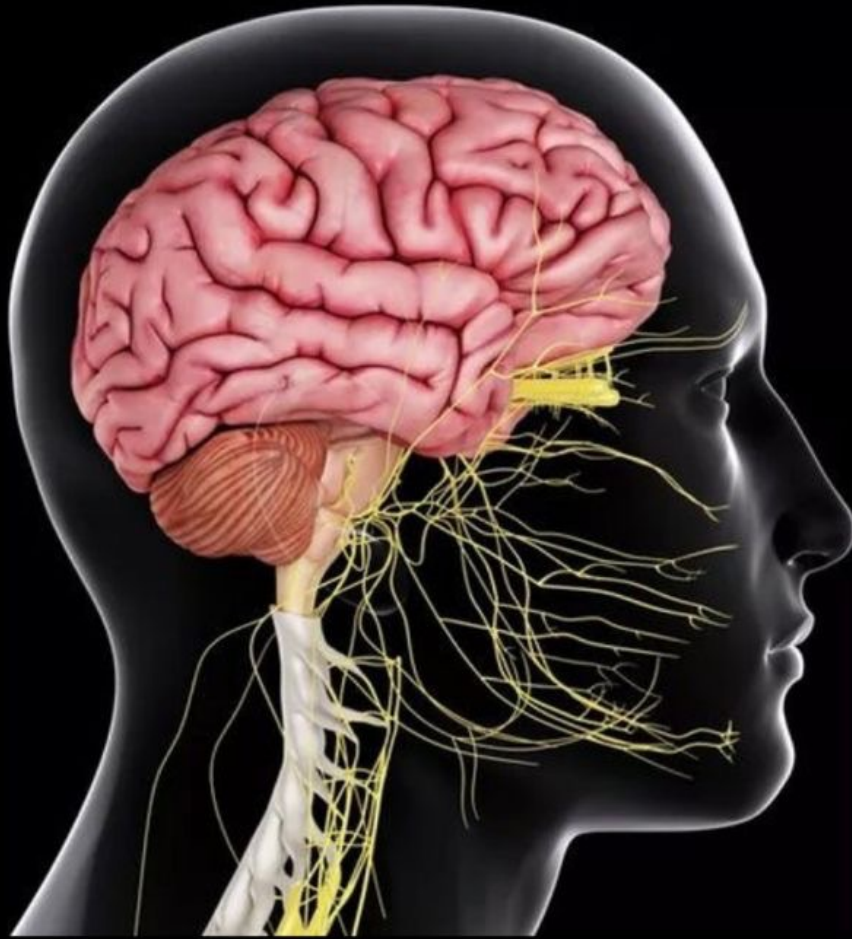




CENTRAL NERVOUS SYSTEM



SUBJECT : Biochemistry

LEC NO. : 2

DONE BY : Batool ALzubaidi

وَقُلْ رَبِّ زِدْنِي عِلْمًا

Biochemistry of CNS Neurotransmitters II

Glutamate

- Glutamate is the main excitatory neurotransmitter and may be responsible for 75% of the excitatory transmission in the CNS.

It means that we can't have glutamate in presynaptic neurons if it doesn't cross the BBB » we will need precursors that can enter the presynaptic neuron and there it will be transformed into glutamate

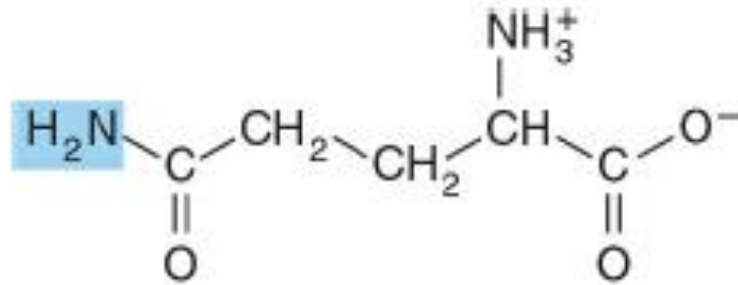
- Nonessential amino acid that **does not cross the blood brain barrier** and must be synthesized in neurons from local precursors that can get into the brain.

Non essential amino acid that can cross the blood brain barrier, it's mostly synthesized in the muscles and stored there

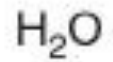
- In the brain, **glutamine** is the fundamental building block for glutamate from glutamine using an enzyme called **glutaminase**.

Mitochondrial enzyme

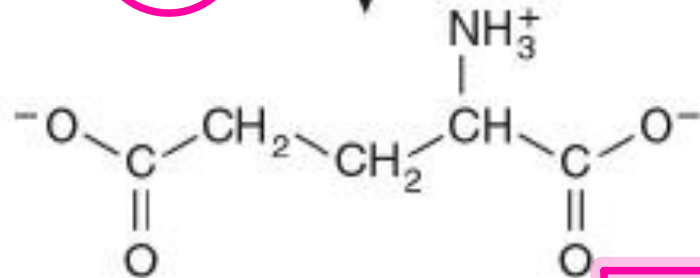
- The most prevalent biosynthetic pathway synthesizes glutamate from glutamine using an enzyme called glutaminase.



