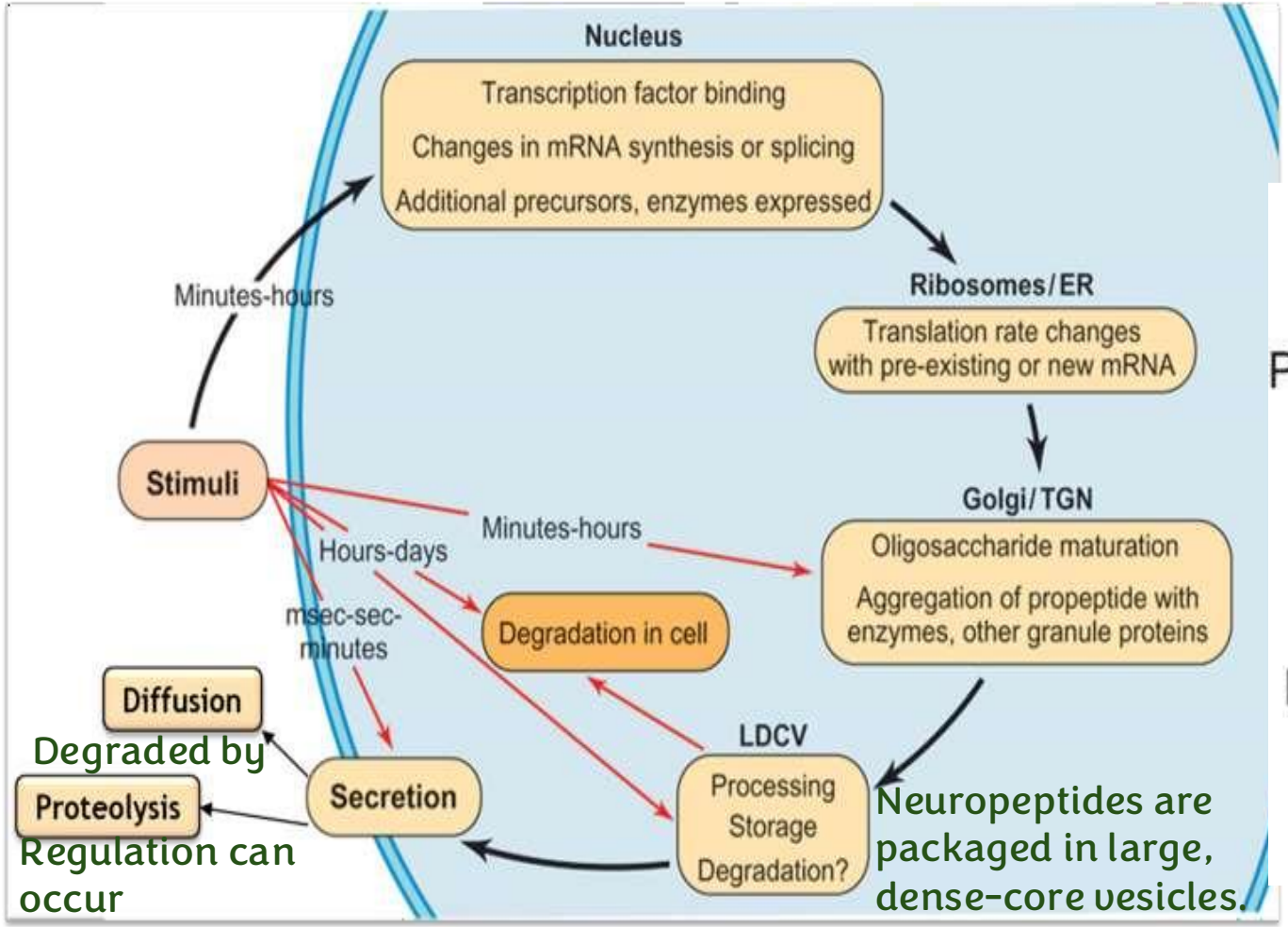
Substance P and Neurokinin A:
Mature mRNA in the nervous system produces Substance P, while in both the peripheral tissue and the nervous system, alternative splicing generates the Neurokinin peptides.



POMC(proopiomelanocortin) is a large peptide synthesized in the hypothalamus and the anterior lobe of the pituitary. After translation, it undergoes cleavage into different peptide hormones. Further cleavage and processing into shorter peptides with different functions can also occur in the hypothalamus, skin, or pars intermedia of the pituitary.



The levels of regulation of neuropeptide expression



Degraded by

Regulation can occur extracellularly

Duration of action is prolonged here

Neuropeptides are packaged in large, dense-core vesicles.

Transcriptional
- Transcription factors
- Histone modifications

Post-Transcriptional
- ncRNA
- RBPs
- mRNA localization
- RNA decay

RNA transport, processing, and splicing

mRNA processing

Where it will go in the cytosol

Binding of miRNA to mRNA effecting its stability

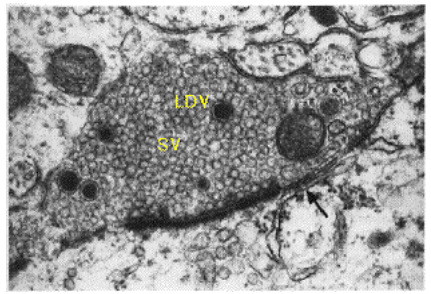
Translational

Post-Translational
- Phosphorylation
- Ubiquitination
- Acetylation
- Glycosylation
- Protein-protein interactions

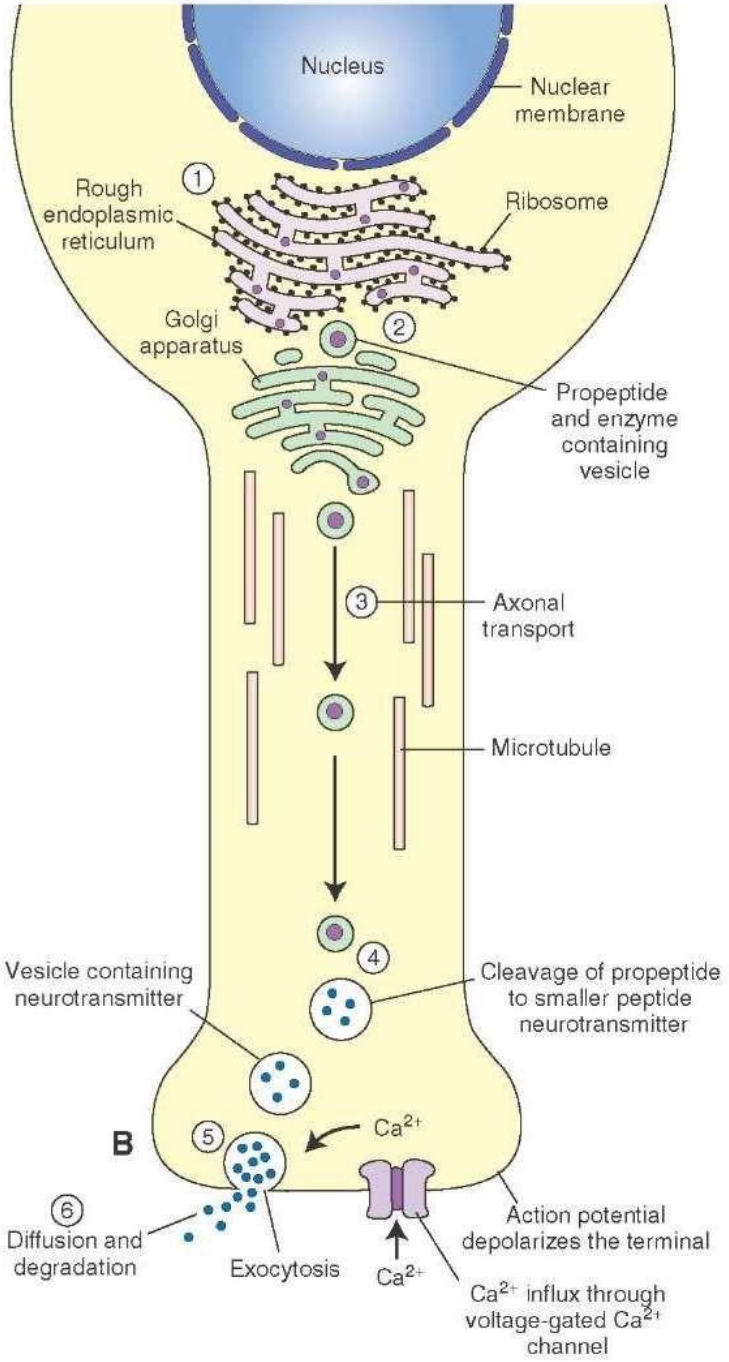
Modification of the neuropeptide, such as cleavage or glycosylation.

Epigenetic mechanisms include DNA methylation, histone modification, and chromatin remodeling into euchromatin or heterochromatin

Stages of action

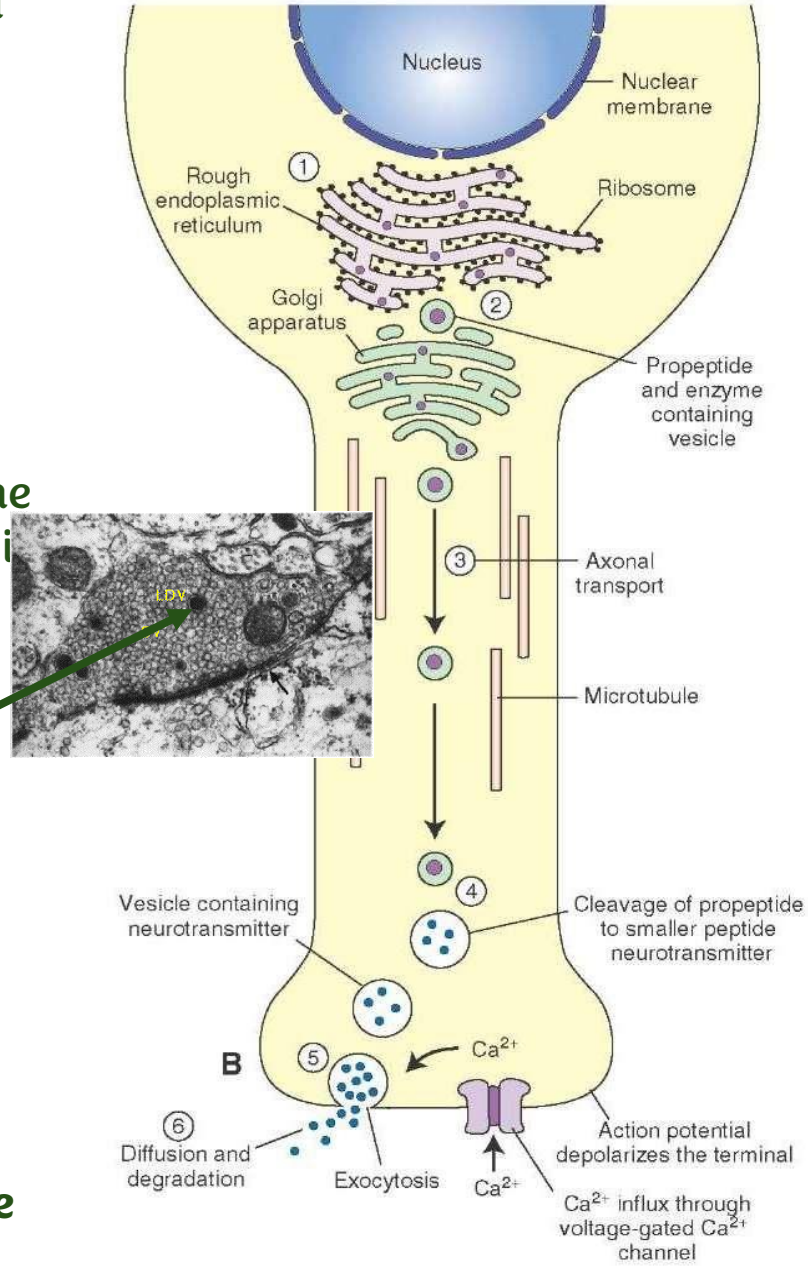


- Synthesized on ER (1) as pre-propeptides then propeptides, and then go into Golgi apparatus (2)
- Packaged into large-dense-core vesicles (with modifying enzymes)
- Transported via (3) **fast**-axonal transport 
  - During the transport, proteases cleave the precursor neuropeptide into the final mature form (4).
- Released (5)
  - Release is gradual over time in response to general increases in the level of intracellular calcium.
- Action (prolonged)
  - Mainly via GPCR
- Termination by diffusion and degradation (6)



Neuropeptide synthesis

1. **Synthesis** begins in the **nucleus** with transcription of the neuropeptide gene.
2. **Translation** occurs at the **rough ER** surface, producing a prepropeptide. The "**pre**" region is a signal sequence (about 70 amino acids) on the ribosome that directs the growing peptide to the ER.
3. As the peptide is **translocated** into the **ER lumen**, the "**pre**" signal sequence i **cleaved off**, leaving the **pro-peptide** (which is still inactive at this stage).
4. From the ER, it moves to the **Golgi apparatus**, where further **modification** occurs before being **packaged** into large dense core **vesicles**.
5. During **axonal transport** to the presynaptic terminal, the vesicles undergo **additional processing**, including **cleavage** of the **propeptide** into the **active** neuropeptide.

