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Astrocyty u neurodegenerativních onemocnění
Astrocytes in neurodegenerative disorders

Bakalářská práce

Školitelka: Ing. Miroslava Anděrová, CSc.

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Prohlášení:

Prohlašuji, že jsem závěrečnou práci zpracovala samostatně a že jsem uvedla všechny použité informační zdroje a literaturu. Tato práce ani její podstatná část nebyla předložena k získání jiného nebo stejného akademického titulu.

V Praze, 7.1.2020

Podpis

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Abstract

Astrocytes are the most abundant glial cells in a mammalian brain. They play an important role not only under physiological conditions, but also during pathological changes. They are involved in miscellaneous functions in a healthy tissue, such as: structural and developmental function, homeostasis maintenance, metabolic support for neurons, or reduction of oxidative stress. In a damaged brain, however, their activity is altered. The most common astrocytic changes in a diseased or injured brain/spinal cord are known as reactive astrogliosis and glial scar formation. Other alterations like cellular atrophy, membrane transporters impairments, or over-expression of certain astrocytic proteins may occur as well. These morphological and physiological changes often lead to an increased excitotoxicity which is one of the factors involved in neurodegeneration. This thesis discusses the astrocytic changes during selected neurodegenerative diseases, namely: Alzheimer's disease, amyotrophic lateral sclerosis, Huntington's disease and Parkinson's disease.

Key words: brain, astrocytes, reactive astrogliosis, homeostasis, neurodegeneration, Alzheimer's disease, Parkinson's disease, Huntington's disease, Amyotrophic lateral sclerosis

Abstrakt

Astrocyty jsou nejpočetnějšími gliovými buňkami v savčím mozku. Hrají důležitou roli nejen za fyziologických podmínek, ale také při patologických změnách. Jsou zapojeni do různých funkcí ve zdravé tkáni, jako jsou: strukturní a vývojová funkce, udržování homeostázy, metabolická podpora neuronů nebo redukce oxidačního stresu. V poškozeném mozku se však jejich aktivita mění. Nejčastější astrocytické změny v nemocném nebo poraněném mozku/míše jsou známé jako reaktivní astroglióza a tvorba gliálních jizev. Mohou se vyskytnout i jiné změny, jako je buněčná atrofie, poruchy membránových transportérů nebo nadměrná exprese určitých astrocytických proteinů. Tyto morfologické a fyziologické změny často vedou ke zvýšené excitotoxicitě, která je jedním z faktorů zapojených do neurodegenerace. Tato práce pojednává o astrocytických změnách během vybraných neurodegenerativních onemocnění, a to: Alzheimerovy choroby, amyotrofické laterální sklerózy, Huntingtonovy choroby a Parkinsonovy choroby.

Klíčová slova: mozek, astrocyty, reaktivní astroglióza, homeostáza, neurodegenerace, Alzheimerova choroba, Parkinsonova choroba, Huntingtonova choroba, amyotrofická laterální skleróza

List of abbreviations

Aβ	- β-amyloid
AD	- Alzheimer's disease
ALDH1L1	- aldehyde dehydrogenase 1 family
ALS	- amyotrophic lateral sclerosis
ANLSH	- astrocyte neuronal lactate shuttle hypothesis
AQP4	- aquaporin 4
ATP	- adenosine triphosphate
CA	- carbonic anhydrase
CNS	- central nervous system
COX-2	- cyclooxygenase
Cx30	- connexin 30
Cx43	- connexin 43
EAAT	- astrocytic excitatory amino acid transporter
fALS	-familial amyotrophic lateral sclerosis
FDH	- 10-formyltetrahydrofolate dehydrogenase
GABA	- gamma-aminobutyric acid
GFAP	- glial fibrillary acidic protein
GLAST	- glutamate-aspartate transporter
GLUT1	- glucose transporter
GS	- glutamine synthetase
GSH	- glutathione
γGT	- γ-glutamyl transpeptidase
HD	- Huntington's disease
iNOS	- inducible nitric oxide synthase
Kir	- inwardly rectifying potassium channels
LDHA	- lactate dehydrogenase isoenzyme A
LDHB	- lactate dehydrogenase B

LPS	- lipopolysaccharide
MCT-1/2/4	- monocarboxylate transporters 1/2/4
NBC	- Na+-HCO3- cotransporter
NFT	- neurofibrillary tangles
NG2 cells	- polydendrocytes
NHE	- sodium-hydrogen exchange
NO	- nitric oxide
PD	- Parkinson's disease
ROS	- reactive oxygen species
S100B	- calcium and zinc binding protein, astrocytic marker
sALS	- sporadic amyotrophic lateral sclerosis
SN	- substantia nigra
SOD1	- copper-zinc superoxide dismutase
TGFβ1	- transforming growth factor-β1
TNF-α	- tumor necrosis factor α
TNFR1	- tumor necrosis factor receptor 1

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Introduction

A couple of decades ago the neurobiological research was focused almost exclusively on neurones. Only recently were the neurocentric concepts compromised by novel views on this matter, promoting the importance of glial cells in both physiological conditions and in the course of brain pathologies.

Glia, previously considered only as components with passive, mostly structural functions, are now believed to be substantially involved in all sorts of mechanisms accounting for healthy functioning of central nervous system (CNS). Astrocytes, in particular, represent cells that play a crucial role in the maintenance of brain homeostasis. Furthermore, they are involved in neuronal metabolic support (Gandhi et al., 2009), antioxidant protection against oxidative stress (Makar et al., 1994), and provide for many other CNS functions. Based on this knowledge we may assume that astrocytes essentially contribute to all sorts of pathological events in the CNS, since all neurological defects are basically characterised as disruptions of neuronal tissue homeostasis (Verkhratsky & Parpura, 2010).

Neurodegeneration is a process of progressive neuronal loss leading to extreme cognitive defects. The thesis focuses on the role of astrocytes during onset and progression of several neurodegenerative disorders, namely Alzheimer's disease, Amyotrophic lateral sclerosis, Parkinson's disease, and Huntington's disease. The precise mechanisms underlying the astrocytic behaviour in the course of neurodegeneration are only yet to be resolved. Nevertheless, there is a wide range of evidence suggesting that the astrocytic morphology and physiology may be altered during the neurodegenerative process (Bélanger & Magistretti, 2009).

The aim of this work is to highlight the importance of astrocytes and to outline their principal roles in both physiological and pathophysiological conditions. The second part is dedicated to the introduction of the major characteristics of selected neurodegenerative diseases and the description of astrocytic changes accompanying the process of neurodegeneration. In the conclusion a comparison of the similarities in astrocytic responses to a previously mentioned neurodegenerative disorders is given.

Astrocytes

Neuroglia

Glial cells are regarded as the most numerous cells of the CNS (Azevedo et al., 2009). This broad cell group consists of microglia and macroglia, the latter comprising astrocytes, oligodendrocytes, NG2 cells and ependymal cells. They all contribute to maintaining the homeostasis of the CNS and have a major role in supporting the neurons (Simard & Nedergaard, 2004).

The term Neuroglia (Nervenkitt) was first introduced in 1856 by a German pathologist Rudolf Virchow. However, his idea of glia was somehow different from the one acknowledged nowadays. He ascribed Nervenkitt to a sort of connective tissue functioning only as a substance holding the brain cells together (Virchow, 1858). During the next few decades new staining techniques have been developed, sheading more light on the categorisation of the glial cells and understanding of the importance of their roles in the nervous system (Castellano et al., 1991; Andersen et al., 1992).

Defining an astrocyte

It was in the 1890's when Michael von Lenhossek came with the name „Astrocytes" to refer to the glial cells in the higher vertebrates. By changing the nomenclature from the previous "glia" to astrocytes he pointed out the function of these cells and their equality with the nerve cells, since the original term (glia) indicated the role of these cells to be only passive, glue-like (Lenhossek, 1891).

The etymology of the term astrocyte indicates its stellate morphology hence astrocytes could also be translated as star-like cells. They are the most abundant glial cells in the CNS and are characterised by their leaflet-like processes. Their endfeet forming processes can either give rise to a glial membrane on the CNS surface or a perivascular sheet by attachments to the blood vessel (Simard & Nedergaard, 2004).

For a long time the main attributes that helped distinguish the astrocytes from other glial cells were considered to be their stellate morphology and the presence of the astrocytic fibrils – consisting of glial fibrillary acidic protein (GFAP) (Nolte et al., 2001). As the tools used for identifying astrocytes improved the categorization of these cells became less straightforward.

Despite all the confusions surrounding astrocyte definition, there is one property unifying the cells described as the astrocytes. This characteristic resides in their function of maintaining the homeostasis of the nervous system. They play a key role in all sorts of processes including blood-brain barrier formation (Simard et al., 2003), ion and neurotransmitter regulation (Rothstein et al., 1996), mediation of trans-cellular communication via gap-junctions and other (Nagy & Rash, 2003).

Classification

Astrocytes belong to a very diverse and heterogeneous class of cells divided into several subclasses depending on various criteria. Based on their location and morphology, astrocytes were divided into two subtypes: fibrous and protoplasmic. This classification was first introduced back in 1890's by Kolliker and Andriezen (Andriezen & Lond,1893) .