L-Glutamine



Glutaminase



L-Glutamate

I

It's action results with ammonia, it needs phosphate which comes from hydrolysis of ATP which accompanies the depolarization process

It's a non essential amino acid » can be synthesized inside the body

Glutamate

First source of glutamate in presynaptic neurons

- Glutamine a **nonessential AA**, is the **most abundant** of the twenty AA's the body **uses to build proteins**.
- Most glutamine is **made and stored in muscle**.
- It is one of the few amino acids that can **directly cross the blood brain barrier** so the **glutamine pool in muscle can be used to support the brain**.

Glutamate

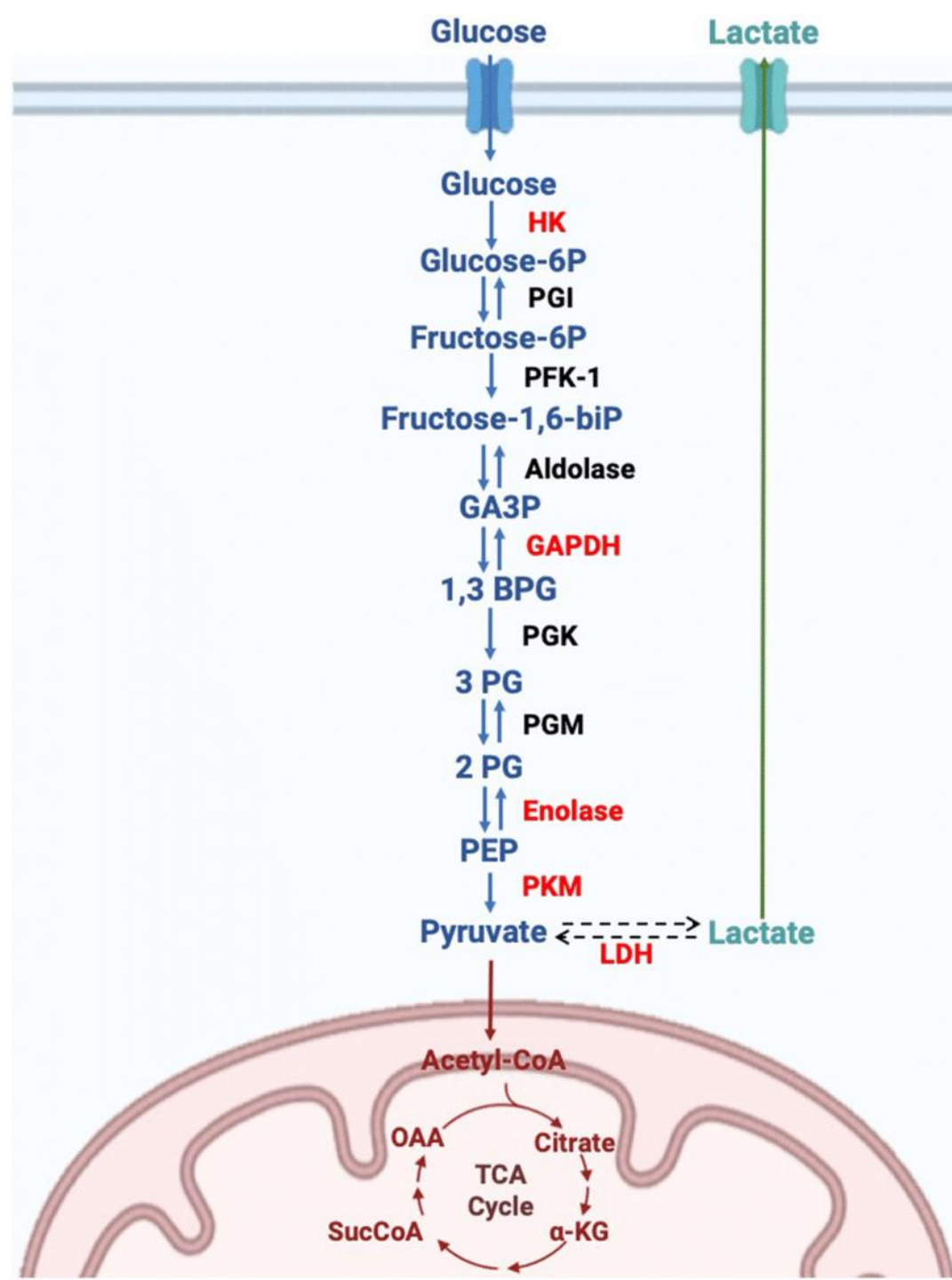
Synthesized indirectly

Second source of glutamate
in presynaptic neurons

- Glutamate can also be produced from **glucose through glycolysis**.
- The glycolysis pathway yields **pyruvate** which enters the **tricarboxylic acid cycle (TCA)**.
- The TCA cycle forms **multiple important intermediates**. One of these intermediates is **α -ketoglutarate (α -KG)**.