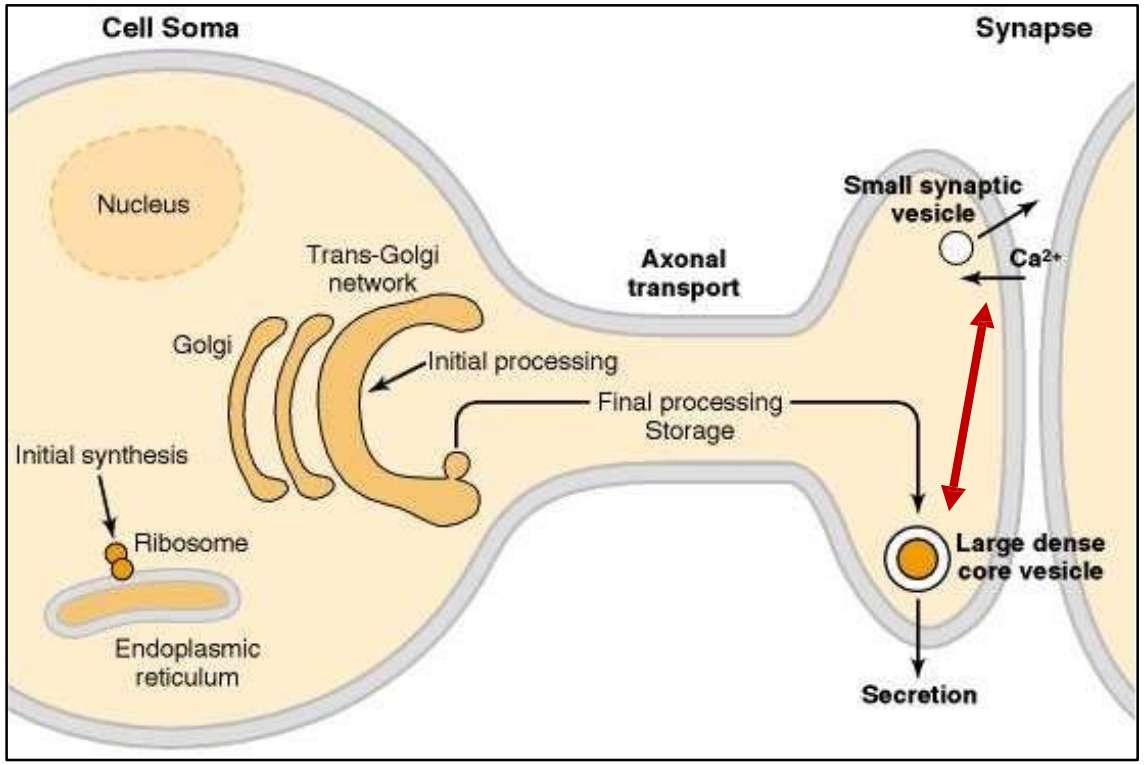


6. Neuropeptides are **released** gradually when **calcium** influx triggers **exocytosis** of the dense core vesicles.
7. The postsynaptic effect is prolonged because neuropeptides typically bind to G protein coupled receptors (**GPCRs**), activating slower, longer lasting second messenger pathways.
8. Once released into the synapse, some neuropeptides can enter the bloodstream and travel to distant target sites (neurohormone function).
9. Neuropeptides are eliminated through diffusion away from the synapse or by proteolytic degradation (enzymatic breakdown), rather than reuptake.

Role of Ca^{2+} ions

Needed for the fusion of the vesicle into the membrane

Their site of calcium entry can be far from where the vesicles are localized, and the concentration of calcium needed for release is lower than that required for small molecule neurotransmitters



- Vesicles are located further away from the presynaptic membrane and away from the area of Ca^{2+} ions influx
- Ca^{2+} ion influx can be from external of internal sources and at lower concentrations than required for small-molecule neurotransmitters.

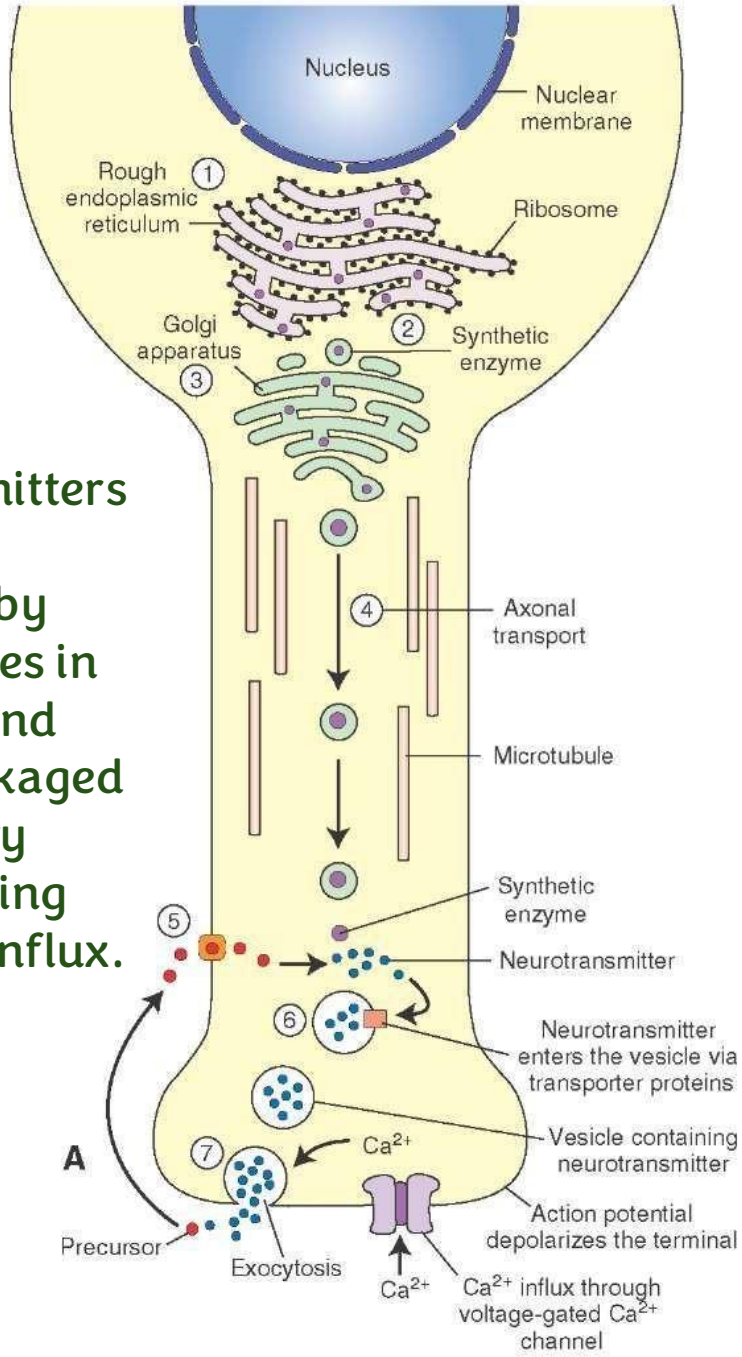
SMALL-MOLECULE NEUROTRANSMITTERS

Stages of synthesis and action

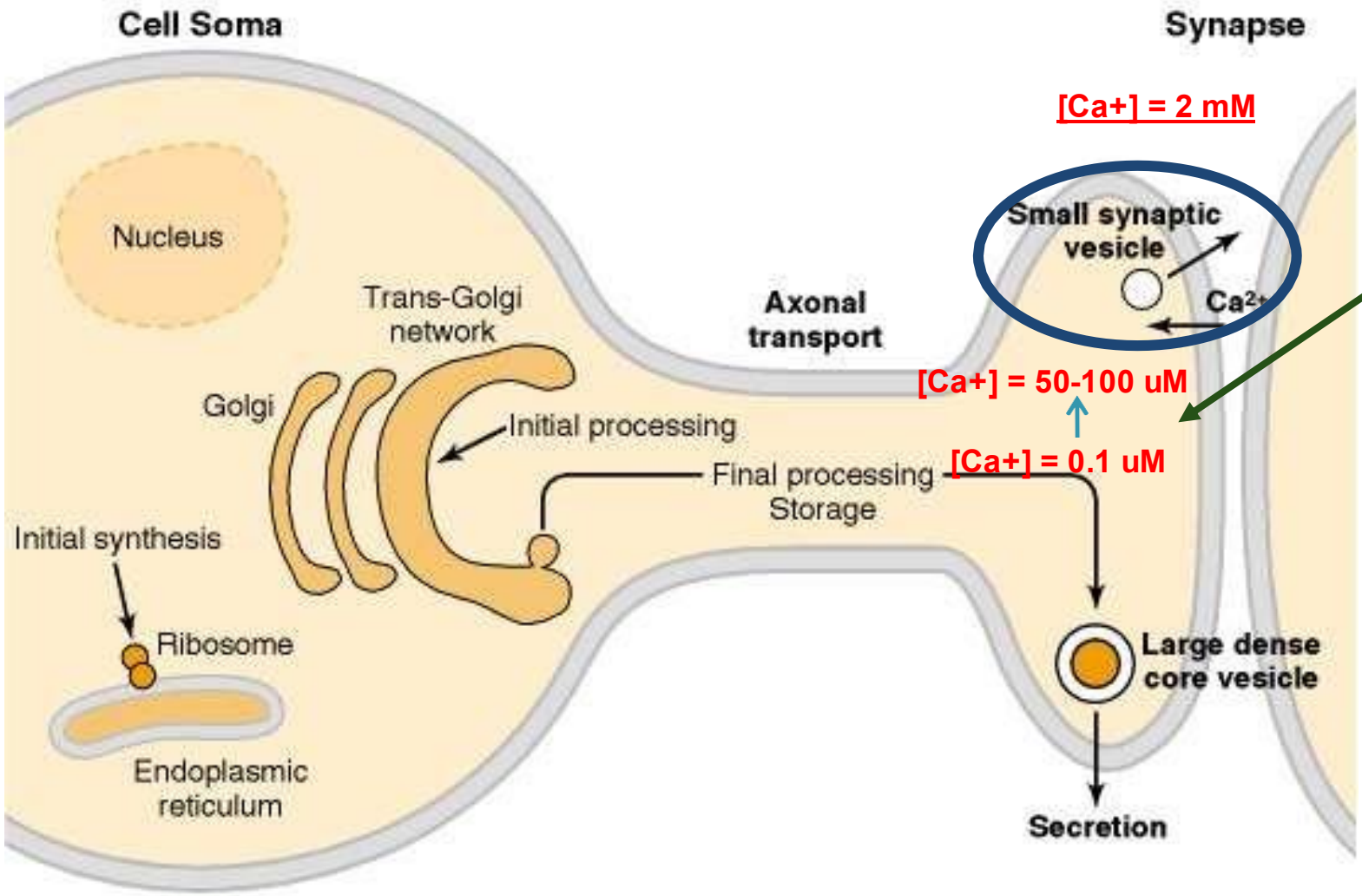
- Synthesis of **the enzymes** in ER (1) and Golgi apparatus (2) where they are modified (3).
- Transport of **soluble** enzymes via **slow** axonal transport by microtubules (4)
- Neurotransmitter precursors are taken up into the cells via transporter proteins located in the plasma membrane of the nerve terminal (5), and the neurotransmitter is synthesized in the presynaptic nerve terminal and then packaged in small synaptic vesicles (6).
- Release is stimulated by brief pulses each time an action potential triggers the influx of calcium (7).
- Action (short)
- Termination by diffusion, re-uptake (repackaged into the presynaptic neuron or glial cells), glial cell uptake, or enzymatic inactivation

Neurotransmitters will be synthesized by these enzymes in the cytosol and then get packaged into secretory vesicles waiting for calcium influx.

