

The Cell Regulation Mechanism of Neurovascular Unit

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Abstract: Ischemic cerebrovascular disease is one of the three deadly diseases. It is characterised by high mortality and high morbidity. Because of no effective treatments of recombinant tissue plasminogen activator (rt-PA) and neuroprotectant, there are more and more research focus on neurovascular unit (NVU), which is composed of brain microvascular endothelial cells (BMECs), neuron, astrocyte(AS) and so on. Cell-cell signaling and coupling between these different compartments form the basis for normal function and repair of brain injury. In this mini-review, we will describe the relationship of CMECs, neuron and AS.

Keywords: Neurovascular unit, Brain microvascular endothelial cells, Neuron, astrocyte.

INTRODUCTION

Ischemic cerebrovascular disease is caused by focal cerebral dysfunction caused by ischemic of vessels. After stroke, microglia induce the releasing of proinflammatory cytokine and adhesion factor, it also can induce the happening of inflammation, breaking Tight junction protein and increasing permeability of blood brain barrier (BBB). Neuron and glial cells begin to die, astrocytes and neurons is virtually all died in 7-14 days [1]. But right now, the therapy of Ischemic cerebrovascular disease (ICVD) is restricted by target and therapeutic window. The thrombolytic agents such as tissue plasminogen activator (t-PA) are available to treat patients with an acute ischemic stroke, but it must be used between 3 hours and 4.5 hours after stroke, or it may make encephaledema become severe [2]. In the view of the fact that the treatment focus on single target not effect enough, there are more researchers pay more attention to research on protection of BMECs, neuron, AS, microglia, pericyte basement membrane and extracellular matrix. The concept of NVU emphasized the importance of overall research is put forward. It provides a conceptual framework for the study of the connection between neurons, glial cells and vascular endothelial cells. In this paper we will review the relationship between neuron, BMECs and AS, we hope this review will provide a new train of thought to the research of protecting NVU.

THE STRUCTURE AND FUNCTION OF NERVE VASCULAR UNIT

NVU composed mainly of neurons, AS, BMECs and extracellular matrix, pericyte and other support cells

such as microglia, etc. [3]. The important members of NVU are neurons, AS, BMECs. AS and BMECs and is involved in the formation of blood brain barrier. Under normal physiological conditions, BBB strict control the substance to enter the brain, maintain the ionic balance of the brain, maintain normal physiological functions of neurons, promote the input of nutrient and the output of the metabolites, and can prevent the harmful substances into the brain and accumulate in the brain. BMECs is the main element and structure foundation of the BBB [4]. The tight junction between the BBB is the main material basis of BBB physiological functions [5,6].

AS is the most number of parenchyma cells in the brain, it is divided into virgin pulp AS and fibre AS, they differ in morphology, distribution, but are same with in basically function. Many branches of astrocytes and pseudopodia wrapped around the BMECs and neurons, they are an important part of the function of the BBB, at the same time, and they have function of supporting isolation, insulation, participating in ion transfer and neurotransmitters, hormones and metabolism, and so on [8]. The neuron complete brain traditional functional activity and it has fast signal processing, and transfer the signals to each other or the effector cells [9].

After cerebral ischemia occurs, BMECs can secrete a variety of factors, such as prostaglandins, endothelial diastolic factor to protect nerve cells, promote the survival of the neuron [10]. When brain damage occurs, the BMECs secretion of VEGF to prompt the differentiation of neural stem cells to neurons, and the newborn blood vessels provide nutrition and oxygen for neuron of ischemic penumbra, and provide nutrients for nerve regeneration and synaptic regeneration [11]. In recent years, a number of studies have shown that,