Krebs cycle

Can be transformed into glutamate via 2 ways » glutamate dehydrogenase and transamination



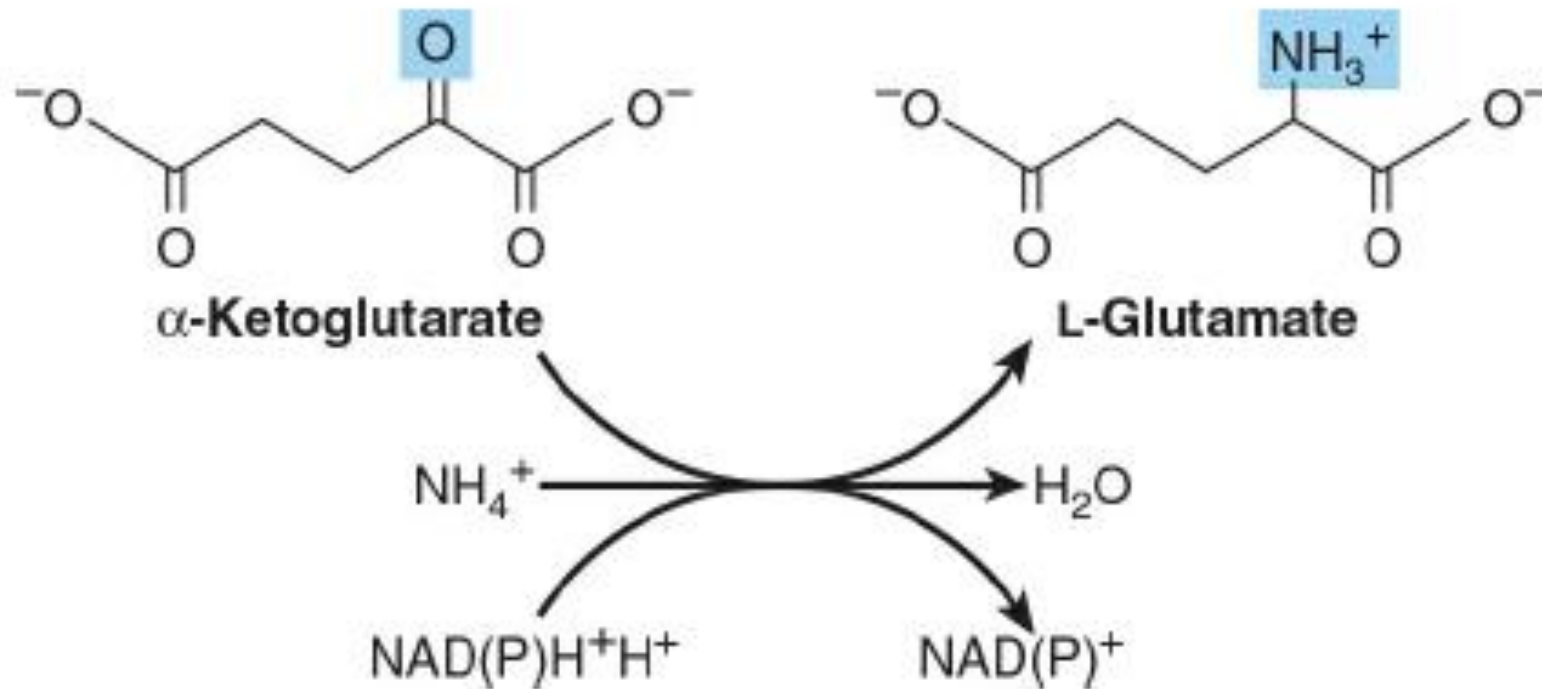
Glutamate

- α -KG can be used to produce glutamate.

- An enzyme called **glutamate dehydrogenase** which uses **vitamin B3 (NAD⁺)** as a **coenzyme**, is responsible for this reaction.

Or NADP

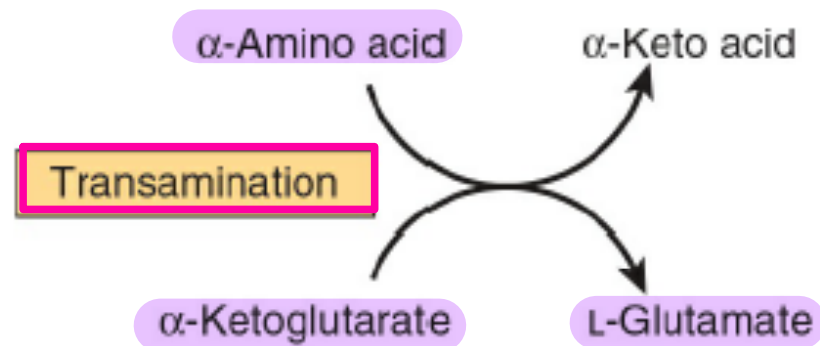
- The **GDH** enzyme is a **mitochondrial enzyme** found primarily in **liver, kidney, and cardiac muscle**, with lower levels in brain, skeletal muscle, and leukocytes.



