Brain Damage due to Excessive Glycine Diet Related to NR2B Protein

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Abstract

Damaged brain will lower the quality of life. Several efforts to prevent brain damage have been performed, including the studies to understand the brain damage mechanisms and to find the drugs to prevent further damages. One of the brain damage mechanisms after the trauma caused by ischemia, infection, etc. is the exitotoxic reaction triggered by excessive Ca^{2+} influx into the neuron. The influx is facilitated by ion channel at neuron, and the performance of this channel is influenced by its subunits, such as NR1, NR2A-D, and NR3A,B. The NR1 subunit helps the opening of the channel after binding Glycine, while the NR2B subunit helps the opening after binding Glutamate. Glycine and Glutamate are ligands working as coagonist to each other. The presence of excessive Glycine is considered to increase the Ca^{2+} influx, which can lead to neuronal death through apoptotic pathway. Therefore, brain-damage patients' intake of foodstuff rich in Glycine should be controlled to prevent them from further damage.

Keywords: NR2B, exitotoxic, glycine

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Kerusakan Otak Akibat Diet Glisin Berlebihan dalam Hubungannya dengan Protein NR2B

Abstrak

Kerusakan otak mengakibatkan turunnya kualitas hidup seseorang. Berbagai usaha untuk mencegah kerusakan otak telah dilakukan, mulai dari mempelajari mekanisme kerusakan otak sampai dengan mencari obat yang dapat mencegah kerusakan lebih lanjut. Salah satu mekanisme kerusakan otak, setelah terjadi trauma pada otak akibat iskemia, infeksi, dsb. adalah reaksi eksitotoksik yang dipicu oleh influks Ca^{2+} berlebihan ke dalam neuron. Influks ini difasilitasi oleh saluran ion pada neuron, dan kinerja saluran ion ini dipengaruhi oleh komposisi subunit penyusunnya, yang antara lain terdiri atas NR1, NR2A-D, dan NR3A,B. NR1 merupakan subunit yang akan turut membuka saluran ion setelah mengikat Glisin, sedangkan NR2B turut membuka saluran ion setelah mengikat Glutamat. Glisin dan Glutamat merupakan ligan yang bekerja sebagai koagonis satu terhadap yang lain. Kehadiran Glisin yang berlebihan diperkirakan meningkatkan influks Ca^{2+} dan dengan demikian dapat menyebabkan kematian neuron melalui jalur apoptosis. Oleh karena itu, bahan makanan kaya Glisin perlu dipertimbangkan untuk dihindari oleh penderita kerusakan otak.

Kata kunci: *NR2B, eksitotoksik, glisin*

Introduction

Most of brain functions are based on ionic transport mechanisms at neuron. This transport is facilitated by ion channel. This channel may consist of some subunits, each may also be receptor for certain ligand controlling the channel opening. Ion channel for Ca^{2+} may contain receptor protein (named NR2B) for a synthetic chemical which resembles Glutamate, named NMDA (N-Methyl-D-Aspartate). The Ca^{2+} channel can be opened by binding certain ligand, in this case, Glycine at NR1 subunit and Glutamate (or NMDA) at NR2B subunit.[1]

Studies concerning the ligands of NMDA receptors (NMDARs) can find their application in controlling exitotoxic reaction caused by its excessive activation at some chronic and acute neurodegenerative disorders. [1] Figure 1 shows some pathways where brain damage can even be worse through cellular response laboring NR2B-containing ion channel which permits excessive Ca^{2+} influx, followed by steps resulted in cell death.

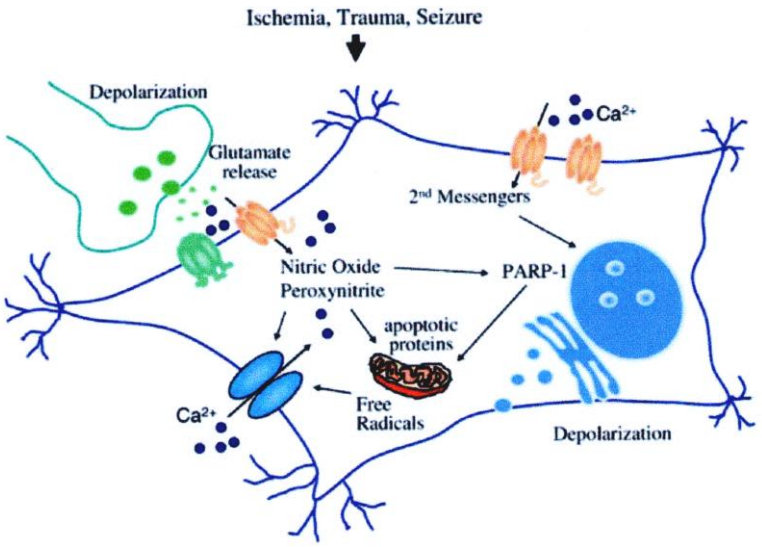

Figure 1. Intracellular Events that Leads to Cell Death after Ischaemia, Trauma, or Epilepsy.[2] PARP = poly(ADP-ribose) polymerase.