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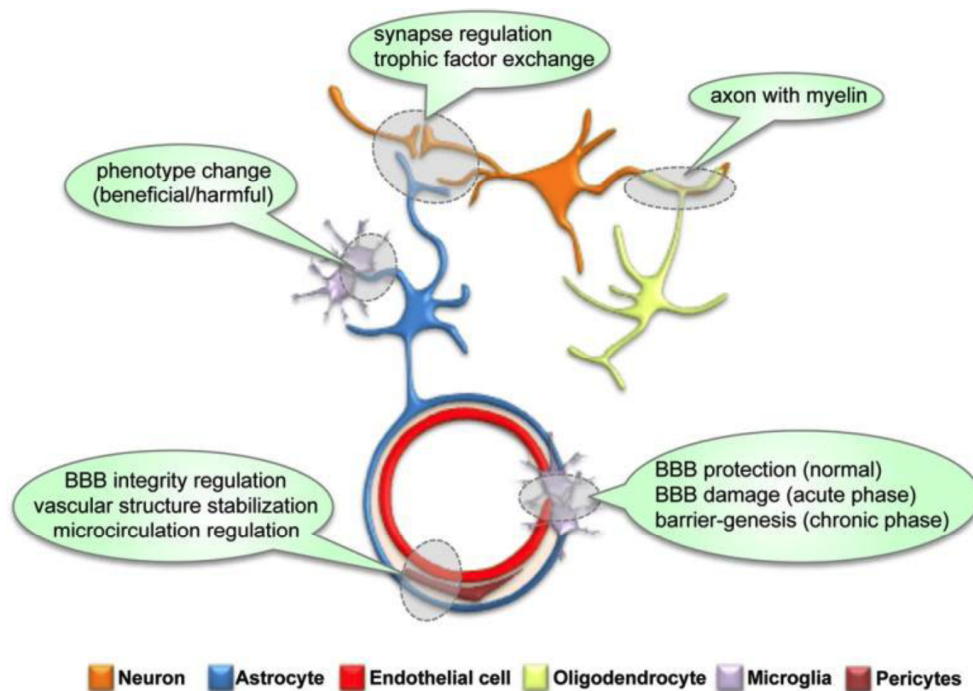


Figure 1: Schematic of the neurovascular unit [7].

astrocytes provide nutritional support for neurons, transfer signal and guide the growth of synapses [12,13]. When neuron damage occurs, AS can fight oxidative stress to protect the neurons. AS can induce the formation of tight junction in endothelial cells, participate in endothelial cell signal transduction, and regulate the blood flow [14]. Endothelial cells can induce the differentiation of astrocytes, when blood anoxic injury occurs, the endothelial cells can increase the oxidative stress of astrocytes.

2. THE INTERACTION BETWEEN NEURONS, BMECs AND AS

2.1. The Interaction between Neurons and BMECs

2.1.1. BMECs to Promote Proliferation of Neurons and Protection Neurons

BMECs can release some soluble factors, including interleukins, interferon, colony stimulating factor, platelet-derived growth factor, transforming growth factor, insulin-like growth factor, basic fibroblast growth factor (bFGF), vascular endothelial growth factor (VEGF), Brain derived neurotrophic factor (BDNF) to promote the survival of the nerve cells [15-17]. The bFGF is a kind of neurotrophic factors, can promote the proliferation and migration of neural stem cells, and with other nutritional factors jointly promote the differentiation of neural stem cells. BDNF is mainly secreted by endothelial cells, effects on neurons,

promotes the survival of the neurons, increase prominent plasticity and neurogenesis. After combination of BDNF and TrkB, intracellular area is activated. It causes TrkB itself phosphorylation, activating the Ras - MAPK pathway, finally activating CREB in CAMP response element binding protein serine loci. CREB play a role by increasing the BDNF gene and anti-apoptosis protein gene expression of BCL - 2 [18]. Under the anaerobic environment, cerebral microvascular endothelial cells secretion of VEGF increased [19], it have direct neuroprotective effects, can stimulate the growth of axons and improve the survival of the nerve cells. Studies have shown that after VEGF role in nerve cells 24h, the expression neural specificity protein increased, such as calcium binding protein, nestin, glial fibers acidic protein (GFAP), etc. VEGF can promote the growth of synapses [20]. In addition to VEGF can increased vascular permeability, promoting angiogenesis, maintain blood vessel function, repairing damage blood vessels, studies have shown that it have effect on stem cells, can protect neural stem cells to survive in anoxic condition [21]. Huang et al. trained BMECs with neural stem cells, found that the number of neural stem cells is directly proportional to the time, and after trained 24h, the content of VEGF in medium markedly increased. After giving VEGF antagonists, the proliferation of neural stem cell was decreased, they guess the BMECs may through the secretion of VEGF

to promote the proliferation of neural stem cells [22]. In addition, neural stem cells can also be collaborative vascular system stimulate angiogenesis and synaptic reconstruction to promote nerve repair [23].