The reaction catalyzed by glutamate dehydrogenase

Glutamate

- Glutamate can also be synthesized by transamination of α -Ketoglutaric acid (2 oxoglutarate).
- Hence, some of the glucose metabolized by neurons can also be used for glutamate synthesis.



The Glutamate-Glutamine Cycle in the Brain

- Within the CNS there is an interaction between the cerebral blood flow, neurons, and the protective astrocytes that regulates the metabolism of glutamate, glutamine, and ammonia.
- This process is referred to as the glutamate glutamine cycle and it is a critical metabolic process central to overall brain glutamate metabolism.

هون الدكتور راحت على الرسمة بسلايد ٢٢ و شرحت عليها المكتوب بالسلايدات الجاي ف اي اشني زيادة كتبتة عند مكانه بالسلايدات عشان ما انسح كل الحكي عند الرسمة

The Glutamate-Glutamine Cycle in the Brain

Glutamate enters via vesicles by vesicular transporter

1. Using presynaptic neurons as the starting point, the cycle begins with the release of glutamate from presynaptic secretory vesicles in response to the propagation of a nerve impulse along the axon.
 - The release of glutamate is a Ca^{+2} dependent process that involves fusion of glutamate containing presynaptic vesicles with the neuronal membrane.

Calcium is released upon depolarization

The Glutamate-Glutamine Cycle in the Brain

2. Following release of the glutamate into the synapse it must be **rapidly removed to prevent over excitation of the postsynaptic neurons.**
3. Synaptic glutamate is removed by three distinct process. It can be **taken up into the postsynaptic cell**, it can **undergo** **reuptake into the presynaptic cell** from which it was released, or it can be **taken up by a third non neuronal cell, namely astrocytes.**

When glutamate is released into the synaptic cleft it has 3 options » 1. it binds to it's receptors on the postsynaptic neuron either on kainate receptor in NMDA or AMPA resulting with it's excitatory action 2. Reuptake by presynaptic neuron 3. Reuptake by other cells called astrocytes

The Glutamate-Glutamine Cycle in the Brain

- ❖ Postsynaptic neurons remove little glutamate from the synapse and although there is active reuptake into presynaptic neurons the latter process is less important than transport into astrocytes.
- ❖ The membrane potential of astrocytes is much lower than that of neuronal membranes and this favours the uptake of glutamate by the astrocyte.

The Glutamate-Glutamine Cycle in the Brain

- **Glutamate** uptake by astrocytes is mediated by Na independent and Na dependent systems.
- **The Na dependent systems** have high affinity for glutamate and are the **predominant glutamate uptake mechanism** in the central nervous system.
- There are two distinct astrocytic Na dependent glutamate transporters identified as **EAAT 1** (Excitatory Amino Acid Transporter 1) and **EAAT 2**.

The Glutamate-Glutamine Cycle in the Brain

- The outcome of astrocytic glutamate uptake is its conversion to glutamine.
- **Glutamine thus serves as a “reservoir” for glutamate but in the form of a non-neuroactive compound.**
- Release of glutamine from astrocytes allows neurons to derive glutamate from this parent compound.

The Glutamate-Glutamine Cycle in the Brain

- Astrocytes readily convert glutamate to glutamine via the **glutamine synthetase** catalyzed reaction as this **microsomal enzyme** is abundant in these cells.
- Indeed, histochemical data demonstrate that the **glia are essentially the only cells of the CNS that carry out the glutamine synthetase reaction.**
- ❖ The **ammonia** that is used to generate glutamine is derived from either the **blood or from metabolic processes occurring in the brain.**

Why does glutamate get transformed into glutamine? Why it doesn't stay as it is for reuse? 1. For buffering ammonia 2. it must be in the form of glutamine to be reuptaken by presynaptic neurons