Protoplasmic astrocytes predominate in the grey matter. In terms of their morphology, they are characteristic for the "bushy" structure of their processes also referred to as spongiform. These abundantly branched processes facilitate intimate associations with synapses (Bushong et al., 2004; Miller & Raff, 1984). The white matter is where fibrous astrocytes can be found. The orientation of their fine and poorly branched processes is along the axon tracts (Miller & Raff, 1984; Oberheim et al., 2009). Both of these cell subtypes are consistent in making extensive contacts with blood vessels (Marín-Padilla, 1995).

There are other types of astrocytes as well. Radial glia, for example, function during the development of the brain (Anthony et al., 2004). In the cerebellum these cells are called Bergmann glial cells (Grosche et al., 2002). Their function is related to the development of cerebellum while the retinal development is associated with Müller glial cells (Bernardos et al., 2007).

Molecular markers

The most common astrocytic marker is GFAP. It has been widely used ever since the development of the gold chloride-sublimate staining technique in early 20th century (Kálmán & Hajós, 1989; Vaughn & Pease, 1967). GFAP is an intermediate filament, 8-9 nm in diameter, expressed mainly in mature astrocytes. It is involved in structural stabilisation of the astrocytic processes resulting in cell motility and shape modulations of the astrocytes (Eng et al., 2000).

Another molecule widely used as a marker for astrocytes is a calcium and zinc binding protein S100B (Ogata & Kosaka, 2002; Baudiers, 1986). Most of the GFAP-positive proliferating astrocytes are not immunostained for this protein, suggesting that it is mainly expressed in mature astrocytes. It was proved that even multipotent astrocytic stem cells lack its expression (Deloulme et al., 2004).

Aldehyde dehydrogenase 1 family, member L1 (ALDH1L1) also known as 10-formyltetrahydrofolate dehydrogenase (FDH) is an astrocytic enzyme playing an important role in the cell division and growth. This folate enzyme has been proposed to be an astrocyte specific protein and is now used as a relatively new marker for astroglial visualisation (Yang et al., 2011).

A number of studies have revealed the presence of glutamine synthetase (GS) in the astrocytes. Nevertheless, this enzyme converting glutamate to glutamine has not been confirmed to be exclusively astrocytic protein, it is considered to be one of their markers (Miyake & Kitamura, 1992).

Astrocytic functions

Developmental function

Astrocytes were proved to be actively involved in neurogenesis regulation by releasing a number of growth factors, interleukins and other proteins with ability to control neuronal maturation and survival. Astrocytes also function as brain stem cells – they serve as a source for both neurons and glial cells (Song et al., 2002; Ashton et al., 2012; Wilhelmsson et al., 2012). Moreover, they control synaptogenesis (Song et al., 2002) and provide a scaffold for cells during neuronal migration (Bozoyan et al., 2012; Kaneko et al., 2011).

Structural function

The micro-architecture of the grey matter is divided into individual astrocytic domains. Each structural unit is established by separate protoplasmic astrocyte and is almost independent. Astrocytes provide for extensive connections with neurons, synapses and blood vessels within each astrocytic domain (Bushong et al., 2002; Bushong et al., 2004; Halassa et al., 2007).

There is only slightest overlap between these territories, but these areas are pivotal for further interconnections. The individual astrocytic domains are coupled through gap junctions and integrate into syncytia. The gap junctions are concentrated in the peripheral astrocytic processes and mediate a transfer of the information enabling the inter-glial communication, broad signalling and intercellular transfer of glucose and lactate (Nagy & Rash, 2003; Ball et al., 2007; Rouach, 2008). The gap junctions are channels composed of two connexons, also called “hemichannels”, localised in the plasma membrane of neighbouring cells. Connexins are the structural subunits, which are involved in connexon formation. The most prevalent connexins in astrocytes are Cx43 and Cx30 (Nagy & Rash, 2003). It is noteworthy that there are immense variations of the gap-junctional connectivity extent of glial cells among the individual brain regions and even within one region (Wallraff et al., 2004)

Homeostatic function

Astrocytes are extensively involved in homeostasis maintenance in CNS. The properties of the astrocytic membrane play an important role in regulating the extracellular ion concentration, pH, brain water, and neurotransmitter homeostasis (Figure1) (Verkhratsky & Parpura, 2010).

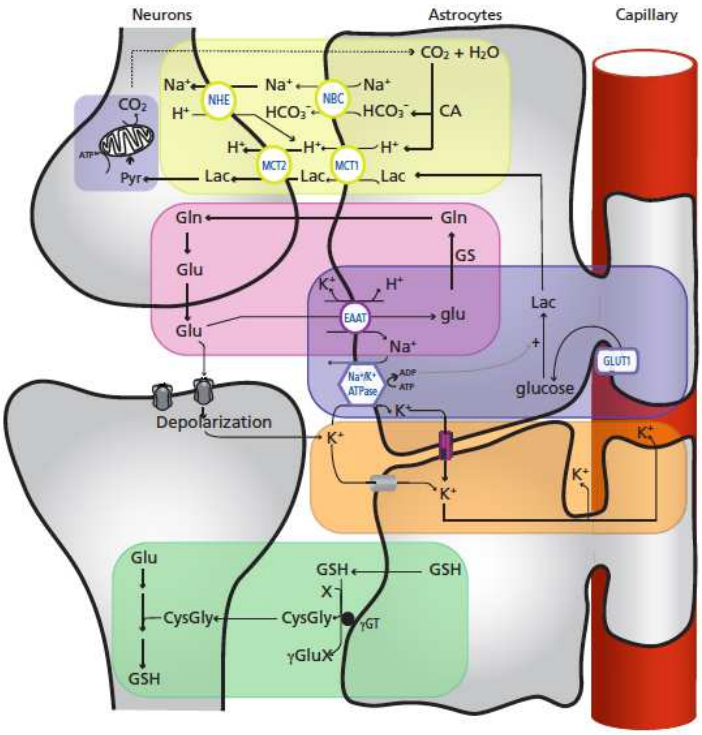

Figure 1: **Summarization of the principal homeostatic functions of astrocytes.** Pink box: glutamate-glutamine cycle. Astrocytic uptake of glutamate is mediated by excitatory amino acid transporters (EAATs). Glutamine synthetase (GS) converts glutamate into glutamine, which is then resynthesised back to glutamate. Violet boxes: lactate shuttle. Na^+ entry accompanies glutamate uptake by astrocytes. Increased ADP triggers glucose intake from blood vessel through the glucose transporter (GLUT1). Lactate is transferred to neurons via monocarboxylate transporters (MCT-1, MCT-2) where it is converted to pyruvate. Yellow box: pH buffering. CO_2 is broken down to H^+ and HCO_3^- by carbonic anhydrase (CA). Na^+- HCO_3^- cotransporter (NBC) mediates transport of HCO_3^- and Na^+ into extracellular space. Orange box: K^+ buffering. Astrocytes distribute K^+ from areas with higher to sites with lower K^+ concentration. Green box: Glutathione metabolism. Astrocytic γ-glutamyl transpeptidase (γGT) cleaves glutathione (GSH) from astrocytes to CysGly, which is a neuronal precursor for GSH (Bélanger, Magistretti, 2009).

Ion homeostasis

The concentrations of extracellular ions need to be controlled and maintained to preserve an accurate neuronal excitability (Newman et al., 1984). Astrocytes are known to play an important role in this process thanks to their unique membrane properties. Even though astrocytic channels and receptors do not differ a lot from those expressed in neurons, their proportion and density vary between these cell types (Papanikolaou et al., 2019).

Astrocytes control the level of extracellular K^+, which is important for repolarization phase of action potential. In the process known as potassium spatial buffering astrocytes disperse local extracellular K^+ from areas with increased concentrations to sites with low [K^+] (Figure 2). This mechanism of K^+ redistribution through astrocytic syncytium is preceded by K^+ uptake mediated by a

number of ion channels and transporters. Inwardly rectifying K^+ channels (Kir), with their ability to favor the influx of K^+ to astrocytes, play an important role in spatial buffering (Takumi et al., 1995; Higashi et al., 2001). The influx of K^+ needs to be accompanied by either efflux of other cations or anion influx. Thus, presence of Na^+/K^+ ATPase pumps (Larsen et al., 2014) and $(Na^+)/K^+/Cl^-$ cotransporters is not insignificant and they are considered to be one of the key players in K^+ uptake (Lenart et al., 2004). Furthermore, astrocytes express calcium and voltage-dependent K^+ channels (Quandt & MacVicar, 1986; Bevan et al., 1985).

The presence of functional ion channels is not the only factor influencing the ion homeostasis in the CNS. The transfer of ions is also interconnected with aquaporin channel-mediated brain water transport. The predominant channel enabling the influx of water in astrocytes is aquaporin 4 (AQP4), localised in the astrocytic perivascular endfeet and perisynaptic processes, usually in a close proximity to potassium channels Kir 4.1 (Figure 2) (Nagelhus et al., 1999; Lunde et al., 2015). It has been shown that a decreased level of perivascular AQP4 results in malignant K^+ buffering (Eid et al., 2005).