Once released, neurotransmitters will go on to bind to receptors on the postsynaptic neuron membrane.



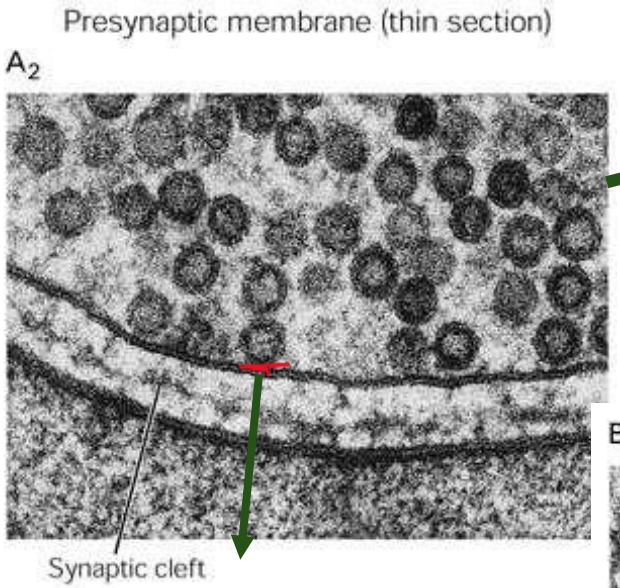
Role of Ca^{2+} ions



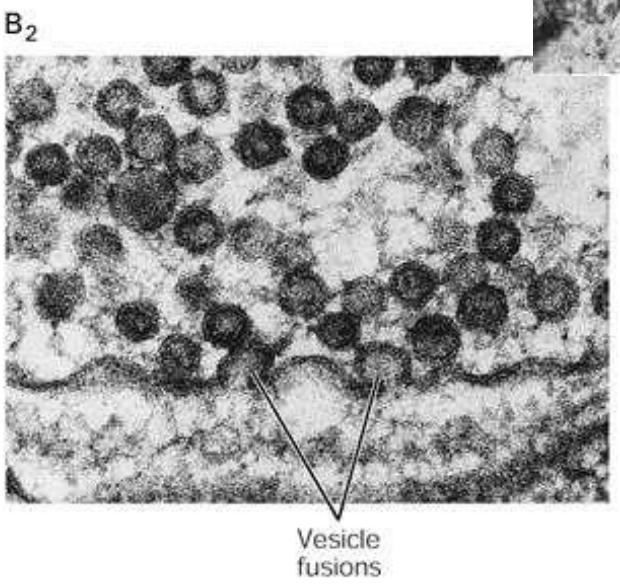
The site of calcium influx will be at the location of the presynaptic secretory vesicles. Here, influx will be extremely high, causing a huge, sudden, acute increase
Note the increase in the calcium ion concentration

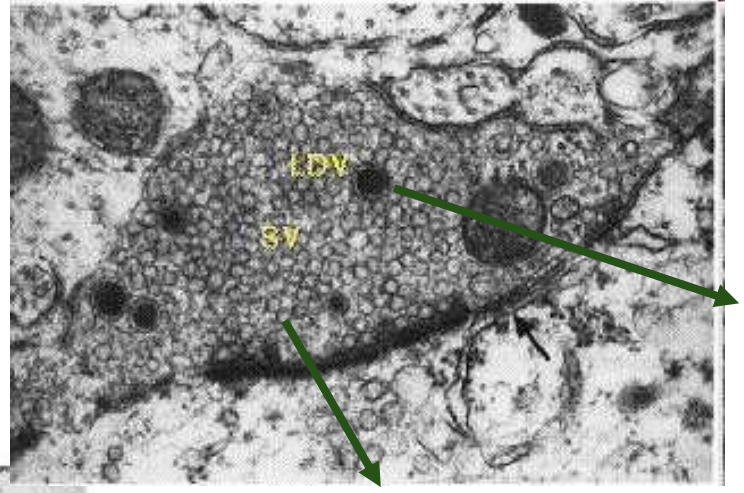
Vesicles are located near the presynaptic membrane and the area of Ca^{2+} ions influx.

Role of Ca^{2+} ions



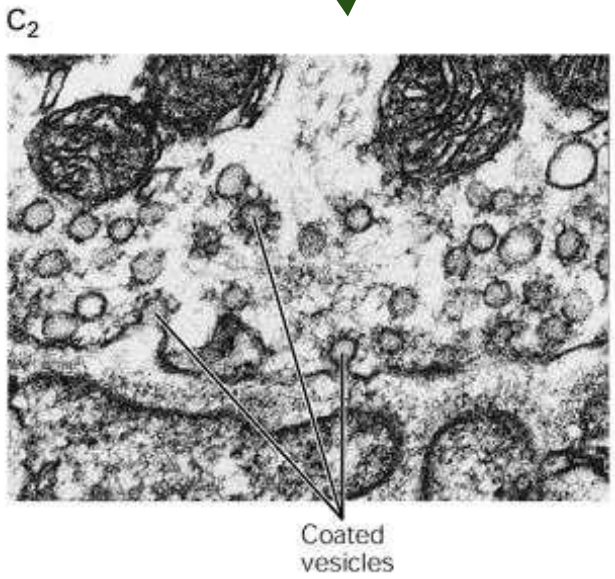
There is a space between the vesicle and the presynaptic membrane that prevents fusion. When calcium rushes in, it brings them closer together, causing the vesicle to dock, fuse with the membrane, and release the neurotransmitter





Large dense core vesicles for neuropeptides

Secretory vesicles will accumulate near the cell surface



The influx of Ca^{2+} ions allows the vesicles to fuse with the presynaptic membranes.

TYROSINE-DERIVED NEUROTRANSMITTERS

Dopamine, norepinephrine, and epinephrine

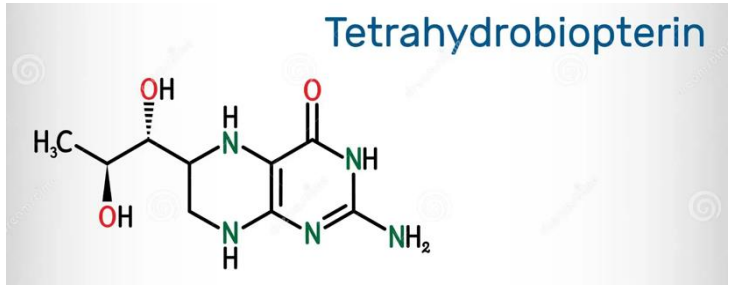
- They are well studied, we have wealth of information regarding small molecule neurotransmitters.
- Their regulation applies to other small molecule neurotransmitters.

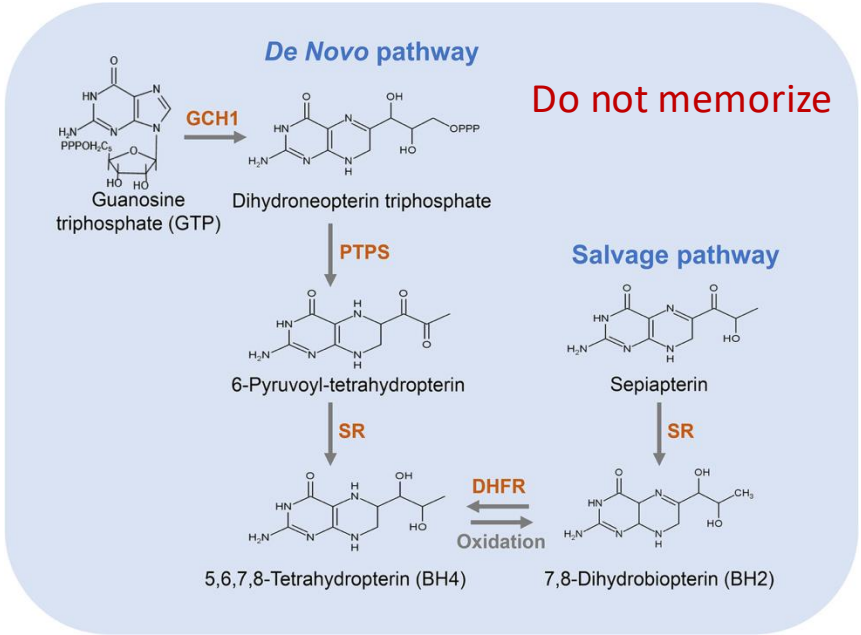
Notes

The synthesis of these neurotransmitters: (dopamine, norepinephrine, epinephrine) requires a number of co-factors.

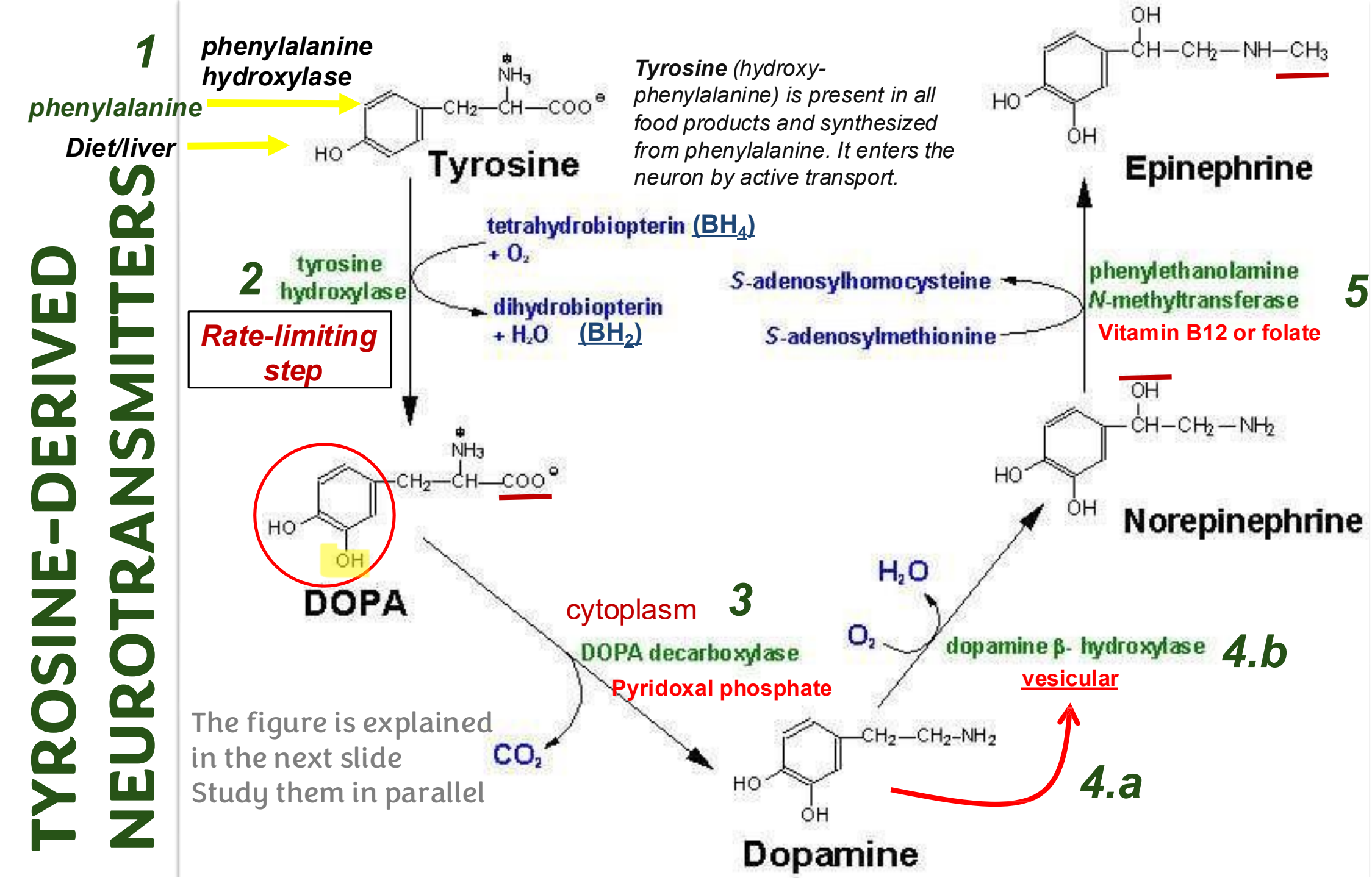
Role of cofactors

1. S-adenosylmethionine (methyl transfer)
   - keep in mind that it requires folate and vitamin B12
2. Pyrodoxal phosphate (vitamin B6)
   - transamination, decarboxylation
3. Tetrahydrobiopterin (BH4)
   - An organic co factor. It is synthesized from GTP and it exists as dihydrobiopterin or tetrahydrobiopterin (structure below).
4. Vitamin B12
5. Folate