The Glutamate-Glutamine Cycle in the Brain

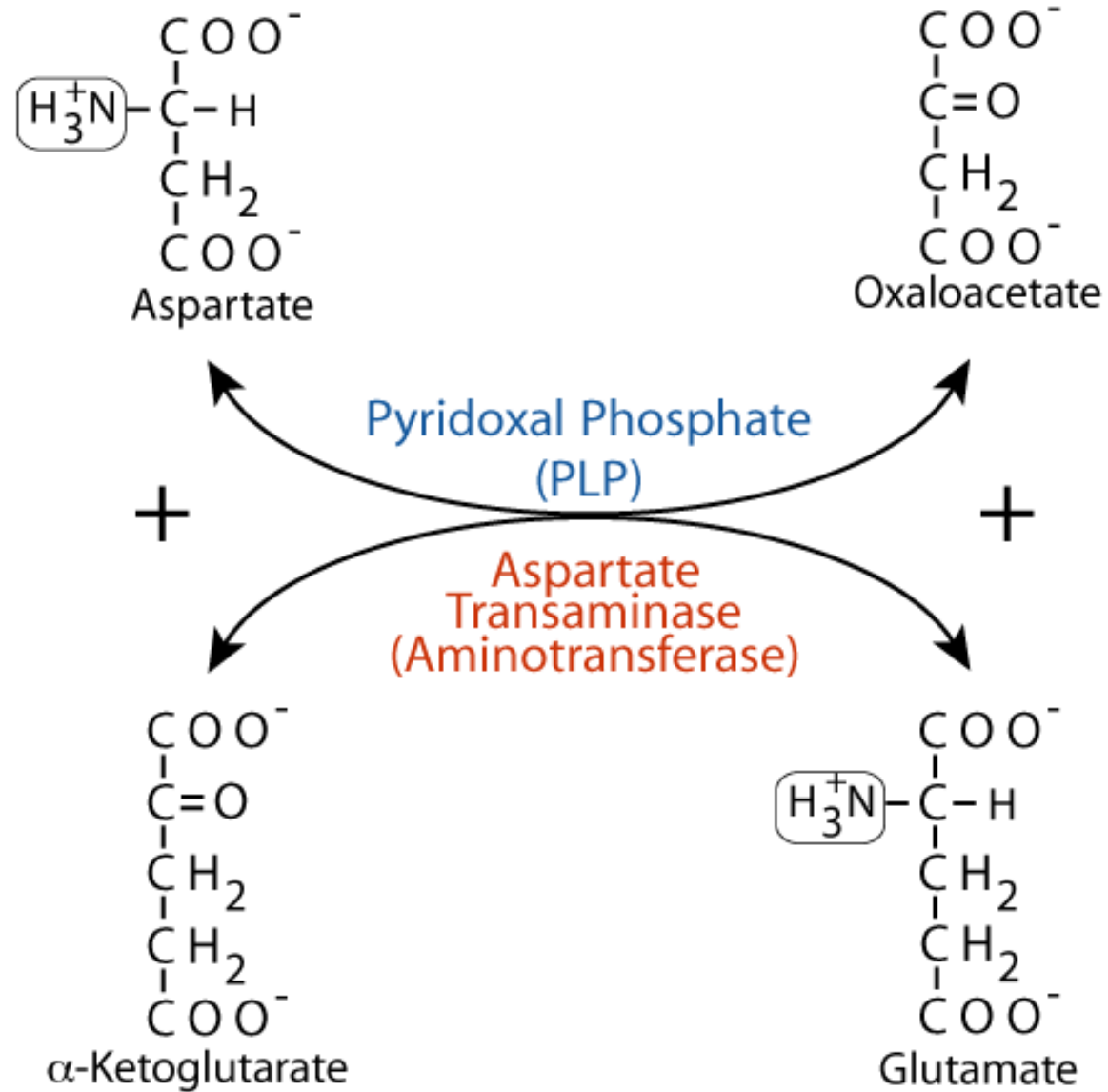
- The predominant metabolic fate of the glutamine taken up by neurons is **hydrolysis to glutamate and ammonia** via the action of the mitochondrial form of glutaminase.
- This form of glutaminase is referred to as **phosphate dependent glutaminase (PAG)**.
- The inorganic phosphate (Pi) necessary for this reaction is primarily derived from the hydrolysis of ATP.
- During depolarization there is a sudden increase in energy consumption.
- The hydrolysis of ATP to ADP and Pi thus **favors the concomitant hydrolysis of glutamine to glutamate via the resulting increased Pi**.