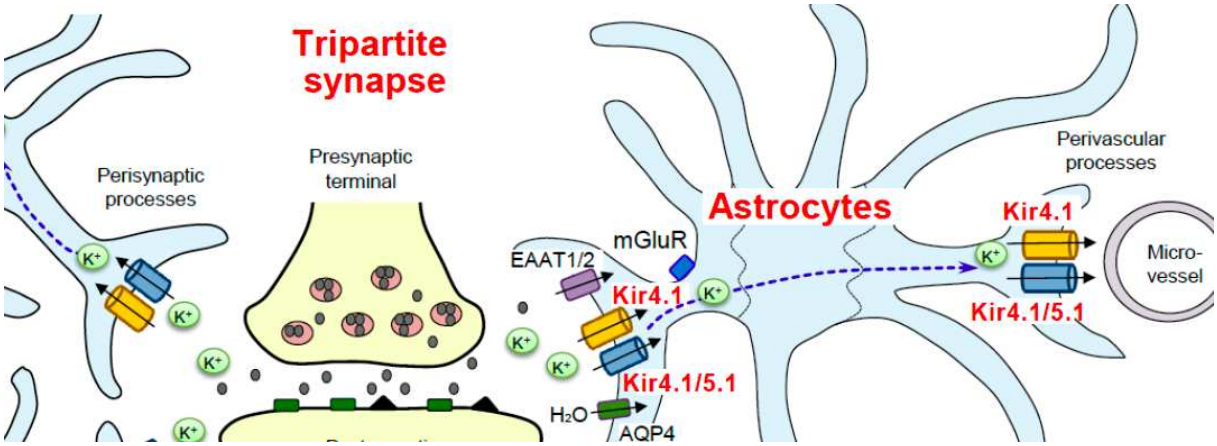

Figure 2: **K^+ spatial buffering**. The action of K^+ buffering is mediated by several subtypes of channels for example Kir 4.1 and Kir 4.1/5.1 which are sited in perisynaptic and perivascular astrocytic processes. Glutamate and water intake to astrocytes by excitatory amino acid transporters (EAATs) and aquaporin 4 (AQP4) are both functionally coupled with potassium channels. (Ohno et al., 2018).

Neurotransmitter homeostasis

Astrocytic membrane is enriched with numerous transporters taking part in transfer of various neurotransmitters like glutamate, γ-aminobutyric acid (GABA), and glycine from the synapses (Bezzi et al., 2004; Guastella et al., 1990; Shibasaki et al., 2016). After the uptake by astrocytes, these neurotransmitters are enzymatically converted into precursors, which are subsequently recycled to synapses for their reconversion into active neurotransmitters (Sibson et al., 1997).

The principal amino acid engaged in neuronal excitability is glutamate (Ehrhart-Bornstein et al., 1991). The role of astrocytes is to maintain low extracellular concentrations of the glutamate via its uptake (Rothstein et al., 1996). The process is mediated by the two main glutamate transporters: excitatory amino acid transporter 1 (EAAT1), known as glutamate-aspartate transporter (GLAST) in rodents (Storck, et al., 1992); and excitatory amino acid transporter 2 (EAAT2) also referred to as glutamate transporter 1 (GLT1) (Pines et al., 1992). Both transporters are predominantly localized in astrocytic processes. The transport of glutamate is an electrogenic process and requires energy of transmembrane Na^+ gradient. The glutamate transporters exchange one glutamate and three Na^+ (or two Na^+ and one H^+) for one K^+ and OH^- (Illarionova et al., 2014; Cholet et al., 2002).

Glutamate uptake is followed by conversion of glutamate into non-toxic glutamine, a process catalysed by astrocytic enzyme glutamine synthetase. Afterwards, glutamine is transported back to presynaptic terminal of a neuron via extracellular space. The final step in glutamate – glutamine shuttle, necessary for glutamatergic neurotransmission, is a recycling of glutamine back to glutamate (Figure 1) (Ottersen et al., 1992; Sibson et al., 1997).

pH regulation

Numerous studies have proven a contribution of astrocytes to pH maintenance in the CNS. A constant pH within a brain is one of the essential requirements for the proper protein modifications and processing, organelle integrity, cargo directing, and other CNS processes (Carnell & Moore, 1994; Chanat & Huttner, 1991; Linstedt et al. 1997, Puri et al., 2002). Various astrocytic transport mechanisms take part in proton transfer, these include bicarbonate transporters (Ko et al., 1999), Na^+/H^+ exchanger (Wada et al., 2005), proton ATPase (Philippe et al., 2002), and monocarboxylic transporters (Bröer et al., 1997; Mac & Nałęcz, 2002). The reaction of carbonic anhydrase, an enzyme reversibly converting carbon dioxide and water into proton and bicarbonate, is the major process preserving the acid-base homeostasis (Chen & Chesler, 1992).

Antioxidant function

The CNS is notably vulnerable to damages caused by oxidative stress. An imbalance between reactive oxygen species (ROS) production and antioxidant processes frequently appears under neuropathological conditions (Gegg et al., 2003). Both neurons and astrocytes carry a neuroprotective force against oxidative injuries in a form of antioxidant production (Tanaka et al., 1999). A number of different antioxidants like glutathione, ascorbate, and vitamin E are present in astrocytes, together with a variety of ROS-detoxifying enzymes (Makar et al., 1994). The most significant antioxidant molecule in the CNS is glutathione (GSH). Astrocyte-neurone GSH shuttling provides precursors for the neuronal synthesis of GSH (Figure1, green box). Astrocytic ectoenzyme γ-glutamyl transpeptidase (γGT) converts a GSH, released from astrocytes to extracellular space, into CysGly, which is either directly

transferred to neurons or cleaved first into glycine and cysteine and transferred afterwards (Dringen et al., 1997; Dringen et al., 1999).

Metabolic function

Glycogen functions as a major energy source for the CNS cells. The distribution of brain glycogen is uneven throughout the brain, with astrocytes being the primarily sites for its storage. Active neurons are provided with energy substrate by astrocytes, which have a substantial location in the CNS organization, with processes contacting both blood vessels and neurons (Phelps, 1972; Cruz & Dienel, 2002; Cataldo & Broadwell, 1986).

Under the physiological conditions blood glucose is taken up by the astrocytes via glucose transporters (GLUT1) and is then converted into pyruvate in the process of aerobic glycolysis (Morgello et al., 1995). Lactate is a product (pyruvate being the substrate) of the reaction catalysed by lactate dehydrogenase isoenzyme A (LDHA) expressed in astrocytes which is transported into neurons, where it serves as a fuel for neuronal processes (Figure 3) (Gandhi et al., 2009). This mechanism has been introduced as astrocyte neuronal lactate shuttle hypothesis (ANLSH). This process can be stimulated either by glutamate, which increases the uptake of glucose from capillaries or by glycogen breakdown (Pellerin et al., 1998; Pellerin & Magistretti; 1994).

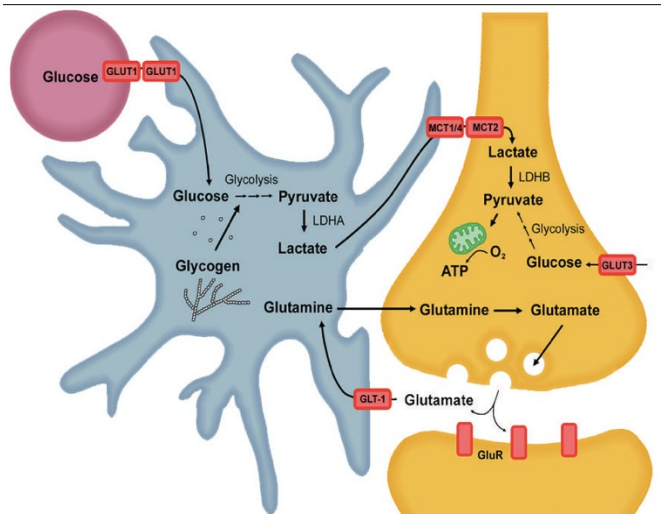

Figure 3: **Lactate shuttle hypothesis.** The transfer of neurotransmitter glutamate into astrocytes is mediated by glutamate transporters (GLT-1). It is transformed to glutamine in astrocytes. Glycolysis and astrocytic glucose uptake (by glucose transporters GLUT1) are both increased as an accompanying processes of glutamate uptake. Lactate dehydrogenase isoenzyme A (LDHA) in astrocytes converts pyruvate to lactate which is transported out of astrocytes by monocarboxylate transporter 1 or 4 (MCT1/4) for then being imported into neurones by MCT2. Lactate is then converted back to pyruvate by another lactate dehydrogenase (LDHB) in neurones (Newington et al., 2013).