After a brain injury, acute neuronal damage follows, which involves excitotoxicity, inflammation, necrosis, and apoptosis (the last two mean the cell death)[2]; the injury activates NMDA receptor (the NR2B protein).[3] These processes start when cerebral blood flow is impaired after such an injury, followed by the impairment of ionic gradients. Membrane potential is lost and neuron depolarizes, then excitatory amino acids are released and bound to their receptors to start excitotoxic steps/cascades leading to cell death. In this case, enzymatic degradation is activated, peroxynitrite (a free radical) is formed, disruption of normal mitochondrial function (causing oxidative stress) takes place, and activation of pro-apoptotic protein named PARP-1 occurs.[2]

The finding of new neuroprotective drugs that work as an antagonist of NMDA-type and Glutamate receptor, and can be tolerated by human body, gives hope to deal with further brain damage processes after pathological attack.[4]

Finally, proper knowledge about food constituents can be useful to evaluate how save they are, in relation to *NR2B* gene expression. It is known that soybean extract of 40-50% calori caused moving disorder at testing animals; the cerebellum was damaged with 18% weight decrease.[5] The high Glycine containing food becomes the focus of this paper.

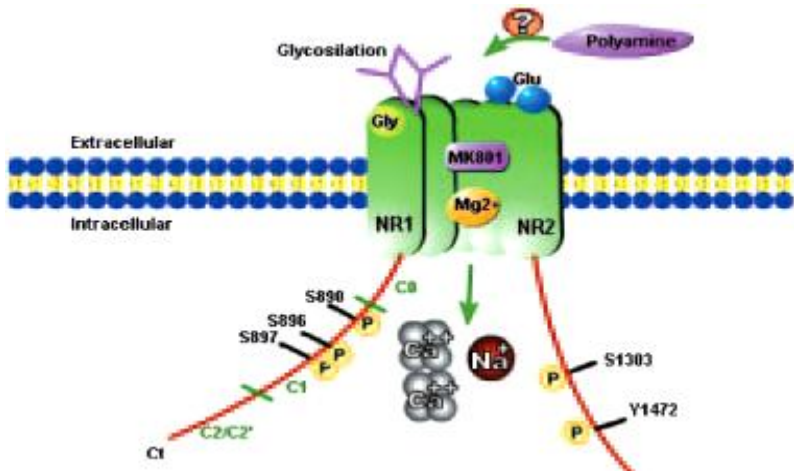

Figure 2. The Schema of NMDA Receptors Construct an Ion Channel. Some Binding Sites of Agonist, Antagonist, and Modulator Molecules, also Phosphorylation and Glycosilation Sites are Shown. [9]

The NR2B Protein

NR2B is a Subunit of Ca^{2+} Ion Channel

NMDA Receptors (NMDARs) may consist of the subunit NR1, subunit NR2 (NR2A, NR2B, NR2C, NR2D), and NR3 (NR3A, NR3B).[6] This NMDA Receptor ion channel is very permeable to monovalent ions and Ca^{2+}.[1]

The Expression and Distribution of NR2B

Each subunit is expressed both overlappingly and specifically at certain specific location in brain.[7] The predominant NR2 subunits found at adult neocortex and hippocampus are NR2A and NR2B. NR2A is found predominantly at adult synaps, while NR2B is mainly distributed extrasynaptically. NR1 subunit must be present at NMDA receptor and it interacts with subunit NR2A to D, resulting in various functions.[8] The schema of ion channel laboring NR1 and NR2 can be seen in Figure 2.

The Two Main Roles

The first role of NR2B is in learning and memory processes. Learning is defined as the mechanism of how new information is gained, while memory is the mechanism of retaining that.[10] NMDA Receptors have important roles in neuron plasticity, learning, memory, even in brain development. Basically, they are the result of the high permeability of Ca^{2+}.[1,8] Hippocampus is a part of brain important in learning process since NR2B is found abundantly in this part.[8] Besides that, the integrity of connections among hippocampus, subiculum, and cortex areas is important in the synthesis of all components in spatial learning.[10] The second main role

of NR2B is its involvement in brain pathological condition, which results in neuron death, something to be explained further next in this paper.