The Glutamate-Glutamine Cycle in the Brain

ATP will be replenished by transformation of glutamate into α -ketoglutarate

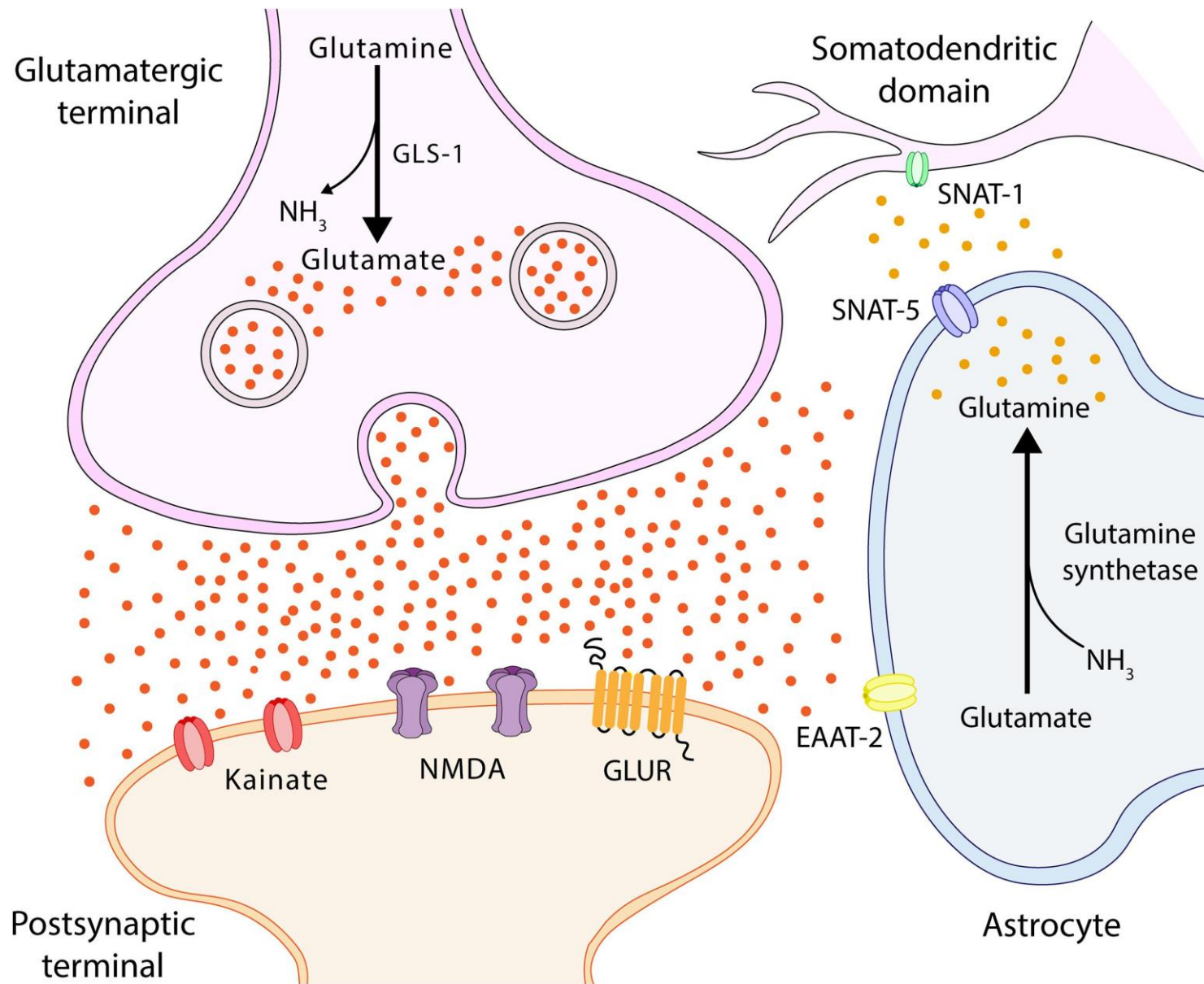
- Because there is a need to replenish the ATP lost during neuronal depolarization, metabolic reactions that generate ATP must increase.
- It has been found that not all neuronal glutamate derived from glutamine is utilized to replenish the neurotransmitter pool.
- A portion of the glutamate can be oxidized within the nerve cells following transamination.
- The principal transamination reaction involves aspartate aminotransferase and yields α -ketoglutarate (2 oxoglutarate) which is a substrate in the TCA cycle.