Signalling function

Regarding the signalling function, astrocytes have the ability to regulate synaptic transmission. As it was mentioned previously, the concentration of neurotransmitters in the synapses can be controlled via uptake by specific astrocytic transporters. Another way of modulating synaptic transmission is by the release of gliotransmitters (Araque et al., 1998). Gliotransmitters are molecules produced by and/or stored in glia. They induce rapid responses in nearby cells and play an important role in both physiological and pathological processes. Several miscellaneous gliotransmitter release mechanisms have been described in astrocytes, such as a release through: volume-regulated anion channels (Li et al., 2012; Takano et al., 2005), unpaired connexons (hemichannels) (Ye et al., 2003), ionotropic purinergic receptors (Duan et al., 2003), cystine-glutamate antiporters (Warr et al., 1999), Ca^{2+}-dependent exocytosis, etc. (Parpura et al., 1994). There are two principal categories of gliotransmitters expressed in astrocytes. The first category consists of amino acids and their derivates, comprising molecules like glutamate (Araque et al., 2000), aspartate (Rutledge et al., 1998), D-serine (Schell et al., 1995), GABA (Bowery et al., 1976), taurine (Shain & Martin, 1984); while the second group consists of nucleotides and their derivates like ATP (Anderson et al., 2003), UDP-glucose and others (Kreda et al., 2009).

Ca^{2+} waves

Astrocyte signalling across the syncytium is mediated by gap junctions (see "Structural function") and this mechanism is a means of Ca^{2+} waves spreading. Ca^{2+} waves are generated either spontaneously or as a result of neuronal activity and have an effect on several pathways, including those engaged in modulation of synaptic transmission and plasticity (Stout et al., 2002).

Vascular function

Blood brain barrier

The blood brain barrier is a selective physical barrier composed of microvessel endothelial cells enclosed by basal lamina and astrocytic endfeet (Simard et al., 2003). Endfeet are astrocytic structures adapted for the direct interaction with the vessels (Gordon et al., 2007). Tight junctions between endothelial cells play an important role in the permeability of the blood brain barrier (Morita et al., 1999). Only small lipophilic molecules and gases like oxygen, carbon dioxide (CO_2), and ethanol are able to penetrate a lipid bilayer of the endothelium. All the other components (small polar solutes, large molecules,…) need a specific carrier to transport them into or out of the CNS (Bartus et al., 1996). Morover, it is known that astrocytes promote the tight junction formation and tightness (Janzer & Raff, 1987).

Blood flow regulation

As it was previously mentioned, astrocytes are connected with both neuronal synapses and brain vessels. This location represents an excellent position for neurovascular coupling or, in other words, a blood flow regulation in response to neuronal changes. Astrocytes are able to control the physiology of blood vessels leading to either vasoconstriction (Mulligan & MacVicar, 2004) or vasodilation (Straub et al., 2006). The direction in which a vessel diameter changes depends, on the astrocytic molecular mediators produced by the arachidonic acid conversion. This process is also regulated by nitric oxide (NO) levels (Gordon et al., 2007).

Astrocytes under pathological conditions

Glial cells, and astrocytes in particular, are significant for their irreplaceable role in the maintenance of CNS homeostasis. Thus, the pathological conditions brought about by brain injuries, diseases or other impairments have a noticeable impact on astrocytic physiology and morphology. Despite all the differences between the individual CNS disorders, there are some common features accompanying these diseases, for instance: inflammation, ROS accumulation, excitotoxicity, and metabolic failure. Majority of these processes are under astrocytic control in a healthy brain (Bélanger & Magistretti, 2009).

The most common astrocytic response to the physiological changes in a diseased or injured brain is undoubtedly the one known as reactive astrogliosis. A wide range of different astrocytic changes and reactions has been observed in a process of astrogliosis, some of them being contradictory or only poorly explained. However, Sofroniew suggested a model in his review on Molecular dissection of reactive astrogliosis and glial scar formation which comprises general characteristics of the process in these four points:

1. Reactive astrogliosis includes a range of astrocytic changes occurring during CNS injury or disease of any form and severity.
2. The changes in astrocytes depend on the nature and the severity of the damage and vary along a gradated continuum of progressive alterations in molecular expression, progressive cellular hypertrophy and proliferation and scar formation which is the most severe.
3. The changes accompanying the astrogliosis are regulated by the precise signalling pathways with the ability to customize their nature and degree, in a context-specific manner.
4. Astrocytic activity during reactive astrogliosis is altered through gain and loss of functions that have the ability to affect the surrounding cells both positively and negatively (Sofroniew, 2009).

There is a vast array of factors in astrocytic microenvironment, such as: cytokines, adhesion molecules, growth factors, and various signals from glial, neuronal and other cells, all contributing to a modulation of astrocyte reaction (Balasingam et al., 1994; Krum et al., 2002). Even though astrogliosis represents a gradated continuum of changes it has been categorized into three stages: mild, moderate, and severe (Figure 4). All the three present with altered gene expression resulting in GFAP up-regulation and cellular hypertrophy. Mild to moderate reactive astrogliosis usually appears during some forms of trauma and in bacterial or viral infections, further from focal CNS lesions. Severe reactive astrogliosis on the other hand is accompanied by astrocyte proliferation resulting in tissue reorganization and astrocyte domain loss. These structural changes result in a so called glial scar formation, isolating the area of the tissue damage, necrosis, infection, or inflammation by compact border formation around it. Glial scar formation is known to be induced by CNS impairments like invasive infections, chronic neurodegenerative changes, neoplasm, trauma and others (Drögemüller et al., 2008; Voskhul et al.,

2009). Glial scar separates defective brain areas from the healthy tissues, prevents axon regeneration and hinders inflammation from spreading. This barrier persists for a long period of time and it requires astrocytes with proliferative potential for its formation (Wanner et al., 2013; Silver et al., 1993; Faulkner et al., 2004).

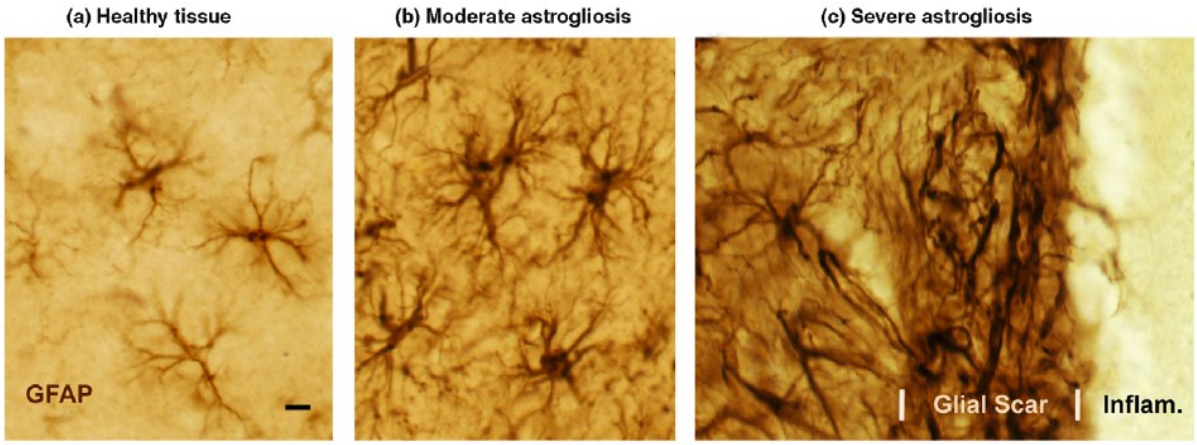

Figure 4: **Visualization of different types of reactive astrocytes from murine cerebral cortex**. Astrocytic cytoskeleton is visualized via GFAP immunohistochemical staining. (a) Healthy cortex. GFAP is detectable only in some astrocytes. (b) Moderately reactive astrocytes with GFAP visible in all astroglial cells, due to injection of bacterial antigen lipopolysaccharide (LPS). Astrocytes are hypertrophied. (c) Scar border (SB) formed by proliferated reactive astrocytes is easily visible and is formed in the close vicinity of the lesion (L) caused by severe traumatic injury. Astrocytes no longer form individual domains but are overlapping with intermingled processes. Scale bar520 mm. (Photos courtesy of the Sofroniew laboratory) (Sofroniew, 2009).

Whether the impact of reactive astrocytes on CNS is rather positive or negative is a question that needs to be further investigated to be resolved properly. There is an evidence of both beneficial and deleterious astrocytic responses to CNS insults. A recent study on reactive astrocytes suggests the existence of two distinct types of astroglia, one characterised as damaging and the other one supportive, named “A1” and “A2” respectively. Neuroinflammation instigates an activation of A1 astrocytes which subsequently up-regulate the pathways involved in synapse destruction, unlike the ischemia-induced A2 astrocytes which implement their protective potential by stimulating neurotrophic factors production (Liddelow et al., 2017).

In general, all CNS disorders could be characterised as homeostatic failures. Astrocytes may contribute to these imbalances in the homeostasis in various ways, often by altered expression or function of transporters and channels (Figure 5).