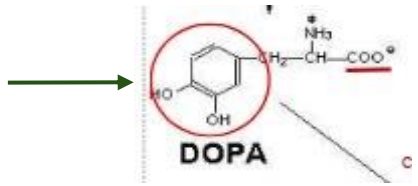
TYROSINE-DERIVED NEUROTRANSMITTERS



TYROSINE-DERIVED NEUROTRANSMITTERS

The doctor said: "Synthesis (of these three neurotransmitters) takes place in the **liver**.", however, the liver synthesizes tyrosine from phenylalanine.Catecholamine neurotransmitters are synthesized within neurons from tyrosine.

1. We can get phenylalanine from diet. We can **synthesize** tyrosine from phenylalanine by phenylalanine hydroxylase.
    - Phenylalanine is an essential amino acid, we must get it from diet, but once we do, we can synthesize tyrosine. Tyrosine is known as a conditional essential amino acid or conditional nonessential amino acid.
2. Tyrosine is hydroxylated again by tyrosine hydroxylase into an intermediate known as DOPA, and this is a **rate limiting reaction** because tyrosine hydroxylase is highly regulated, so it's a slow reaction, and it **requires tetrahydrobiopterin**. *(Tetrahydrobiopterin is needed for hydroxylating rings)*. This reaction happens in the **cytosol**.
    - This ring is called **catechol**, which is why those 3 neurotransmitters are known as catecholamines



3. This reaction also happens in the **cytosol**. We have decarboxylation of DOPA to make dopamine. It requires vitamin B6 (pyridoxal phosphate).
4. a: Dopamine gets packaged inside vesicles. b: Inside vesicles, it gets converted by dopamine beta-hydroxylase enzyme into norepinephrine
5. Norepinephrine then leaves the vesicle, and it gets converted by methyltransferase into epinephrine.

- ❖ Sequence of reactions: hydroxylation, decarboxylation, hydroxylation, methyltransferation
- ❖ Now we are going to take the neurotransmitters one by one.

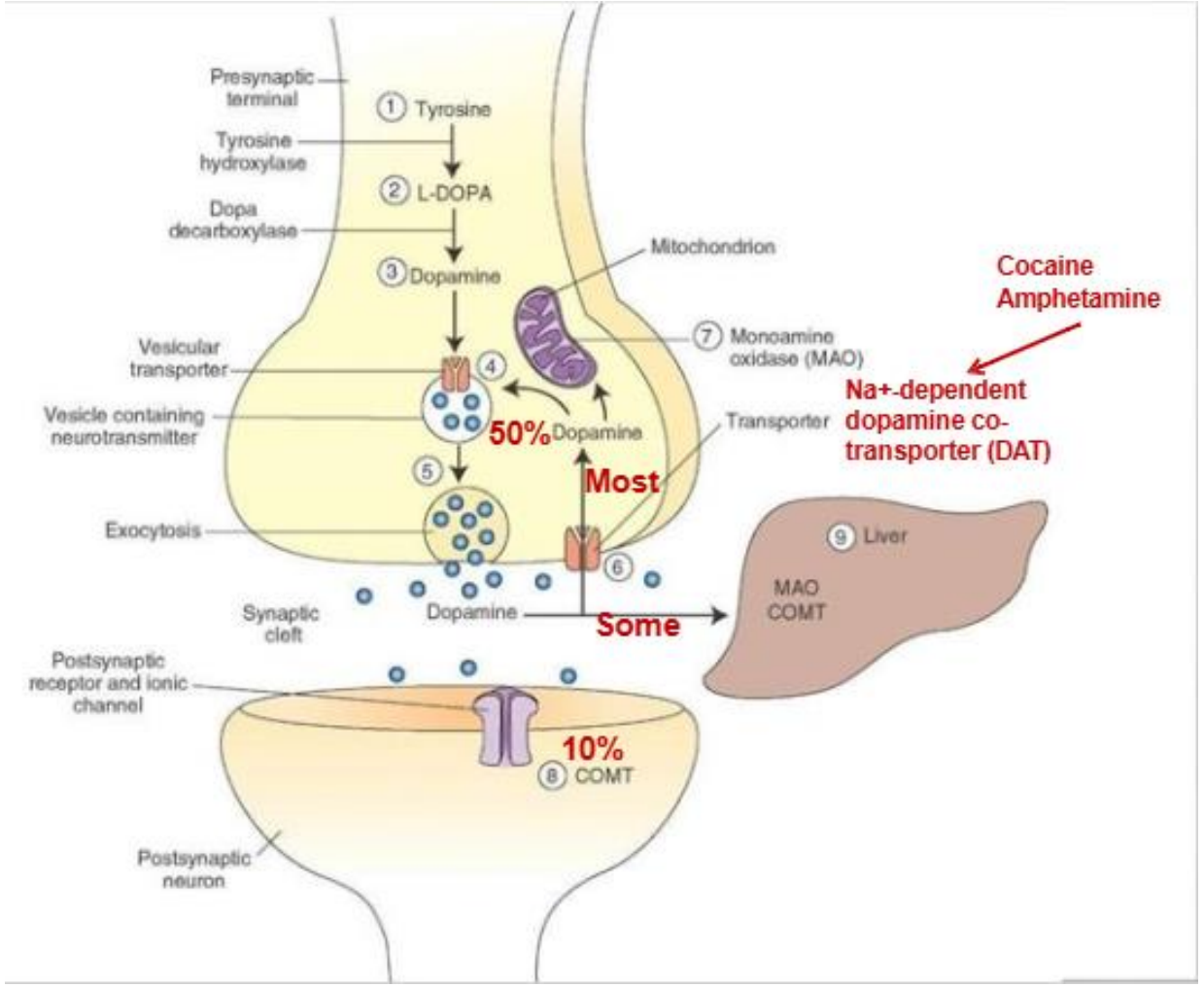
Dopamine

Tyrosine is converted to dopamine, these reactions happen in the **cell periphery.**

Dopamine is then packaged into vesicles. These vesicles wait for the influx of calcium ions, and they fuse with the plasma membrane releasing dopamine outside.

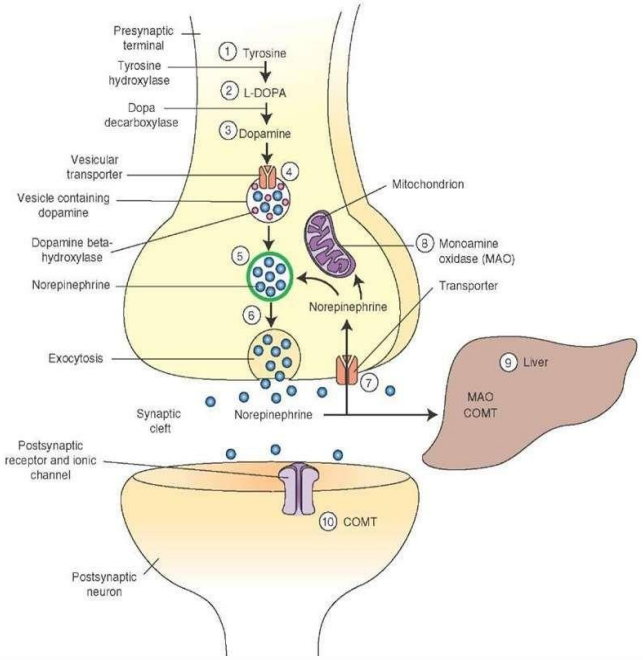
Dopamine binds to a receptor on post-synaptic nerve, and then it has to be inactivated, and it gets inactivated by:

1. Reuptake: Most of it would be taken up again by the pre-synaptic neuron, and inside cells it can be repackaged, or it can be inactivated by monoamine oxidase in the mitochondria.
    - This reuptake takes place via a transporter, sodium ion dependent transporter, and this is the target of cocaine and amphetamine, leaving dopamine in the synaptic cleft for a longer time so that it can induce excitation, energy, and the person would be like fully aware and excited, in other words.
2. Some of the dopamine would leak out into the bloodstream and transported into the liver, where it gets inactivated by also these two enzymes (more on them later)
3. It can be taken up by neighboring cells, including the post-synaptic neuron, where it can also be inactivated.



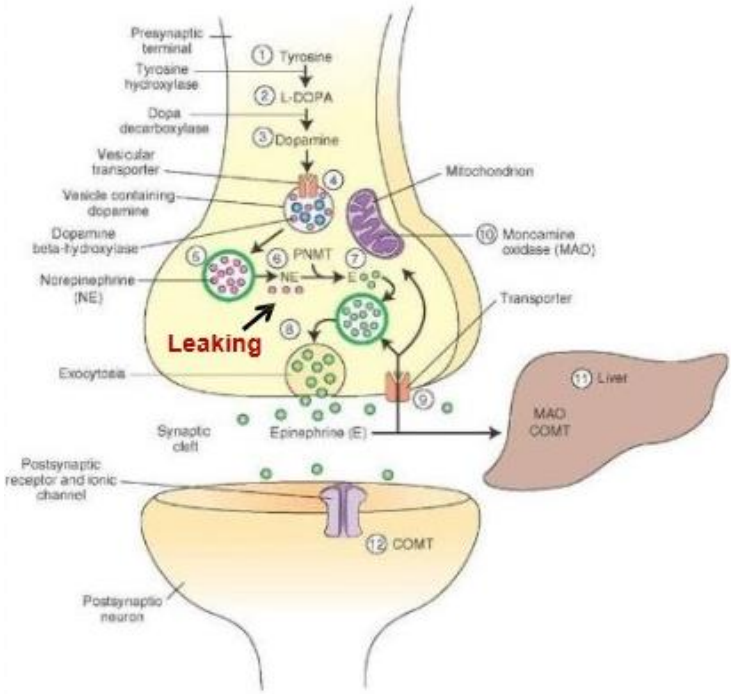
Norepinephrine:

Packaging of dopamine inside vesicles, and in these vesicles there's conversion of dopamine to norepinephrine Then calcium ion influx, fusion of vesicles with the plasma membrane, release of norepinephrine into the synaptic cleft, and the same process of inactivation or termination of signaling: reuptake, uptake by neighboring cells, diffusion, enzymatic inactivation and so on.