2.1.2. BMECs Induce Neuron Differentiation

Neurons was considered the most important brain cells, it is responsible for processing information quickly and delivery information to neurons and other cells [9], therefore, it is important that restore the function of neurons supplement the number of neurons as soon as possible after the neuron. According to traditional opinion, the neurons are non-renewable, when neurons damaged and even death only through differentiation of neural stem cells to compensate for the damage. Neural stem cells are neuronal precursor cells. After stroke in the adult animals' brain, the number of neural stem cells will increase rapidly, and they are concentrated at the edge of the damaged region [24]. Neural stem cells can differentiate into astrocytes, neurons and oligodendrocytes, and grow into other cells rate is greater than the differentiation to neurons. Other substances are required for neural stem cells differentiation to other cells. In current, there are two methods are used to induce neural stem cells differentiate to neurons has two kinds: one is to add the factors associated with neurogenesis, such as VEGF, BDNF, etc. Second one is cell culture, neural stem cells were co-cultured with glial cells or BMECs, etc., and it is better that neural stem cells were co-cultured with BMECs [25-28]. BMECs promote differentiation and proliferation of neural stem cells by the secretion of bFGF, BDNF and VEGF.

2.1.3. Neurons Promote Angiogenesis

Angiogenesis is one of the body's self-repair mechanisms after cerebral ischemia. There are some factors that can promote angiogenesis: VEGF, PDNF, angiotensin (Ang) and nitric oxide (NO). The VEGF is recognized as the most critical factor in the process of AG. And it continuous high expression in the process of AG, VEGF and its receptors can promote endothelial cell proliferation, adhesion and migration, start AG [29]. The central nervous system can secrete VEGF in Embryonic period, which can induce the formation of blood vessels [30]. After brain injury, neurons and neural stem cells can also promote angiogenesis by VEGF. PDGF is mainly expressed by neurons, PDGF bind with its receptor to induce endothelial cells and pericyte involved in vascular remodeling and repair process after ischemia. Thus, angiogenesis and nerve regeneration have mutual promoted effects.

2.2. The Interaction between Astrocytes and Neurons

2.2.1. Astrocytes Support Neurons with Nutrition and Protect Neurons

Astrocytes maintained steady state of the brain, and provide a good environment for the brain reconstruction [31]. Neurons can't provide energy by itself, but by astrocytes to get energy supply. Astrocytes are the main storage area of neurons glycogen. When the anoxic injury occurs, astrocytes can convert glucose into pyruvic acid salt, and then converted to lactic acid salt secrete cell provides energy for neurons. Astrocytes secrete glial cell derived neurotrophic factor (GDNF), BDNF, bFGF, ciliary neurotrophic factor, laminin, fiber adhesion protein and insulin-like factor and so on, these factors have a nutritional effect on neurons, can promote the survival of the neurons, help neuronal cell metabolism [12]. The GDNF can continue to increase after brain ischemia, inhibit expression of casepase-3 to protect the neurons. BDNF is important for neuronal survival, differentiation, growth and development, and can prevent nerve damage, promote the repair, regeneration and differentiation of neurons, it is necessary to neuron survival and maintain normal physiological function. Experiments show that dopamine neurons 9d can survive alone, but after 10d, if neurons without nutritional support of astrocytes, neurons will quickly death [31].