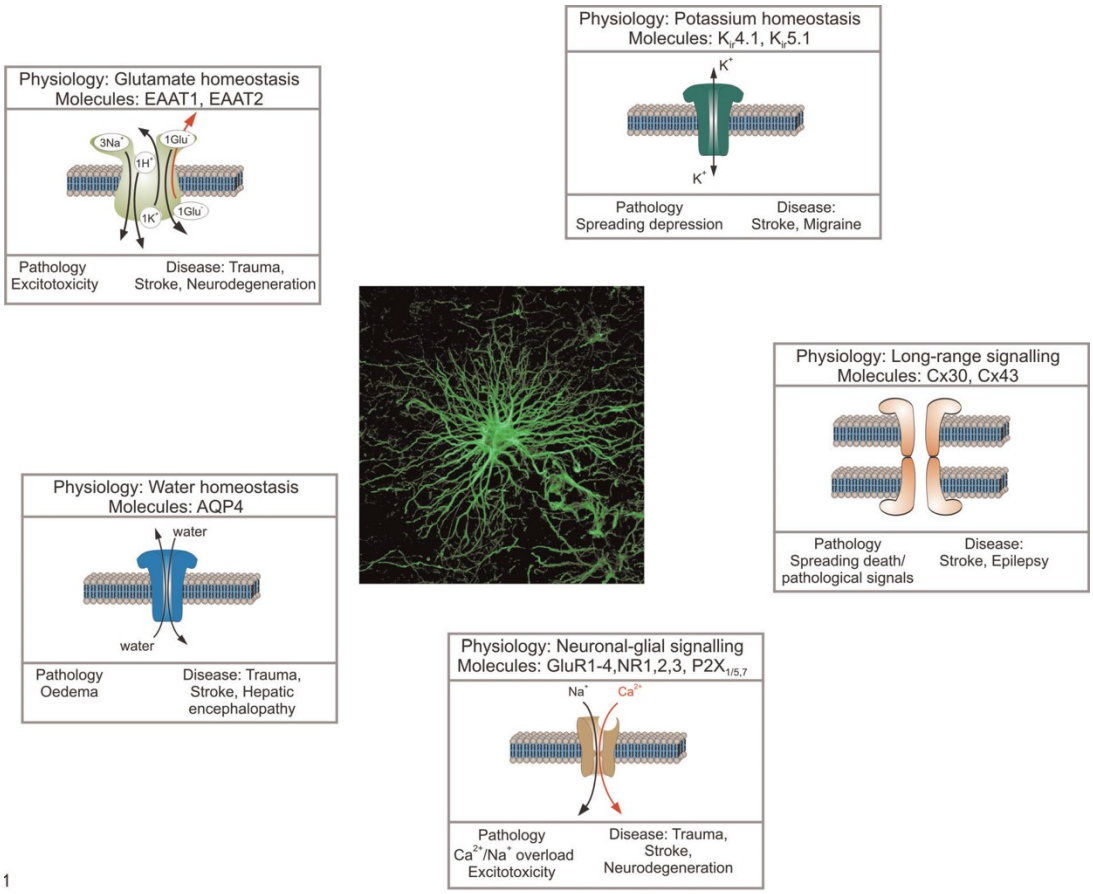

Figure 5: **Astrocytic pathophysiology.** Astrocytes maintain potassium, glutamate and water homeostasis and are involved in signalling. Several types of transporters and channels are involved in the process of the homeostasis maintenance. These processes are disrupted under pathological conditions, hence they contribute to the brain damage. (Verkhratsky et al., 2012).

One of the examples of such failures in ion homeostasis is represented by the alterations in potassium channel expression and location. Miscellaneous CNS pathologies, either chronic or acute, have the ability to trigger defects in K^+ channel functions, leading to an impaired K^+ spatial buffering. As a result of dysfunctional K^+ uptake a spreading depression may occur, mediated by extracellular K^+ accumulation. Disproportion of the ionic distribution leads to depolarization of the astrocytes, which in turn inflicts the glutamate transport defects. Together with the affiliation of other proteins like aquaporins and connexins the whole process ends up with systemic collapse of homeostasis contributing to the neuronal damage (Tong et al., 2014)

Another abnormality observed in astrocytic behaviour under pathological conditions is the process called astrocyte swelling. An increased uptake of water via AQP4 on astrocytic endfeet impels the cells to extend their volume, contributing to a condition described as cytotoxic edema (MacAulay et al., 2001; Schneider et al., 1992).

Neurodegeneration

Neurodegeneration is a chronic and progressive process of neuronal death, leading to brain atrophy and impairments of CNS connectivity. The onset and progression of neurodegenerative disorders can be triggered by miscellaneous factors from traumatic and infectious attacks to genetic susceptibility or sporadic errors accumulation. Astrocytes play a significant role in health of neuronal cells, and therefore their contribution to the development of neurodegeneration is critical (Heneka et al., 2009; Verkhratsky et al., 2013).

Alzheimer's disease

Alzheimer's disease (AD) is a neurodegenerative disorder manifesting itself by dementia and was first described by Alois Alzheimer in 1907 (Alzheimer, 1907; Stelzmann et al., 1995). From all types of dementia, Alzheimer's disease is the most frequent one among the elderly. The clinical symptoms of this neuropsychiatric disorder include progressive memory loss and dysfunction of higher cognitive domains. Histological features accompanying development of AD are senile/neuritic plaques and neurofibrillary tangles (NFT) formation. Neuronal degeneration and death are the consequences of these pathological changes (Arriagada et al., 1992). The senile plaques were shown to be formed by an extracellular aggregation and deposition of the β-amyloid peptide (Aβ) fibrils (Mawuenyega et al., 2010). Tau protein is the principal component engaged in NFT formation (Tapiola et al., 1997).

Alzheimer's disease occurs in two distinct forms: sporadic and familial. These two forms have strong phenotypical similarities and it is often difficult to distinguish between them. Familial form of the disease is usually induced by mutations in AD-related genes like amyloid precursor protein, presenilin 1 or presenilin 2, which all contribute to Aβ accumulation (Goate et al., 1991; Levy-Lahad et al., 1995; Sherrington et al., 1995). Even though the genetic factors may be involved in the onset and progression of the disease, it seems that majority of AD cases are of non-familial origin (Maccioni et al., 2001; Selkoe, 2001).

There are three phases of the disease: pre-symptomatic phase, prodromal phase, and dementia. In the first one (pre-symptomatic phase) the cognitive functions are not affected, however, the patients may have AD pathological changes (Knopman et al., 2003; Price & Morris, 1999). The prodromal phase is also known as mild cognitive impairment. It presents itself with the earliest cognitive symptoms, such as episodic memory deficits (Morris et al., 2001). Dementia is the final and the most severe phase of AD and is characterised by extreme impairments in multiple domains causing loss of function (Visser et al., 2009).

Braak stageing is used to distinguish between six distinct pathological stages of AD. Changed intraneuronal neurofibrils in AD are found in three forms: neuritic plaques with variable distribution,

neurofibrillary tangles and neuropil threads. The latter two are distributed in a specific manner. The distribution patterns determine the stageing (Braak & Braak, 1997a; 1997b).

Astrocytes in Alzheimer's disease

One of the most controversial roles of astrocytes in AD, however, seems to be the one they play in the metabolism of amyloid β. Various studies have revealed contradictory results on this matter. Astrocytes in AD are known to be affected by Aβ aggregates in miscellaneous ways. The changes are visible not only on the morphological level but also metabolism and other astrocytic functions are altered (Pike et al., 1994; Allaman et al., 2010; Söllvander et al., 2016). It has been proposed that Aβ can be generated not only by neurones but also by astrocytes. Thus, astrocytes were suggested to be directly implicated in the formation of Aβ aggregates in the course of AD (Zhao et al., 2011).

A release of gliotransmitters such as glutamate, GABA, ATP, or D-serine during AD is altered in reactive astrocytes as well. GABA is a primary gliotransmitter involved in the inhibition of neuronal transmission. The activation of microglial and astrocytic GABA receptors during inflammatory processes in brain results in a decreased release of proinflammatory cytokines, reducing the neurotoxicity (Lee et al., 2011). On the other hand, deleterious effects of the over-expressed astrocytic GABA were also reported. Hypertrophy of astrocytes and GABA accumulation, which were observed in the AD mouse models, are both occurring in the proximity of amyloid aggregates. Furthermore, a contribution of elevated astrocytic GABA release to cognitive decline was proposed (Jo et al., 2014; Wu et al., 2014). Another recent study revealed that GABA accumulation in astrocytes appears not only in the presence of amyloidosis, but also as a result of neuronal hyperactivity (Brawek et al., 2018). Another common gliotransmitter – D-serine was also proposed to be implicated in the process of neurodegeneration in the course of AD (Balu et al., 2019).

As mentioned above, AD occurs predominantly among elderly hence, some of the deleterious processes related to aging may contribute to the disease onset and progression. There is an evidence of age-associated changes in astrocytic functioning, which subsequently conduce to AD pathogenesis. One of such astrocytic dysfunctions results from decreased insulin-like growth factor receptor (IGFR) signalling, which then affects aging of neurons and neurodegeneration. A study on both mouse models and astrocyte cultures revealed a number of negative effects of reduced IGFR levels on nervous system, such as defective ATP synthesis, increased gliosis, augmented level of mitochondrial ROS, and moreover, altered glucose and Aβ uptake by astrocytes (Logan et al., 2018).