Epinephrine:

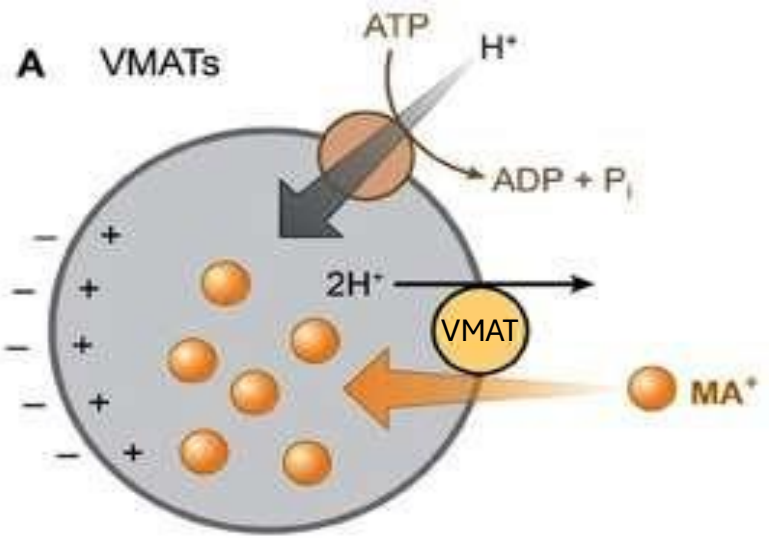
Norepinephrine now is inside vesicles, it leaks out of these vesicles and it gets converted to epinephrine, and then epinephrine is packaged again inside vesicles.. and the same process again: fusion, release, reuptake, uptake by neighboring cells, diffusion into and transport to the liver, enzymatic inactivation and so on.



Packaging into vesicles

The catecholamines (dopamine and epinephrine) are transported into vesicles by an ATP-dependent process linked to a proton pump.

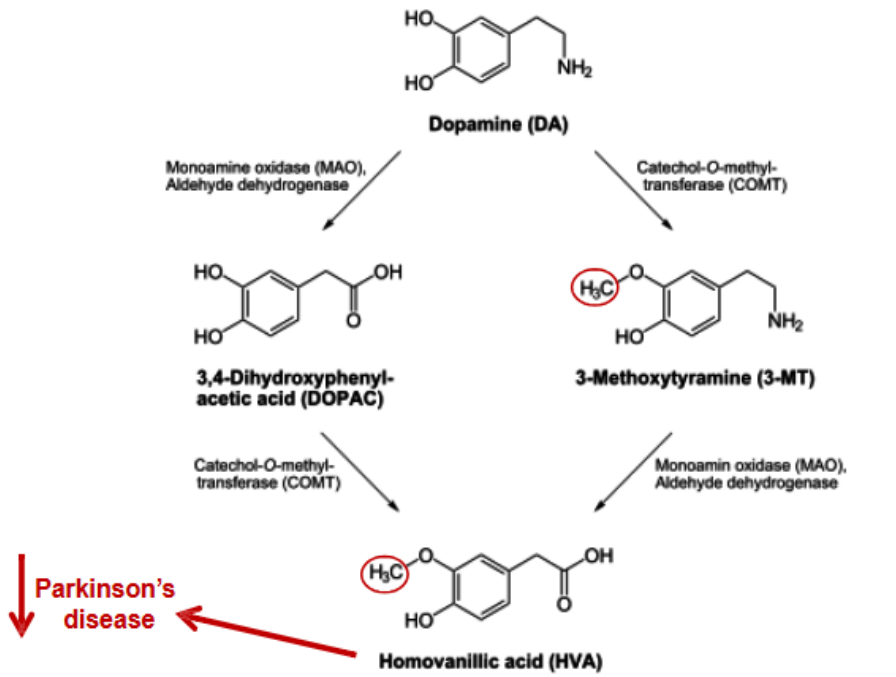
- Protons are pumped into the vesicles by a vesicular ATPase (V-ATPase).
  - ATPase transporter consumes ATP for the entry of protons inside vesicles.
  - The vesicles are acidic inside, low pH, and there are a lot of positive charges inside vesicles versus negative charges outside in the cytosol.
- The protons then exchange for the positively-charged catecholamine via the transporter VMAT (vesicular monoamine transporter)
- Targeting VMATs causes depletion of the neurotransmitters (target for some drugs).

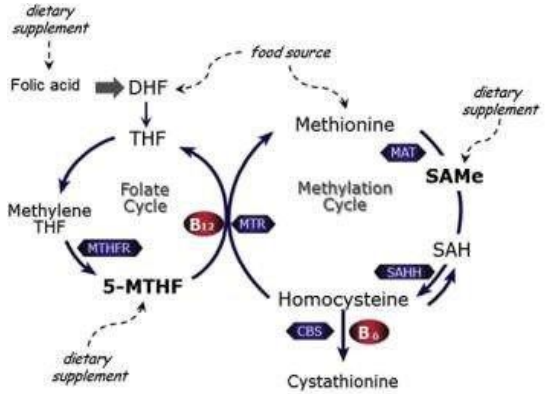


How? See next slide.

COMT and MAO

- Enzymatic inactivation happens via two enzymes:
  - monoamine oxidase MAO
  - catechol-O-methyl transferase COMT.
    - It doesn't matter which one starts first. We could have MAO then COMT, or COMT then MAO.
- **Degradative enzymes are present in the presynaptic terminal and in adjacent cells,** like glial cells, and post-synaptic neurons .
- **Drugs that deplete storage vesicles indirectly increase catecholamine degradation.**
  - because that leaves the monoamine neurotransmitters in the cytosol for a longer time where they can be inactivated enzymatically, and that causes depletion of these neurotransmitters
- Homovanillic acid (HVA) is the end product and it's excreted in urine.
  - High level of dopamine increases HVA in urine.
  - People with Parkinson's disease have reduced level of the HVA in urine (Due to low levels of dopamine).
  - Drugs that inhibit VMAT (e.g., reserpine) prevent vesicular storage of dopamine, increasing its cytoplasmic degradation and thereby increasing urinary HVA levels.





Inactivation is dependent on S-adenosylmethionine (SAM) and, indirectly, vitamin B12 and folate

vitamin B12 and folate are needed for the synthesis of the S-adenosyl methionine inside cells

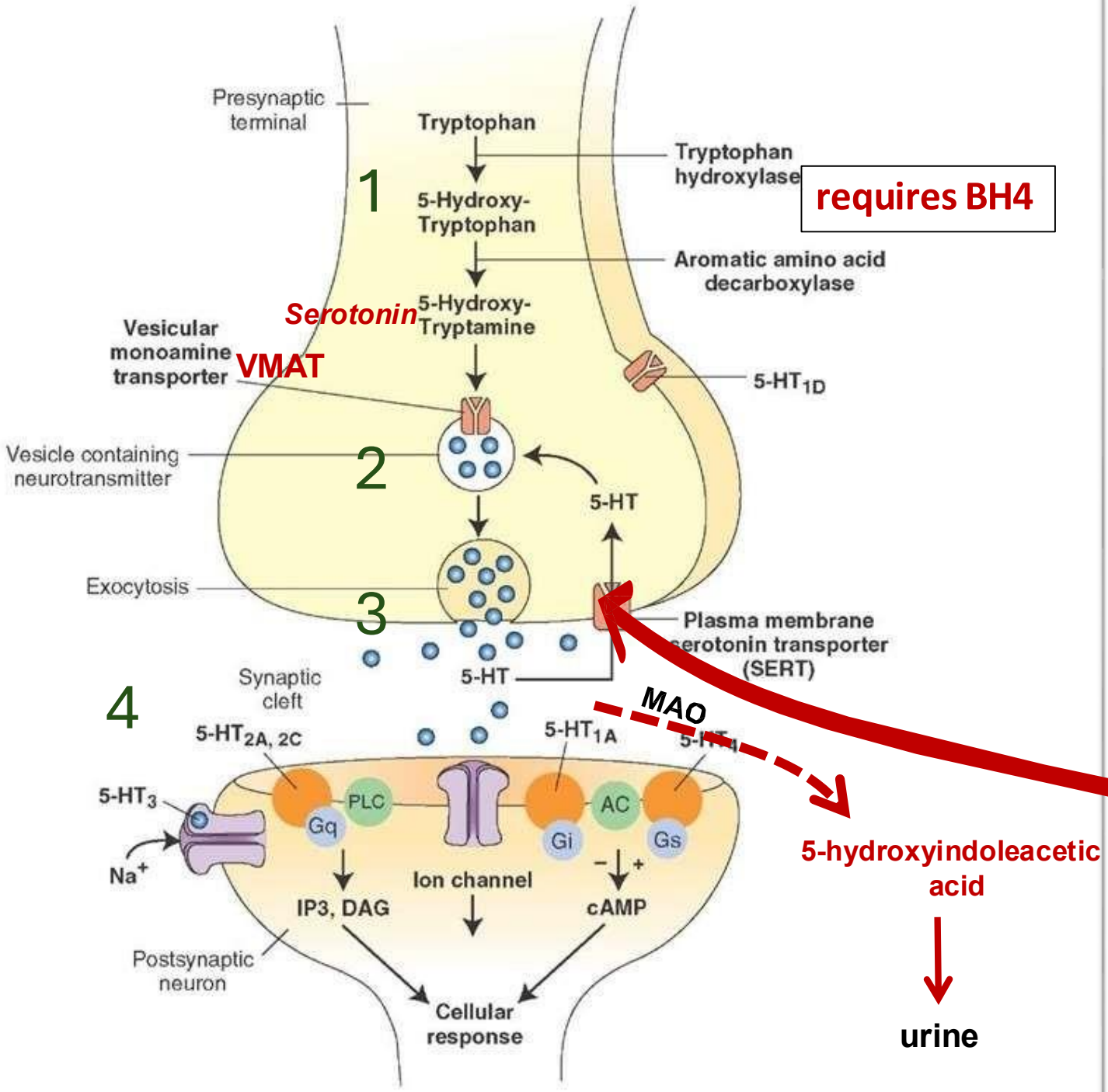
Regulation

- Tyrosine hydroxylase (rate limiting reaction)
  - Short-term regulation (level of enzyme activity)
    - Inhibition by free cytosolic catecholamines, which compete with BH_4 (tetrahydrobiopterin) binding to the enzyme.
    - Activation by depolarization, which activates several protein kinases including PKC, PKA, Ca^{2+}-calmodulin–dependent kinases that phosphorylate tyrosine hydroxylase. This makes the enzyme bind more tightly to BH4 and, consequently, less sensitive to end-product inhibition.
  - Long-term regulation (plus dopamine β-hydroxylase) - level of gene expression (takes hours)
    - Prolonged sympathetic neuronal activity increases the transcription of tyrosine hydroxylase and dopamine β–hydroxylase.

TRYPTOPHAN-DERIVED NEUROTRANSMITTERS

Serotonin and melatonin

Seratonin synthesis

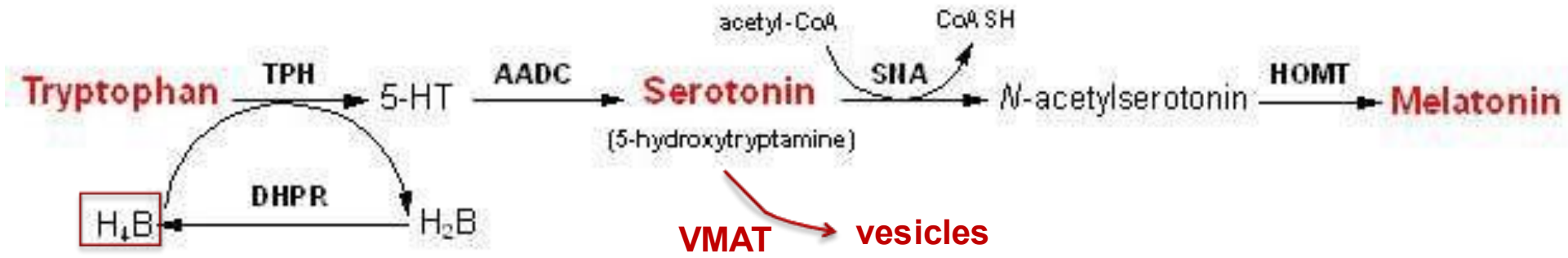


1. Tryptophan is hydroxylated to the final product serotonin, since this is hydroxylation of a ring, we need tetrahydrobiopterin .
2. ***Serotonin is packaged into vesicles by VMAT.***
3. Release: Influx of calcium ions, fusion of the vesicles with the plasma membrane, release of serotonin.
4. Same mechanism of inactivation mentioned before.
5. Serotonin is metabolized by MAO into: 5-HIAA (5-hydroxyindoleacetic acid) This is the main urinary metabolite of serotonin.