Astrocytes have longer synapses, they woven mesh in the brain, the mesh support neuronal cell bodies and fibers and support the neurons migrate to the lower part of the, and guide the growth of neurons axon [13]. When neurons damage, such as hypoxia ischemia, astrocytes can anti oxidative stress to protect neurons, but also can remove oxygen free radicals. Astrocytes can protect the neighboring neurons out from NO, H₂O₂, ultra oxygen anion attack. And when the damage occurs, the astrocytes secretion IL - 1, IL - 2 to induce nerve regeneration [33], astrocytes can also be converted into neurons, so as to substitute damage and even death neurons.

2.2.2. Astrocytes Regulate Neuronal Metabolism and Signaling

Neurons in the brain is divided into glutamate neurons and GABA neurons, when they excited, they will can release glutamate and neurons GABA respectively, both of them can interfere with synaptic transmission, astrocytes can metabolize both of them to glutamine and hand over them to neurons, the glutamine can be used to transmitters precursor substances to make glutamate and neurons GABA,

ensure the normal delivery of information. Between astrocytes and neurons signaling is a two-way street. Astrocytes can regulate neurons by synaptic activity, mainly through the neurotransmitters and concentration of Ca^{+} in the astrocyte cause the changes of synaptic transmission. At the same time, there are many neurons receptor on astrocyte, when the receptors combine with neurotransmitters, it can produce a series of physiological reaction of astrocytes. In addition, astrocytes are rich in clearance connections and a lot of ion channels, they can adjustable neurons inside and outside the ion concentration and PH, especially to the potassium ion, and thus for the signal conduction.

2.3. The Interaction between Astrocytes and BMECs

2.3.1. Astrocytes Promote BMECs Proliferation and its Effect on Promoting Angiogenesis

There are more than 99% astrocytes foot package of the blood vessels, thus to signal transduction and other activities. Astrocytes mainly through the following two ways to influence endothelial cells: one way is the long-distance cell communication; the other is communication between cells [34]. We found astrocytes on BMECs growth mainly played the negative role *in vitro*, but played a positive role on keep BMECs specific functions. Chen Weizhao *et al.* [14] found astrocytes can induce the formation of tight junction and expression endothelial cell transport molecules, such as glucose transporters-1, through co-culture BMECs and astrocytes. And Stewart [35] and fellow found the astrocytes culture with endothelial cells, astrocytes can promote the formation of endothelial cells, and promote vascularization.

Astrocytes through establish close tight junction with capillary endothelial cells, make it directly and quickly perception changes of microenvironment of neurotransmitter, affects synaptic activity, regulate blood vessels and blood flow [36]. Ischemia injury could finally brought astrocyte foot swell, pressure vessels, make a poor blood flow and leads to more massive cell death. At this point, astrocytes can regulate the blood flow by adjusting the contraction of blood vessels.

2.3.3. BMECs Induce Astrocytes Differentiation and Protection it

The endothelial cells play an important role in expression of characteristics of astrocyte [37, 38]. The endothelial cells secrete factors can induce astrocytes differentiation, such as leukemia inhibitory factor. When anoxia/reoxygenation injury occurs, the interaction between endothelial cells and glial cells enhance the

vitality of the antioxidant enzymes, as well as increase the capability of the astrocyte oxidative stress [39].

To summarize, the factors play an important role in NVU physiological process and in the process of repair, such as VEGF, BDNF, BFGF and GDNF. VEGF is secreted by many cells, which can promote endothelial cell proliferation, migration and the formation of blood vessels, but also can promote the proliferation and differentiation of neural stem, promote the survival of the neurons and synaptic growth. BDNF has a direct role in nutrition of neurons, plays an important role in growth and survival of neurons. The bFGF can promote the proliferation, migration and differentiation of the neural stem cells, protect neurons, at the same time can promote BMECs proliferation and migration, maintain the integrity of the newly formed blood vessels. GDNF secreted by astrocytes, have nutrition effect to neurons, and can protect the neurons by inhibiting neuronal apoptosis.

NVU is consisted by neurons, BMECs and astrocytes, the members' interaction, participation brain normal physiological process and repair process after injury. Therefore, if we understand the mutual influence between each member's secretion of some cytokines, it will be good for looking for effective therapeutic targets, and it has a great significance for treatment of ischemic brain injury.

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