Two events are needed to open the channel

NMDA receptor, the NR2B, is in the family of ionotrophic Glutamate receptor.[6] The channel can be blocked by Mg2+ which is opened after depolarization and the binding of agonist.[1]

The Roles of Glycine in Brain

One of food components is amino acid Glycine, which is amino acid constituting both protein and neurotransmitter. It can also function as ligand, the coagonist of Glutamate, that opens the Ca^{2+} ion channel at neuron. In short, Glycine as important chemical plays some roles in normal and pathological functions in brain. The discussion will begin with its metabolism in brain, followed by step by step description of how it eventually plays its role in neuron death.

Glycine Metabolism in Brain

Despite the damage it may cause, Glycine is absolutely needed in brain, where it acts as neurotransmitter and neuromodulator. Glycine is taken up and consumed quickly in brain tissue; astroglial cell metabolizes it into Serine and Lactate. The Glycine metabolism pathway involves SHMT (Serine Hydroxymethyltransferase) enzyme and GCS (Glycine Cleavage System) (Figure 3). Besides Glycine from outside, brain itself produces Glycine.[11]

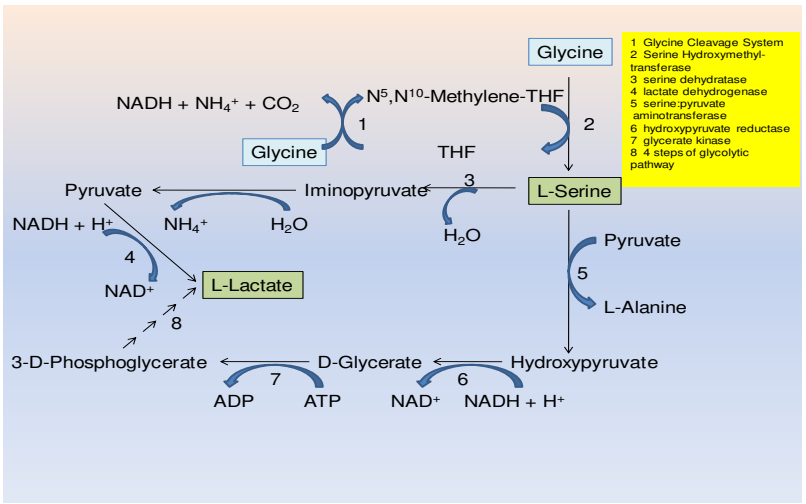

Figure 3. Metabolic Pathway of Glycine in Brain[11]

Note: Glycine intake is followed by the production of its metabolic products (L-Lactate and L-Serine)

The Two Glycine Binding Sites

Brain has two Glycine binding sites, at least. There are Glycine receptor at ion channel for Cl^- in marrow and brain stem; this channel works after binding the ligand. The next is at NMDA receptor. The binding of agonist ligan is needed to open the ion channel.[11]

Interaction of Glycine and Glutamate in Brain

Both Glutamate and its coagonist, Glycine, are needed in opening the ion channel and synaptic activation of NMDA receptors. The allosteric interaction happens between the Glutamate and Glycine binding sites. Simultaneous binding of Glutamate and coagonist Glycine is needed for the activation of NMDARs.[1]

Neuronal Cell Damaging Mechanism

Under pathological condition, following the overactivation of NMDAR, excessive Ca^{2+} influx causes Ca^{2+} overload in mitochondria which results in the formation of oxygen free-radical, activation of caspases, and the release of apoptosis-inducing factor. Besides that, the production of NO (nitric oxide) and $ONOO^-$ (peroxynitrite) increases as the result of Ca^{2+}-dependent activation of neuronal nitric oxide synthase (nNOS). The last is the activation of transcription factors through the stimulation of p38 mitogen-activated protein kinase (p38 MAPK), resulting in neuronal injury and apoptosis. Fig.4 summarizes all.[12]