Antidepressants, called selective serotonin reuptake inhibitors (SSRIs) like Prozac® inhibit the reuptake process resulting in prolonged serotonin presence in the synaptic cleft.

Melatonin

- Serotonin is synthesized in the pineal gland and serves as a precursor for the synthesis of melatonin by the sequential action of N-acetyltransferase and hydroxyindole-O-methyltransferase (HIOMT) which is a neurohormone involved in regulating:
  - sleep patterns
  - seasonal and circadian (daily) rhythms
  - dark-light cycle

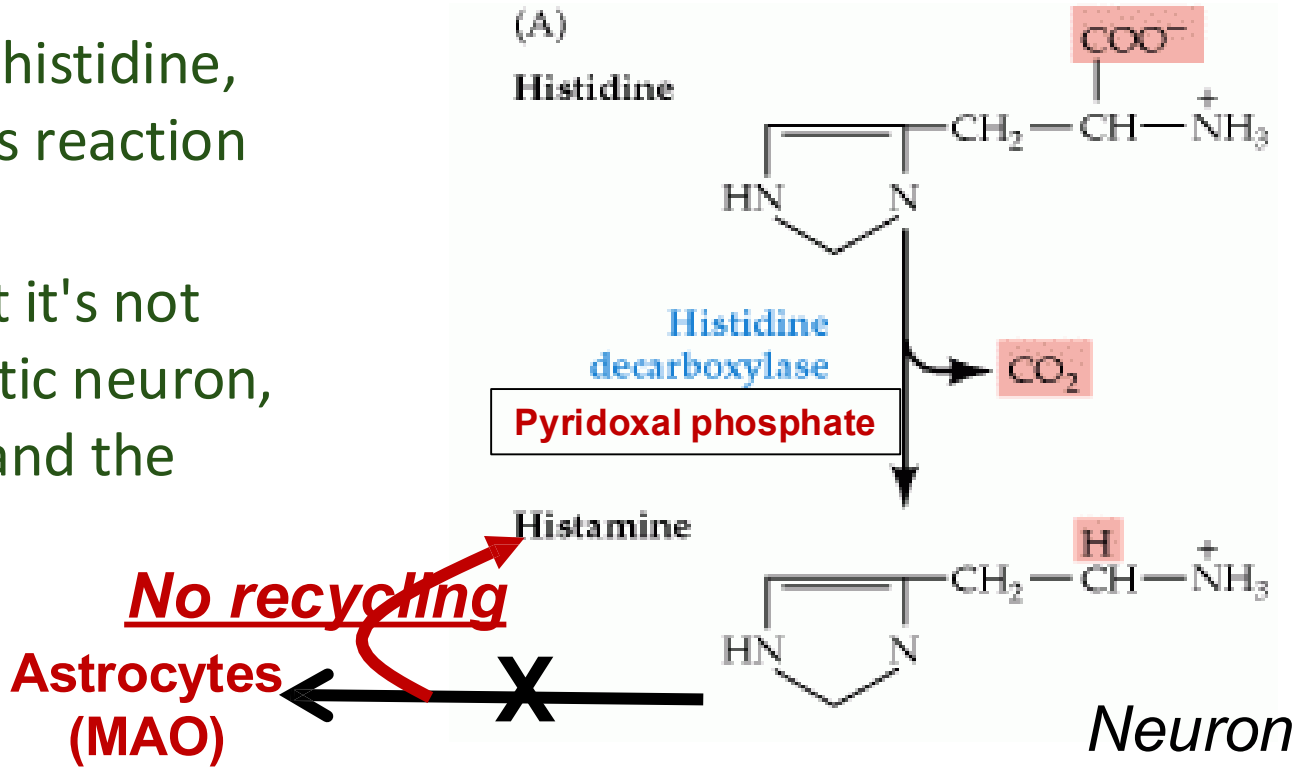


AMINO ACID-BASED NEUROTRANSMITTERS

Some of these amino acids would act as neurotransmitters themselves, or they would be modified a little bit resulting in the production of a neurotransmitter.

Histamine

- It does not penetrate the blood-brain barrier and, hence, must be synthesized in the brain.
- Histamine is inactivated by two enzymes—histamine methyltransferase and diamine oxidase (histaminase).
- **Histamine is packaged into vesicles by VMAT.**
- Synthesis takes place by decarboxylation of histidine, resulting in the production of histamine, this reaction requires vit B6 (pyridoxal phosphate)
- One important thing about histamine is that it's not recycled, it is not taken up by the pre-synaptic neuron, and this is a difference between histamine and the other neurotransmitters.
- In addition, the way it gets inactivated is by monoamine oxidase in astrocytes.

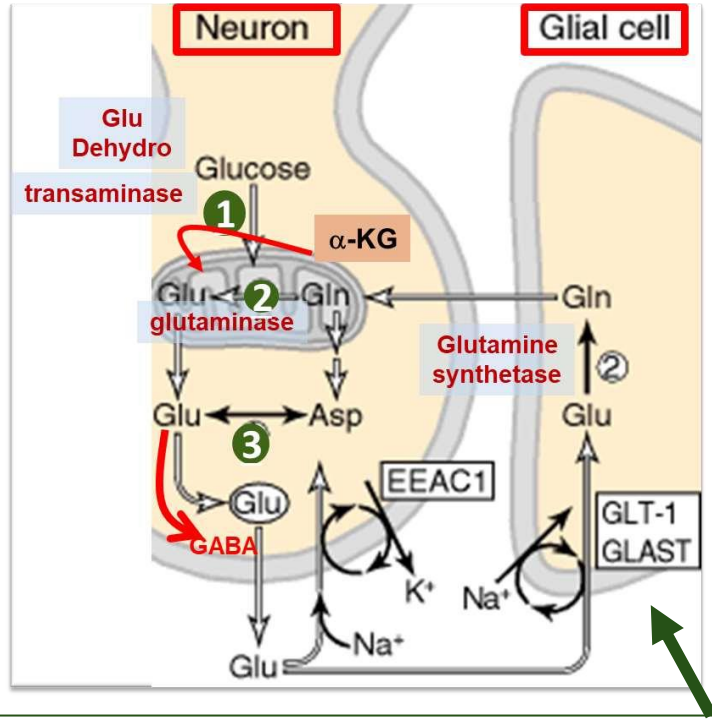


Glutamate and aspartate

- Nonessential amino acids (we can synthesize them ourselves.)
- Do not cross BBB
  - must be synthesized in neurons
- Main synthetic compartments
  - Neurons
  - glial cells
- Both are excitatory neurotransmitters.

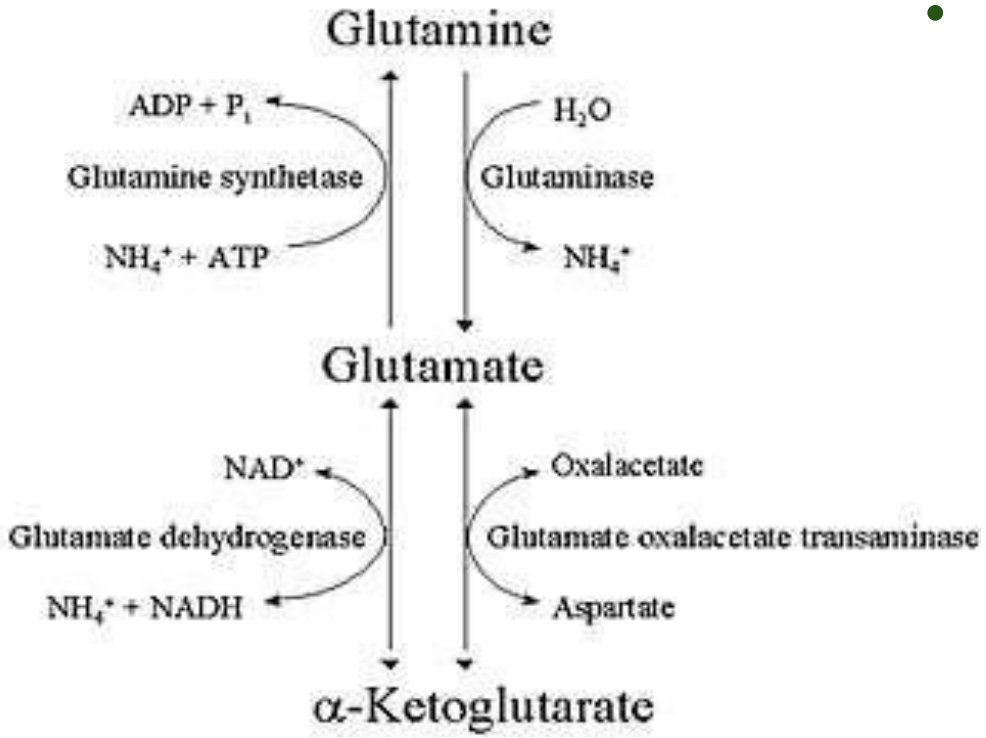
Synthesis of glutamate

- In terms of synthesis of glutamate, three mechanisms or three pathways (Sources):
  1. Glycolysis→Krebs cycle→dehydrogenation of→ ketoglutarate then add an amino group (transamination rxn) → becomes glutamate
  2. Glutamine (deamination)
  3. Aspartate (transamination), also involving α-ketoglutarate, resulting in the production of glutamate.
- Removal
  - Uptake and re-uptake by high affinity
  - transport systems in the nerve terminal (either the pre-synaptic cells or by the post-synaptic cells) and glial cells.



If glutamate is taken up by glial cells, it gets converted to glutamine. Glutamine is transported out to the pre-synaptic neuron where it can be converted to glutamate.

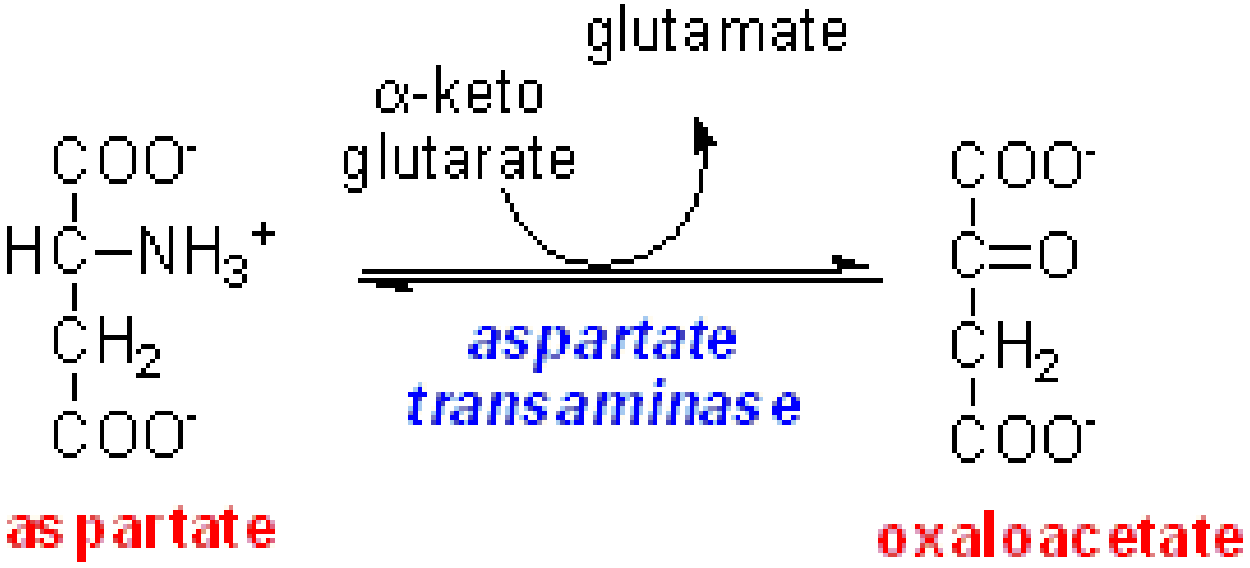
Sources of glutamate *(supplementary)*



- This here just to show you the process
  - Alpha- ketoglutarate can take an amino group from the aspartate. This rxn could be via glutamate dehydrogenase.
  - Glutamate can then be converted to glutamine,
  - And this is the cycle that we learnt before in the metabolism course.