Amyotrophic lateral sclerosis

Amyotrophic lateral sclerosis (ALS) also known as Lou Gehrig's disease is the third most common neurodegenerative disorder after AD and Parkinson's diseases. It is a fatal idiopathic motor neuron disease, which presents with upper and lower motor neuron degeneration and death, localised in the brain and spinal cord. The most probable cause of death of amyotrophic lateral sclerosis patients is due to respiratory failure (Brown & Al-Chalabi, 2017). Not only motor neurons supplying voluntary muscles are affected by ALS, but also lower medullar motor neurons and upper motor neurons in the cerebral cortex (Gordon, 2013).

Population-based studies revealed that ALS is more prevalent among elderly, men being at higher risk of developing the disease than women (Moura et al., 2016). Only 5-10% of ALS cases are of genetic origin. Thus, a vast majority of the patients is diagnosed with a sporadic form of the disease (Chio et al., 2011) .

Many genes have been proved to be involved in both familial and sporadic ALS; however, approximately 20% of cases of fALS and 1% of sALS are related to dominant mutations in the Copper-Zinc superoxide dismutase (SOD1) gene (Rosen et al., 1993).

In about 70% of patients the first symptoms of the disease occur in the limbs (usually in the arms) and are focal and unilateral. They include foot drop, walking difficulties, hand dexterity loss, and weakness in the arms during movements. With the progression of the disease a complete loss of the ability to walk may occur. Older women usually present with bulbar-onset of the ALS, which means a poorer prognosis for the patients. Speech (dysarthria) and swallowing (dysphagia) difficulties are characteristic signs of the bulbar-onset ALS. The possible consequences of these conditions are excessive salivation, anarthria and even malnutrition. Furthermore, as a result of axial weakness, dropped head and kyphosis may also accompany the disease (Gordon, 2013; Kiernan et al., 2011).

Astrocytes in amyotrophic lateral sclerosis

It has been proposed that a loss of adenosine deaminase in defective astrocytes leads to increased neurotoxicity (Allen et al., 2019) A functional adenosine deaminase induces deamination of adenosine into inosine, which is known to have glioprotective properties (Haun et al., 1996).

Astrocytes isolated from mice models overexpressing human SOD1, together with human post-mortem ALS astrocytes and human induced pluripotent stem cell (iPSC)-derived astrocytes were all proved to cause motor neuron death in co-cultures (Almad et al., 2016; Harlan et al., 2016). This effect results from all sorts of ongoing defective processes in ALS astrocytes. For example altered gap junctional coupling was observed in astrocytes derived from $SOD1^{G93A}$ mice. An increased expression of astrocytic hemichannel-forming connexin Cx43 is responsible for these alterations, which

subsequently bring about elevations in the intracellular calcium levels (Almad et al., 2016). One of the options how to diminish an astrocyte-mediated neurotoxicity is by providing an augmented supply of nicotine amide dinucleotide NAD^+ into ALS astrocytes. NAD^+ is participating in mitochondrial metabolism and its increased levels contribute to peroxide toxicity resistance and decreased amounts of mitochondrial reactive oxygen species (Harlan et al., 2016).

Another recent study revealed a correlation between the expression of astrocytic tumour necrosis factor receptor 1 (TNFR1) and a neuronal survival. The TNFR1 promotes a release and synthesis of glial cell line-derived neurotrophic factor (GDNF), which is recognized as a factor implicated in the motor neuron protection. Thus, an astrocytic TNFR1-GDNF axis denotes a new potential therapeutic target in ALS (Brambilla et al., 2016).

Neuroinflammation is also implicated in the progression of ALS. The neuroinflammatory processes have both protective and toxic consequences on neurons, and are induced by several cell types, such as microglia, T-cells and, of course, astrocytes in a non-cell-autonomous manner. A transforming growth factor-β1 (TGFβ1) produced by activated astrocytes is overexpressed during the ALS progression, and has a neurotoxic effect, and therefore, it contributes to the degeneration of neurons. Furthermore, TGFβ1 interfere with neuroprotective action of inflammatory molecules produced by microglia and T-cells (Figure 6) (Endo et al., 2015).

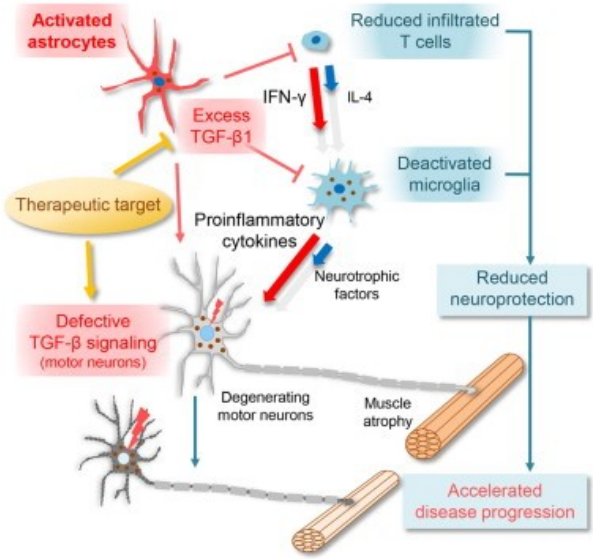

Figure 6: **Excess astrocyte-derived TGFβ1 contributes to disease progression.** TGF-β1= transforming growth factor beta 1, IFN-γ = interferon γ , IL-4= interleukin 4 (Endo et al., 2015).

Parkinson's disease

Parkinson's disease (PD) is a condition characterised by a progressive loss of dopaminergic neurons of pars-compacta - a part of midbrain-located substantia nigra- and a region specific aggregation of α-synuclein, which functions as a negative regulator of dopamine neurotransmission. (Abeliovich et al., 2000). One of the histological hallmarks of PD are structures known as Lewy bodies. These intracellular eosinophilic neurofibrillary tangles are suggested to contribute to neuronal death (Gibb & Lees, 1989; Greffard et al., 2010). The disease comes with a number of symptoms, among which the most significant are tremor, bradykinesia, rigidity and postural instability (Paísan-Ruíz et al., 2004).

Ageing and genetic predisposition are primary risk factors concerning PD. Other risk factors linked to a sporadic form of the disease have been also reported. They include rural living, middle-age obesity, injury of head, shortage of exercise, exposure to chemicals (Dick et al., 2007); but they are all only of minor importance. Nicotine and caffeine are related to the disease as well, and they both contribute to a decreased risk of PD as they induce a dopamine release in striatum. Moreover, cigarette smoke has been suggested to inhibit monoamine oxidase, an enzyme producing reactive oxygen species, which account for increased risk of neuronal damage. Hence, it is not surprising that non-smokers and people with no or very low caffeine intake have higher susceptibility to develop the disease (Fowler et al., 1996, Ascherio et al., 2003).

Astrocytes in Parkinson's disease

Substantia nigra, which is affected by the disease in the first place, is a brain region with a lower abundancy of astrocytes in comparison to the rest of the brain (Damier et al., 1996). This indicates a lack of astroglial support to the neurons, which might play a crucial role in the development of PD (Mena et al., 2002).

Various genes implicated in the PD progression were detected, one of them being a recessive familial gene known as DJ-1, which is associated with all sorts of cellular functions (Ashley et al., 2016). DJ-1 protein was recognized to be involved in eliminating reactive oxygen species with its antioxidant properties, contributing to a protection of the cell (Andres-Mateos et al., 2007). Besides, it functions in the maintenance of mitochondria (Cai et al., 2015). Other studies propose its participation as a chaperone in α-synuclein folding (Shendelman et al., 2004; Zhou et al., 2006). Furthermore, DJ-1 is implicated in the death of neurons via regulating the activity of phosphatase and tension homolog (Choi et al., 2014). Interestingly, DJ-1 when mutated can even induce aberrations in EAAT2, leading to impaired astroglial uptake of glutamate (Figure 7, E) (Kim et al., 2016). Another study showed that mutations in DJ-1 gene modify the expression of pro-inflammatory mediators in astrocytes. Dysregulation of these factors may

lead to a decreased astroglial protection of the neurons, making them more susceptible to neurodegeneration (Ashley et al., 2016).

One of the possible factors decreasing the expression of DJ-1 is NO. Fortunately, we are able to prevent a down-regulation of DJ-1 by supressing the activity of NO, or eventually inducible nitric oxide synthase (iNOS). It has been revealed that cinnamon and its metabolite have exactly the effect described above. The authors of the study focusing on this matter suggest that oral administration of cinnamon has a neuroprotective role (Khasnavis & Pahan, 2014).

It has been also observed that the depositions of α-synuclein in the extracellular space can stimulate the astrocytic inflammatory responses mediated by activation of its toll-like receptor 4 (Figure 7, C) (Rannikko, 2015).