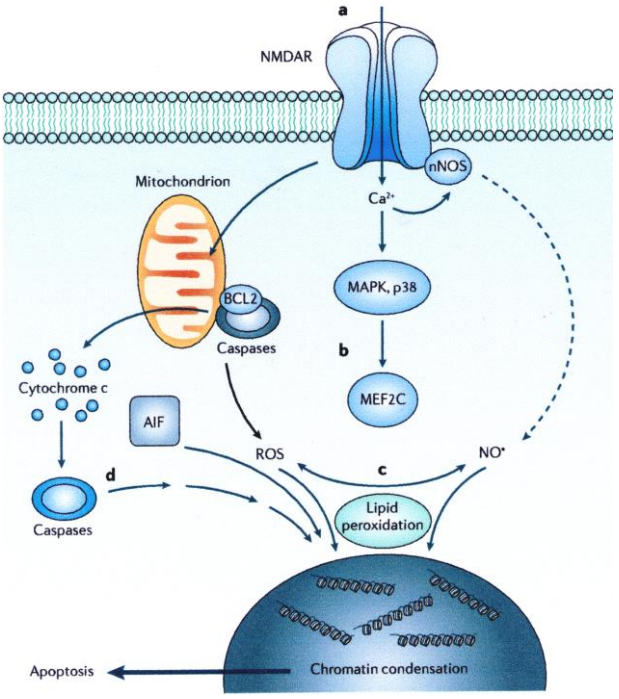

Figure 4. The Schema Of The Apoptotic-Like Cell Injury And Death Pathways Triggered By Excessive NMDAR Activity[12]

Glycine, NMDAR, and Some Brain Diseases

Glycine activates NMDA receptors at neuron; the prolonged activity of NMDA receptors (NMDARs) can happen in the administration of NMDA agonist, like Glycine.[6] This prolonged stimulation results in high influx of Ca^{2+} which causes sitotoxicity ended by cell death in ischemia and other neurodegenerative disorders.[1] Besides, the excessive stimulation takes role in chronic neurogenerative condition, such as in Alzheimer disease, Parkinson, Huntington disease, and dementia, associated with HIV or lateral amyotrophic sclerosis.[8,13] In model animal suffering Glycine encephalopathy, Glycine consentration is high in the blood and cerebrospinal fluid, significantly high in brain tissue. Most patients with this disorder will die soon after birth, while the survivors will suffer neurological problems, like psychomotoric retardation and convultion. In high concentration of Glycine, oral medication using NMDAR antagonist is successful in recovering the patients condition.[14]

Serine can replace Glycine

The influence of Glycine can be observed at some concentrations (100 μM or more), hundreds of time above the dissociation constant of Glycine from NMDA receptors, while D-Serine, the agonist at Glycine binding site is proven to be effective in lower micromolar range.[15]

High Glycine Food Diet

As one of amino acids that constitutes proteins, our protein diet inevitably may contain Glycine. This fact does not say that all kinds of food are the same, instead each kind has its own benefit and risk for the consumers. Soybean (*Glycine max* L.) is relatively high in Glycine content, in compare to some other food stuffs (Table 1); tempeh and soybean cake are among the foods processed from soybean. These Indonesian traditional foods can be found almost anywhere in this country.

Table 1. The Content of Some Neurotrasmitter Amino Acids in Some Food Sources (Per 100 g)[16]

Amino acid	Unit	Soybean	Egg	Beef	Oyster mushroom (raw)
Gly	g	0.77	0.423	1.702	0.1
Glu	g	3.224	1.644	4.197	0.5
Ser	g	0.965	0.936	1.101	0.09
Ala	g	0.784	0.700	1.700	0.18
Asp	g	2.093	1.264	2.547	0.215
Cys	g	0.268	0.292	0.361	0.02

The conversion of human consumption of soybean products has been performed by Firmansyah (2009, unpublished) using soybean cake as a model, with conversion into soybean seed. The result showed that 30 day consumption of soy seed by mice, equal to 62.5 g of the seed for human showed no statistically significant difference from control group which consumed only the half. Despite this, there is a trend of *NR2B* gene expression and brain weight decreases. These trend could be the result of the decrease of brain mass due to neuronal cell death.[5] Excitotoxic reaction is most probably responsible for the death, with the significant NR2B activation by coagonist Glycine.[11] Anyway, high risk patients should avoid any food stuff known to be high of Glycine, in the way of reducing the brain damage progress.

Conclusion

One of the vital organs in human body, even in all animals, is brain. Its damage can be devastating to human`s quality of life. Therefore, efforts protecting the brain from any damage and further damage have to be done. Studies have been in progress for better treatments. The basic mechanisms of brain-further-damage are mostly rooted on molecular base at neuronal cell surface and the following steps. Here, the role of Ca^{2+} ion channel takes the vital role. As the subunit composition of this ion channel varies, efforts are directed to the control of subunit protein activities, even to their gene expression. In this case, protein NR2B as one of the subunits becomes the focus, since it directly influences the channel opening by binding the ligand Glutamate (or NMDA), together with the binding of its coagonist ligand Glycine at subunit NR1. Both ligands work allosterically. Consequently, activity control of NMDAR can also be done by the control of ligand number. New drugs are in queue to emerge, with minimal side effects on brain normal functions. The combination of efforts to minimize brain damage have to be run simultaneously. Then Glycine becomes the focus, since the proper consumption of this neurotransmitter amino acid and the diet may contribute to slowing down of the brain damage progress.

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