Aspartate

- Note: Similar to glutamate
- Precursor: oxaloacetate (transamination)

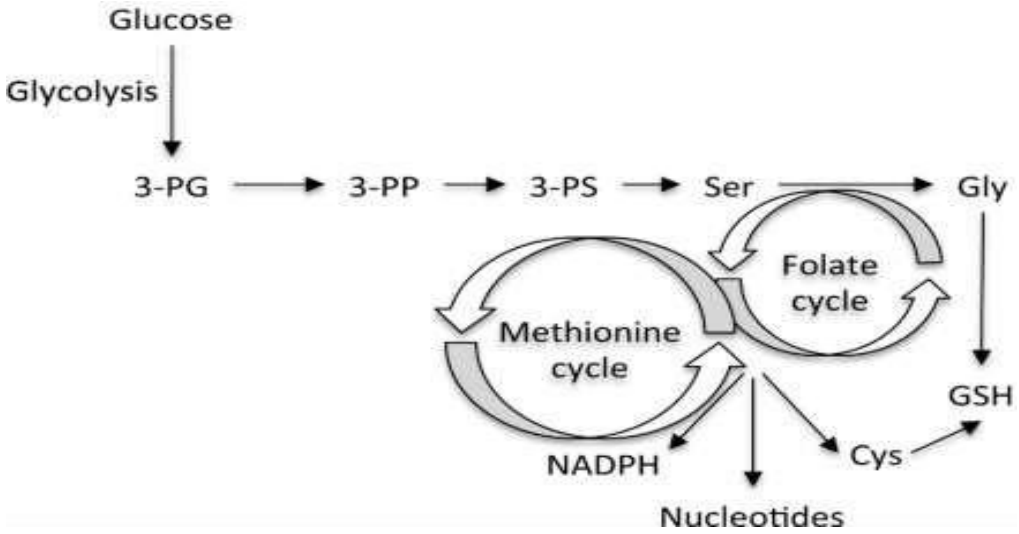


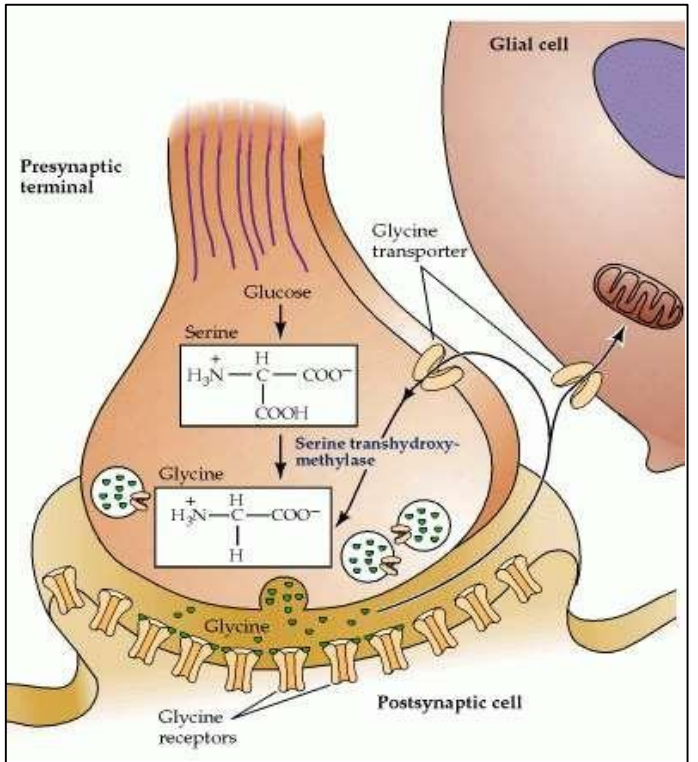
There's controversy on whether aspartate is a neurotransmitter or not, but all we need to know about it is that:-

it gets synthesized from oxaloacetate by transamination reaction, involving alpha-ketoglutarate, glutamate, and aspartate aminotransferase.

Glycine

- A major inhibitory neurotransmitter (in contrast to glutamate and aspartate.)
  - It is synthesized from serine by serine hydroxymethyltransferase through 3-phosphoglycerate. (an intermediate in the glycolytic pathways)
  - Glucose → phosphoglycerate,→ serine →glycine.
- Removal: high-affinity transporter (either by the pre-synaptic neuron or neighboring cells as well.)





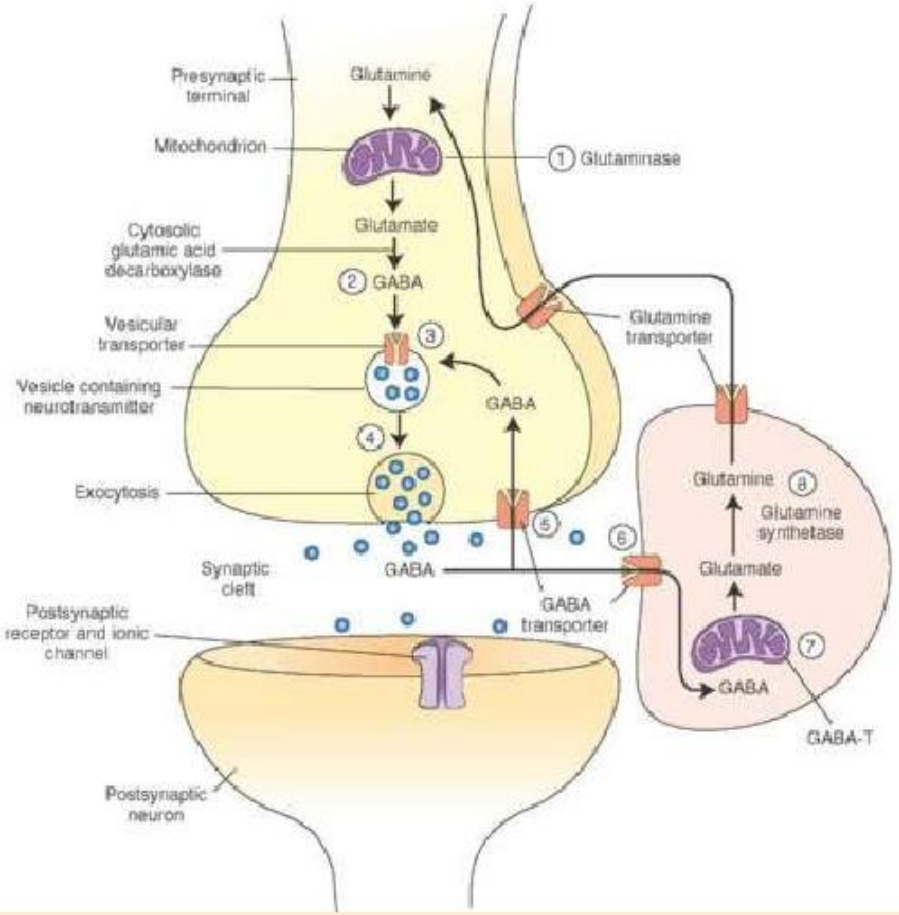
Gamma- aminobutyric acid (GABA)

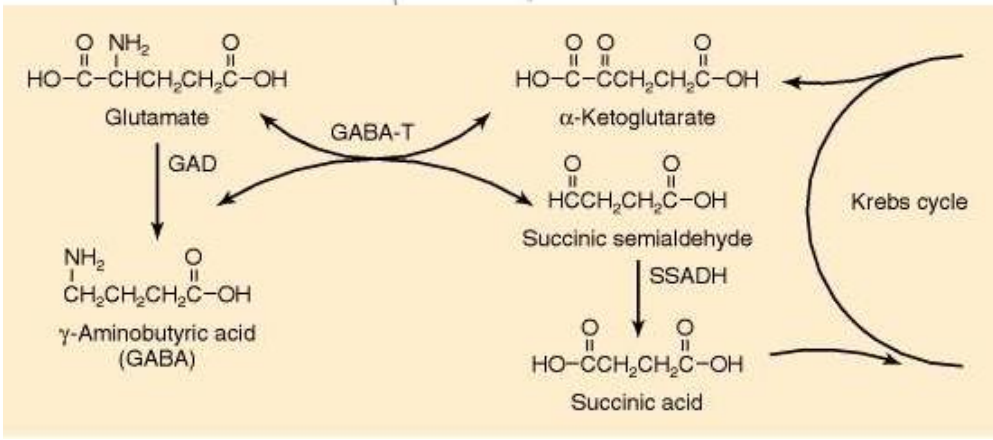
- GABA is present in high concentrations (millimolar) in many brain regions.
  - These concentrations are about 1,000 times higher than concentrations of the classical monoamine neurotransmitters in the same regions.
- The GABA shunt is a closed-loop process with the dual purpose of producing and conserving the supply of GABA.

GABA shunt

A conservation mechanisms of glutamate and GABA

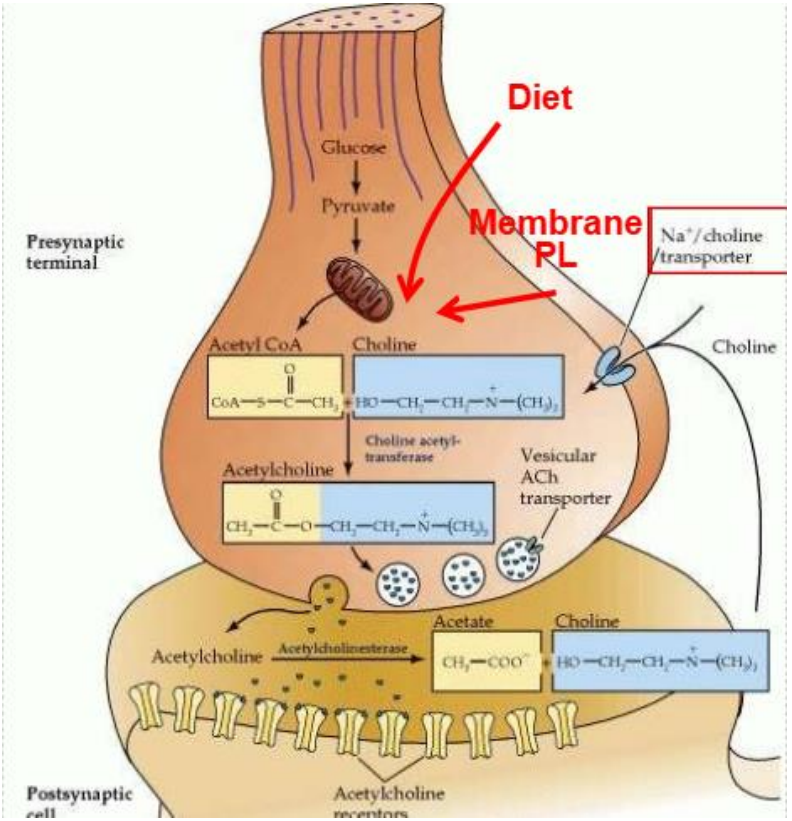
- Synthesis pathway:
  - Gln → Glu by glutaminase.
  - Glu → GABA by glutamate decarboxylase (GAD), which requires pyridoxal phosphate (vitamin B6).
  - GABA is stored in vesicles until released (once you have influx of calcium ions.)
- GABA is either
  - taken up into the presynaptic terminal and repackaged
  - goes into the GABA Shunt where it is taken up into the glia and converted to Glu.
    - Glu is converted into Gln, which is transported into the neighboring nerve terminals to synthesize Glu.