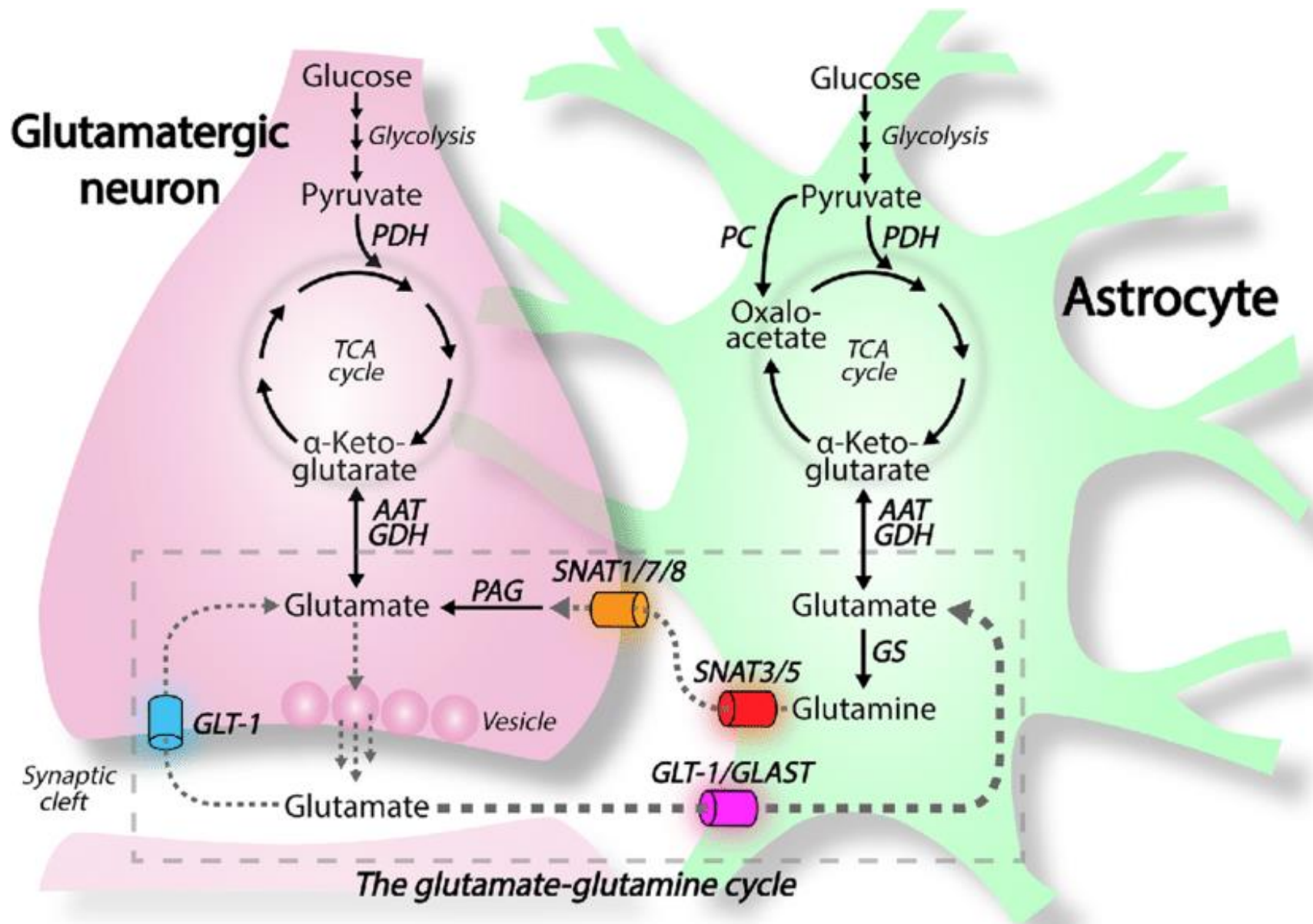
Requires pyridoxal phosphate



The Glutamate-Glutamine Cycle in the Brain

- Glutamine, therefore, is not simply a precursor to neuronal glutamate but a **potential fuel, which, like glucose, supports neuronal energy requirements.**
- Lastly, but significantly, the **incorporation of ammonia into glutamate in the astrocyte serves as a mechanism to buffer brain ammonia.**





Excitotoxicity

- Although glutamate is one of the most abundant neurotransmitters found in the brain, it **exists in very small concentrations**.
- **If the concentration level rises, then neurons become too excited and don't fire in a normal manner.**
- **Glutamate becomes an excitotoxin when it is in excess**; meaning it **overstimulates brain cells and nerves and results in neurological inflammation and cell death.**



Nerve damage

Excitotoxicity

- It is the pathological process by which nerve cells are damaged or killed by excessive stimulation by neurotransmitters such as glutamate and similar substances.
- This occurs when receptors for the excitatory neurotransmitter glutamate (glutamate receptors) such as the NMDA receptor and AMPA receptor are overactivated by glutamatergic storm.