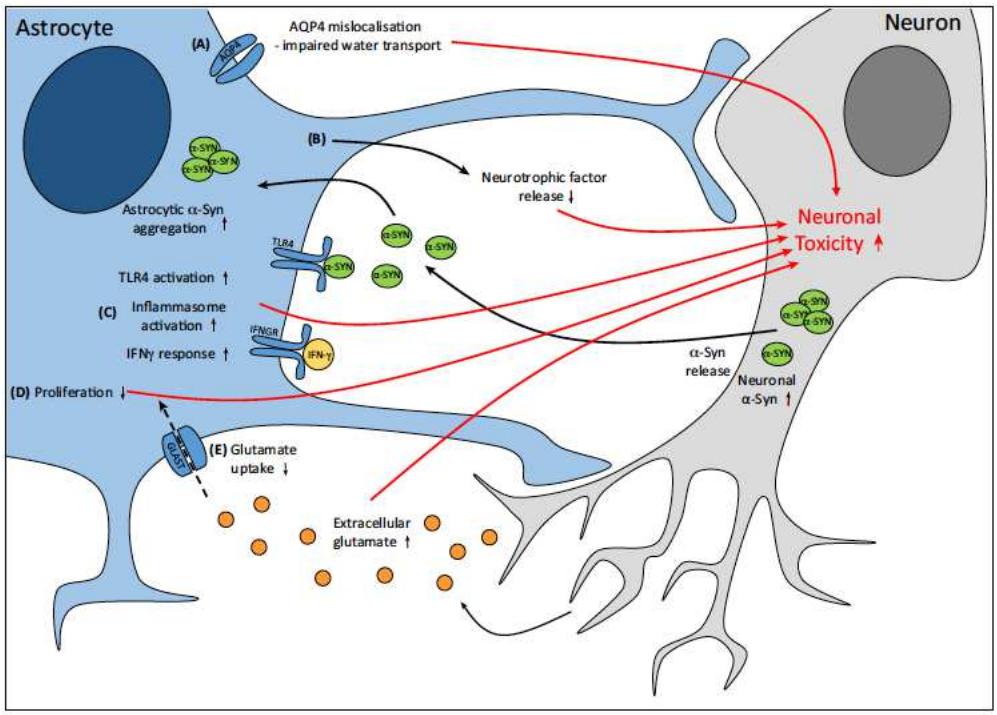

Figure 7: **Astrocytic contribution to neurotoxicity**. Astrocytes can promote toxicity of neurons in different ways: (A) Mislocalisation or disruption of water channels AQP4. (B) Reduced release of neurotrophic factors. (C) Inflammatory responses. (D) Diminished astrocytic proliferation. (E) Decreased activity of glutamate transporters leading to excessive accumulation of glutamate in the extracellular space. (Booth et al., 2017)

Huntington's disease

Huntington's disease (HD) is a progressive genetic disorder with neurodegenerative outcomes, emerging from expansion of triplet CAG repeats in a gene for huntingtin. The onset of HD depends on length of CAG repeats (Rosenblatt et al., 2006). The primary target of neurodegeneration in HD

progression is a striatum. Clinically the disease presents itself with a variety of symptoms from cognitive decline and psychiatric breakdown to a progressive motor dysfunction. The patients diagnosed with HD typically struggle with involuntary movements (chorea), overall motoric slowdown (bradykinesia), incoordination, and rigidity often preceded by behavioural defects (Ross & Tabrizi, 2011).

Astrocytes in Huntington's disease

An increased excitotoxicity which is known to contribute to neurodegenerative processes has been observed in HD. One of the most probable reasons for adverse excitatory neurotransmission detected in HD patients resides in the altered astrocytic metabolism and glutamine release which in turn induce reduced GABA synthesis in neurons. A recent study revealed a down-regulation of EAAT2 glutamate transporter, GS, and the glutamine transporter sodium-coupled neutral amino acid transporter 3 in astrocytes. These factors directly induce the excitotoxic glutamate accumulation in the extracellular space (Skotte et al., 2018). There is still a lot of data missing concerning the astroglial behaviour in HD and a further research would be required to complete a whole picture on the matter. Nevertheless, there is an evidence of various astrocytic alterations, which might play a relevant role in HD progression, such as: increased calcium-dependent glutamate release (Lee et al., 2013), mitochondrial impairments (Oliveira et al., 2007), neuroinflammation mediators' release (Hsiao et al., 2013), Kir 4.1 channels down-regulation (Tong et al., 2014), or abnormal cholesterol production (Valenza et al., 2010).

Conclusion

Astrocytes are a subtype of glial cells and their most important function in CNS is to maintain brain homeostasis. Homeostatic disruption occurs during neurodegeneration, suggesting that astrocytes can play a vital role in the onset and progression of neurodegenerative diseases, or even contribute to their formation. Even though astrocytes are receiving more and more attention from researchers, their role in neurodegeneration is still not fully understood.

Over the past few years, there has been a lot of evidence about both positive and negative effects they have on neurons and their environment. The most typical astrocytic response observed in majority of neurodegenerative diseases is reactive astrogliosis. It is often accompanied by a change in the expression of various genes. A wide range of astrocytic changes occur through different neurodegenerative diseases. Alterations in the metabolism of astrocytes accompany the neurodegenerative processes. Defective glutamate release is one of the most frequently appearing astrocytic changes, observed in most of the neurodegenerative disorders. It is usually caused by down-regulation of one of the glutamate transporters and results in increased excitotoxicity followed by neurodegeneration. Another typical feature of the above mentioned diseases is aberrant ion concentrations and modified signalling. Moreover, the inflammatory processes also contribute to degeneration of the neurons in the course of all AD, PD, ALS, and HD. Pro-inflammatory mediators, however, are produced not only by astrocytes, but by other cell types as well.

In order to obtain a better understanding of the role of astrocytes in neurodegenerative diseases, more detailed investigation and clarification of the processes accompanying reactive astrogliosis is required. At the same time, presuming we want to target the treatment of neurodegeneration to astrocytes, it is important to examine the pathophysiological processes in the neighbouring cells – both neuronal and glial - since the CNS is a dynamic structure, in which an intense communication occurs among its individual interacting components.

Bibliography

Abeliovich A., Schmitz Y., Farinas I., Choi-Lundberg D., Ho W. H., Castillo P. E., Shinsky N., Verdugo J. M., Armanini M., Ryan A., Hynes M., Phillips H., Sulzer D., Rosenthal A. (2000). Mice lacking alpha-synuclein display functional deficits in the nigrostriatal dopamine systém. *Neuron,* 25(1): 239-252.

Allaman I., Gavillet M., Bélanger M., Laroche T., Viertl D., Lashuel H.A., Magistretti P.J. (2010). Amyloid-beta aggregates cause alterations of astrocytic metabolic phenotype: impact on neuronal viability. *The Journal of Neuroscience*, 30(9): 3326-3338.

Allen S.P., Hall B., Castelli L.M., Francis L., Woof R., Siskos A P., Kouloura E., Gray E., Thompson A.G., Talbot K., Higginbottom A., Myszczynska M., Allen C.F., Stopford M.J., Hamingway J., Bauer C.S., Webster C.P., De Vos K.J., Turner M.R., Keun H.C., Hautbergue G.M., Ferraiuolo L., Shaw P.J. (2019). Astrocyte adenosine deaminase loss increases motor neuron toxicity in amyotrphic lateral sclerosis. *Brain,* 142(3): 586-605.

Almad A.A., Doreswamy A., Gross S.K., Richard J.P., Huo Y., Haughey N., Maragakis N.J. (2016). Connexin 43 in Astrocytes Contributes to Motor Neuron Toxicity in Amyotrophic Lateral Sclerosis. *Glia*, 64(7): 1154-1169.

Alzheimer A. (1907). Über eine eigenartige Erlranliung der Hirnrinde. Allgemeine Zeitschrift fur Psychiatrie und phychish-Gerichtliche Medizin, (Berlin) 64: 146-148.

Andersen B.B., Korbo L., Pakkenberg B. (1992). A quantitative study of the human cerebellum with unbiased stereological techniques. *The Journal of Comparative Neurology*, 326(4): 549-560.

Anderson C.M., Bergher J.P., Swanson R.A. (2003). ATP-induced ATP release from astrocytes. *Journal of Neurochemistry*, 88(1): 246-256.

Andres-Mateos E., Perier C., Zhang L., Blanchard-Fillion B., Greco T.M., Thomas B., Ko H.S., Sasaki M., Ischiropoulos H., Przedborski S., Dawson T.M., Dawson V.L. (2007).DJ-1 gene deletion reveals that DJ-1 is an atypical peroxiredoxin-like peroxidase. *Proceedings of the National Academy of Sciences of the United States of America*, 104(37): 14807-14812.

Andriezen W. L., Lond M. B. (1893). The neuroglia elements in the human brain.*The British Medical Journal,* 227-230.

Anthony T.E., Klein C., Fishell G., Heintz N. (2004). Radial Glia Serve as Neuronal Progenitors in All Regions of the Central Nervous Systém. *Neuron*, 41(6): 881-890.