Synthesis of acetylcholine

- Synthesis: Choline + acetyl coenzyme-A by choline acetyltransferase in the cytoplasm.
  - We get choline either from diet, or from the plasma membrane (phosphatidylcholine for example)
- Transported into and stored in vesicles.
- Removal: hydrolysis by acetylcholinesterase
  - this is how acetylcholine is inactivated, it gets converted to choline and acetate, and choline can be taken up by cells again.
  - **Clinical application about sarin gas: Sarin** is an organophosphate nerve agent that irreversibly **inhibits acetylcholinesterase**, leading to accumulation of acetylcholine at synapses and continuous stimulation of cholinergic receptors leading to flaccid paralysis.

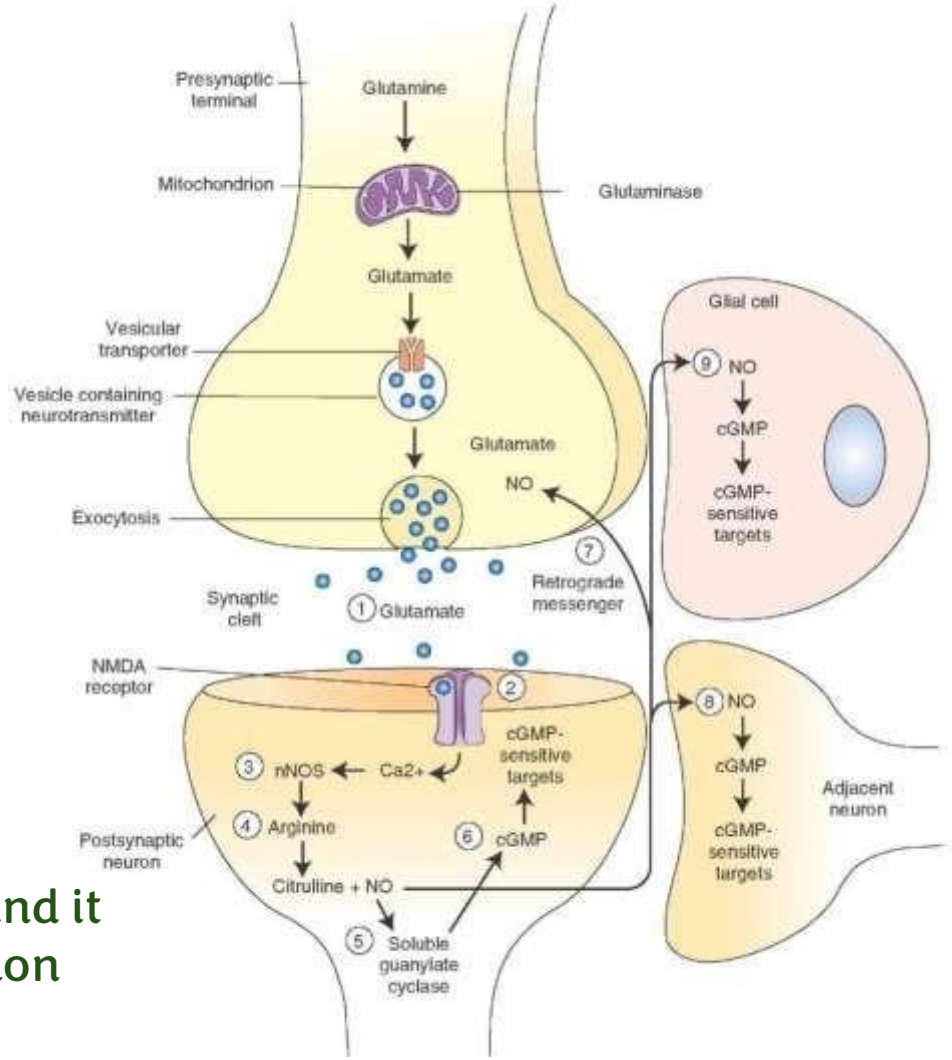


Nitric oxide (NO)

It's a gas and is considered an exception to the neurotransmitter criteria we set earlier.

- Glutamate is released (1) and acts on the N-methyl-D-aspartate (NMDA) receptors located on the post-synaptic neuron (2)
- Ca^{2+} enters the postsynaptic neuron activating nitric oxide synthase (NOS) (3), which forms NO from arginine (4).
- NO stimulates guanylate cyclase forming cGMP (5), which results in a physiological response (6)
- NO can diffuse out:
  - a) to the presynaptic terminal (retrograde messenger) (7) and
  - b) into adjacent neurons (8) and glial cells (9) stimulating guanylate cyclase.
- It has a half-life of 2-4 seconds.
- NO is inhibited by hemoglobin and other heme proteins which bind to it tightly.

Very short action duration and it will be inactivated by diffusion



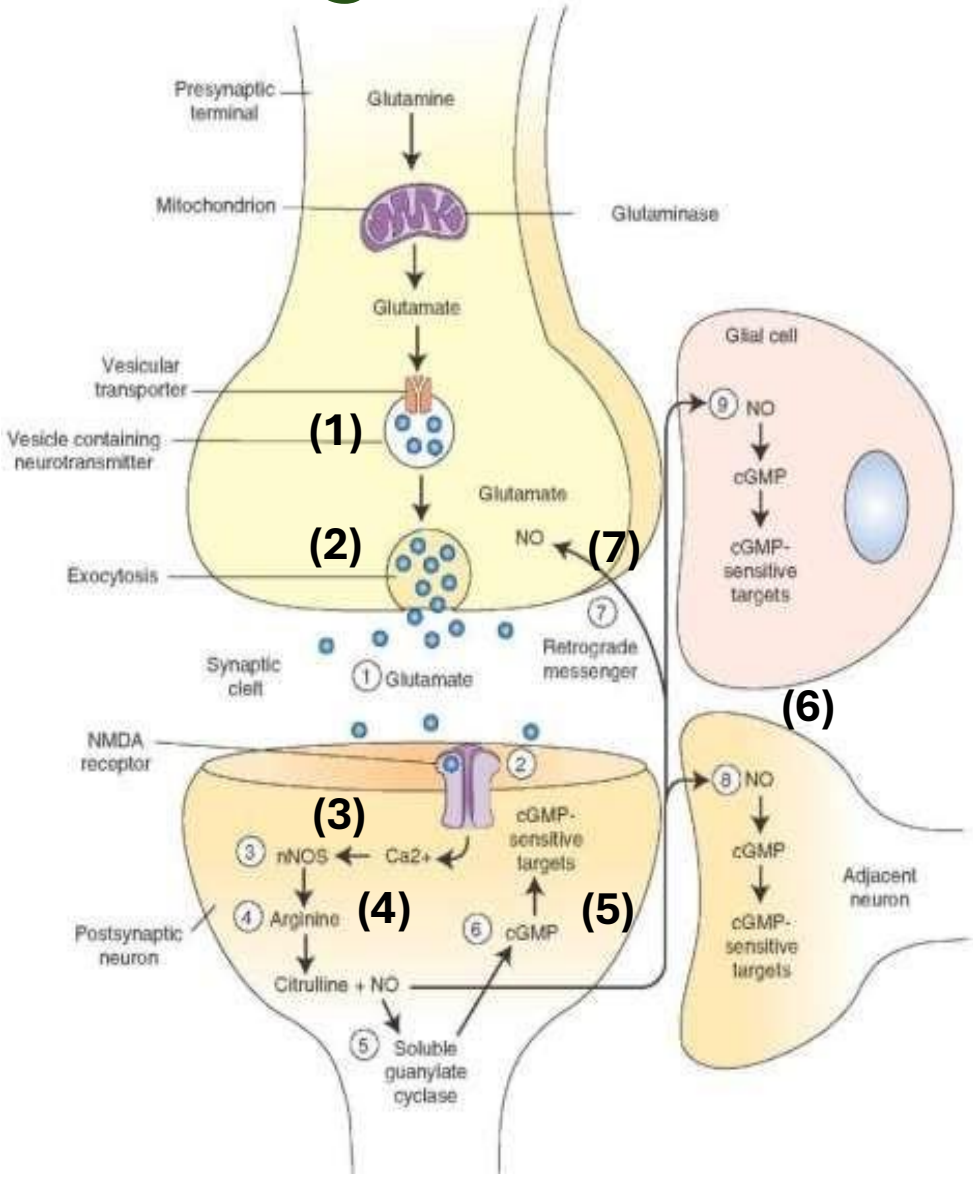
NO synthesis

(1) Glutamate is Packaged inside vesicles

(2) Upon depolarization, glutamate is released into the synaptic cleft, where it binds to postsynaptic NMDA receptors

(3) Once it binds, Ca^{2+} rushes into the postsynaptic neuron, activating NOS inside it.

(4) NOS will convert Arginine into NO



(5) NO will go on to stimulate guanylyl cyclase, which produces cGMP. cGMP then binds to various proteins and enzymes, triggering a physiological effect

(6) NO can also diffuse out and enter neighbouring cells, like glial cells or other neurons.

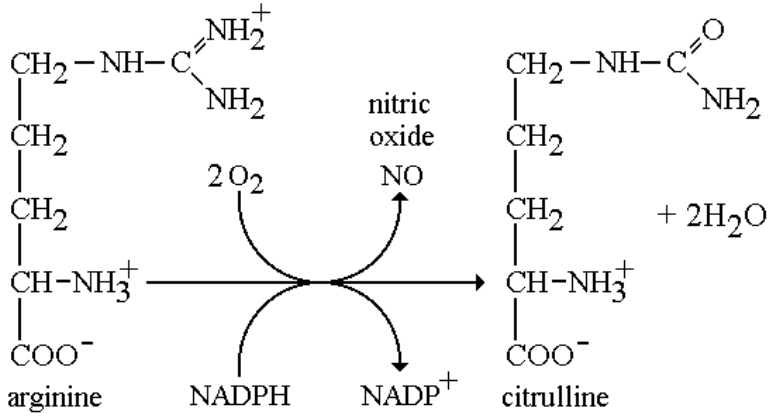
(7) NO can also go into the presynaptic terminal, activating cGMP production in these cells, and this is called a retrograde messenger.

Is NO a neurotransmitter?

- Yes, but:
  - It is not stored in vesicles
  - It is not released by calcium-dependent exocytosis (it diffuses)
  - Its inactivation is passive (there is no active process that terminates its action) (No reuptake but instead it diffuses through the plasma membrane)
    - It decays spontaneously (it has a short half life)
  - It does not interact with receptors on target cells, it goes inside cells
    - Its sphere of action depends on the extent to which it diffuses, and its action
    - is not confined to the conventional presynaptic-postsynaptic direction.
  - NO acts as a retrograde messenger and regulates the function of axon terminals presynaptic to the neuron in which it is synthesized.

NO synthase Just know the isoform found in neurons.

- Isoform I (nNOS or cNOS)
  - Neurons and epithelial cells Target for different drugs
  - activated by the influx of extracellular calcium
- isoform II (iNOS)
  - Macrophages and smooth muscle cells
  - induced by cytokines
- and isoform III (eNOS)
  - Endothelial cells lining blood vessels
  - activated by the influx of extracellular calcium



- All three isoforms require BH2 as a cofactor and nicotinamide adenine dinucleotide phosphate (NADPH) as a coenzyme.

BIOCHEMISTRY QUIZ LECTURE 2

رسالة من الفريق العلمي

اللهم إن عمر عطية في ذمتك وحبل جوارك، فقِه من فتنة القبر وعذاب النار،
أنت أهل الوفاء والحق، فاغفر له وارحمه إنك أنت الغفور الرحيم.

وَءَاتَىٰكُم مِّن كُلِّ مَا سَأَلْتُمُوهُ ۚ ٣٤

And He gave you all that you asked for.

For any feedback, scan the code or click on

Corrections from previous versions:

Versions	Slide # and Place of Error	Before Correction	After Correction
V0 → V1			
V1 → V2			