Especially with NMDA receptors » it has high permeability for calcium

Excitotoxicity

- Excitotoxins like NMDA and kainic acid which bind to these receptors, as well as pathologically high levels of glutamate, can cause excitotoxicity by allowing high levels of Ca^{+2} to enter the cell.
 - Ca^{+2} influx into cells activates a number of enzymes, including phospholipases, endonucleases, and proteases.
 - These enzymes go on to damage cell structures such as components of the cytoskeleton, membrane, and DNA.
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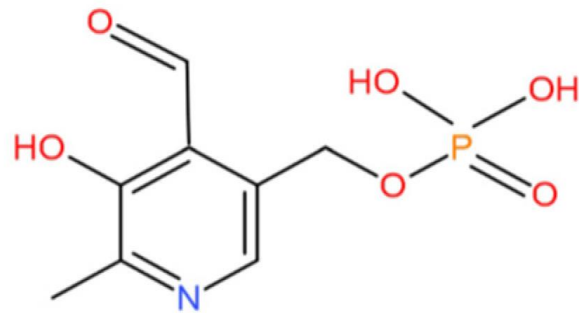
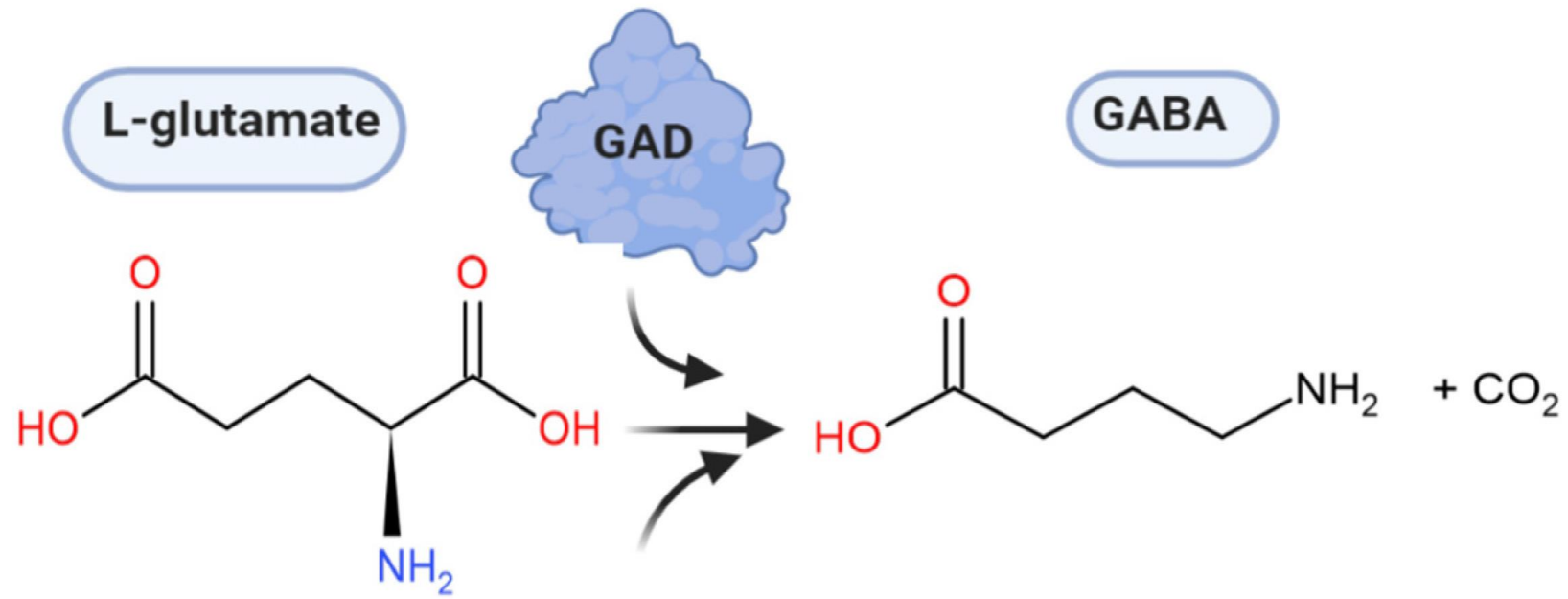


Excitotoxicity

- Glutamate excitotoxicity (sometimes called a glutamatergic storm) refers to the damage to nerve cells caused by excessive stimulation of NMDA and AMPA receptors by glutamate.
 - The excessive rise in the intracellular levels of Ca^{+2} triggers a number of cell damaging processes that ultimately lead to cell death through a process called apoptosis.
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GABA

- GABA is the major inhibitory mediator in the brain.
- GABA is primarily synthesized from glutamate via the enzyme glutamate decarboxylase (GAD) with pyridoxal phosphate as a cofactor.
- This process converts glutamate (the principal excitatory neurotransmitter) into GABA (the principal inhibitory neurotransmitter).



PLP

GABA

- GAD requires vitamin B6 as a coenzyme. Therefore, dietary deficiency of vitamin B6 can lead to diminished GABA synthesis.
- In a disastrous series of infant deaths, it was noted that vitamin B6 was omitted in an infant feeding formula. GABA content in the brain of these infants was reduced. Subsequently, there was a loss of synaptic inhibition that caused seizures and death.

GABA

- After GABA is released from a neuron, a **high affinity GABA transporter allows for its reuptake.**
- **In glia**, GABA is converted to glutamate by a **mitochondrial enzyme, GABA transaminase (GABA T)** as GABA is **metabolized primarily by transamination to succinic semialdehyde** and **then to succinate** in the citric acid cycle.
- **GABA transaminase** enzyme **catalyzes the conversion** of 4-aminobutanoic acid (GABA) and 2 oxoglutarate (α -ketoglutarate) into **succinic semialdehyde** and **glutamate.**
- Succinic semialdehyde is then **oxidized into succinic acid by succinic semialdehyde dehydrogenase** and as such enters the citric acid cycle as a **usable source of energy.**