Araque A., Li N., Doyle R.T., Haydon P.G. (2000). SNARE Protein-Dependent Glutamate Release from Astrocytes. *The Journal of Neuroscience*, 20 (2): 666-673

Araque A., Parpura V., Sanzgiri R.P., Haydon P.G. (1998). Glutamate-dependent astrocyte modulation of synaptic transmission between cultured hippocampal neurons. *European Journal of Neuroscience*, 10 (6): 2129-2142.

Arriagada P.V., Growdon J.H., Hedley-Whyte E.T., Hyman B.T. (1992). Neurofibrillary tangles but not senile plaques parallel duration and severity of Alzheimer's disease. *Neurology*, 42 (3): 631-639.

Ascherio A., Chen H., Schwarzschild M.A., Zhang S.M., Colditz G.A., Speizer F.E. (2003). Caffeine, postmenopausal estrogen, and risk of Parkinson's disease. *Neurology*, 60(5): 790-795.

Ashley, A.K. Hinds A.I., Hanneman W.H., Tjalkens R.B., Legare M.E. (2016). DJ-1 mutation decreases astroglial release of inflammatory mediators. *Neurotoxicology*, 52: 198–203

Ashton R.S., Conway A., Pangarkar C., Bergen J., Lim K.-I., Shah P., Bissell M., Schaffer D. (2012). Astrocytes regulate adult hippocampal neurogenesis through ephrin-B signaling. *Nature Neuroscience*, 15: 1399-1406.

Azevedo F.A., Carvalho L.R., Grinberg L.T., Farfel J.M., Ferretti R.E., Leite R.E., Jacob Filho W., Lent R., Herculano-Houzel S. (2009). Equal numbers of neuronal and nonneuronal cells make the human brain an isometrically scaled-up primate brain. *The Journal of Comparative Neurology,* 513(5): 532-41.

Balasingam V., Tejada-Berges T., Wright E., Bouckova R., Yong V.W. (1994) Reactive astrogliosis in the neonatal mouse brain and its modulation by cytokines. *The Journal of Neuroscience*, 14(2): 846-856.

Ball K.K., Gandhi G.K., Thrash J., Cruz N.F., Dienel G.A. (2007). Astrocytic Connexin Distributions and Rapid, Extensive Dye Transfer Via Gap Junctions in the Inferior Colliculus: Implications for [^{14}C]Glucose Metabolite Trafficking. *Journal of Neuroscience Research,* 85(15): 3267-3283.

Balu D.T., Pantazopoulos H., Huang C.C.Y., Muszynski K., Harvey T.L., Uno Y., Rorabaugh J.M., Galloway C.R., Botz-Zapp C., Berretta S., Weinshenker D., Coyle J.T. (2019). Neurotoxic astrocytes express the d-serine synthesizing enzyme, serine racemase, in Alzheimer's disease. *Neurobiology of Disease*, 130: 104511.

Bartus R.T., Elliott P.J., Dean R.L., Hayward N.J., Nagle T.L., Huff M.R., Snodgrass P.A., Blunt D.G. (1996). Controlled Modulation of BBB Permeability Using the Bradykinin Agonist, RMP-7. *Experimental Neurology*, 142: 14-28.

Baudiers, J., Glasser, N., Gerard, D. (1986). Ions Binding to S100 Proteins. *The Journal of Biological Chemistry*, *261*(18): 8192–8203.

*Bélanger M., Magistretti P. J. (2009). The role of astroglia in neuroprotection. *Dialogues in clinical*

neuroscience, 11(3): 281–295.

Bernardos R.L., Barthel L.K., Meyers J.R., Raymond P.A. (2007). Late-Stage Neuronal Progenitors in the Retina Are Radial Müller Glia That Function as Retinal Stem Cells. *The Journal of Neuroscience,* 27(26): 7028-7040.

Bevan S., Chiu S.Y., Gray P.T.A., Ritchie J.M. (1985). The presence of voltage-gated sodium, potassium and chloride channels in rat cultured astrocytes. *Proceedings of the Royal Society of London. Series B, Biological Sciences*, 225: 299-313.

Bezzi P., Gundersen V., Galbete J.L., Seifert G., Steinhäuser C., Pilati E., Volterra A. (2004). Astrocytes contain a vesicular compartment that is competent for regulated exocytosis of glutamate. *Nature Neuroscience,* 7(6): 613-620.

*Booth H.D.E., Hirst W.D., Wade-Martins R. (2017). The Role of Astrocyte Dysfunction in Parkinson's Disease Pathogenesis. *Trends in Neurosciences*, 40, 358-370.

Bowery N.G., Brown D.A., Collins G.G., Galvan M., Marsh S., Yamini G. (1976). Indirect effects of amino-acids on sympathetic ganglion cells mediated through the release of gamma-aminobutyric acid from glial cells. British Journal of Pharmacology, 57: 73–91.

Bozoyan L., Khlghatyan J., Saghatelyan A. (2012). Astrocytes Control the Development of the Migration-Promoting Vasculature Scaffold in the Postnatal Brain via VEGF Signaling. *Journal of Neuroscience*, 32 (5), 1687-1704.

Braak H., Braak E. (1997a). Diagnostic Criteria for Neuropathologic Assessment of Alzheimer's Disease. *Neurobiology of Aging*, 18, 585-588.

Braak H., Braak E. (1997b). Frequency of Stages of Alzheimer-Related Lesions in Different Age Categories. *Neurobiology of Aging*, 18, 351-357.

Brambilla L., Guidotti G., Martorana F., Iyer A.M., Aronica E., Valori C.F., Rossi D. (2016). Disruption of the astrocytic TNFR1-GDNF axis accelerates motor neuron degeneration and disease progression in amyotrophic lateral sclerosis. *Human Molecular Genetics,* 25(14): 3080-3095.

Brawek B., Chesters R., Klement D., Müller J., Lerdkrai C., Hermes M., Garaschuk O. (2018). A bell-shaped dependence between amyloidosis and GABA accumulation in astrocytes in a mouse model of Alzheimer's disease. Neurobiology of Aging, Vol. 61:187-197.

*Brown R.H., Al-Chalabi A. (2017). Amyotrophic lateral sclerosis. *The New England Journal of Medicine*, 377: 162-172.

Bröer S., Rahman B., Pellegri G., Pellerin L., Martin J.-L., Verleysdank S., Hamprecht B., Magistretti P.J. (1997). Comparison of lactate transport in astroglial cells and monocarboxylate transporter 1

(MCT 1) expressing Xenopus laevis oocytes. *The Journal of Biological Chemistry,* 272: 30096–30102.

Bushong E. A., Martone M. E., Ellisman M. H. (2004). Maturation of astrocyte morphology and the establishment of astrocyte domains during postnatal hippocampal development. *International Journal of Developmental Neuroscience*, *22*: 73–86.

Bushong E.A., Martone M.E., Jones Y.Z., Ellisman M.H. (2002). Protoplasmic astrocytes in CA1 stratum radiatum occupy separate anatomical domains. *Journal of Neuroscience*, 22: 183-192.

Cai Z., Zeng W., Tao, K., Lu F., Gao G., Yang Q. (2015). Myricitrin alleviates MPP(+)-induced mitochondrial dysfunction in a DJ-1-dependent manner in SN4741 cells. *Biochemical and Biophysical Research Communication*, 458 (2): 227-233.

Carnell L., Moore H.P. (1994). Transport via the regulated secretory pathway in semi-intact PC12 cells: role of intra-cisternal calcium and pH in the transport and sorting of secretogranin II. *The Journal of Cell Biology*, 127: 693–705.

Castellano B., González B., Jensen M.B., Pedersen E.B., Finsen B.R., Zimmer J. (1991). A double staining technique for simultaneous demonstration of astrocytes and microglia in brain sections and astroglial cell cultures. *The Journal of Histochemistry and Cytochemistry*, 39(5): 561-568.

Cataldo A.M., Broadwell R.D. (1986). Cytochemical identification of cerebral glycogen and glucose-6-phosphatase activity under normal and experimental conditions. II. Choroid plexus and ependymal epithelia, endothelia and pericytes. *Journal of Neurocytology,* 15(4):511–524.

Chanat E., Huttner W.B. (1991). Milieu-induced, selective aggregation of regulated secretory proteins in the trans-Golgi network. *The Journal of Cell Biology*, 115: 1505–1519.

Chen, J.C.T., Chesler, M., (1992). Extracellular alkaline shifts in rat hippocampal slice are mediated by NMDA and non-NMDA receptors. *Journal of Neurophysiology,* 68: 342–344.

Chio A., Calvo A., Moglia C., Mazzini L., Mora G. (2011). Phenotypic heterogeneity of amytrophic lateral sclerosis: a population based study. *Journal of Neurology, Neurosurgery & Psychiatry,* 82(7): 740-746.

Choi M.S., Nakamura T., Cho S.-J., Han X., Holland E.A., Qu J., Petsko G.A., Yates J.R., Liddington R.C., Lipton S.A. (2014). Transnitrosylation from DJ-1 to PTEN Attenuates Neuronal Cell Death in Parkinson's Disease Models. *Journal of Neuroscience*, 34 (45) 15123-15131.

Cholet N., Pellerin L., Magistretti P.J., Hamel E. (2002). Similar perisynaptic glial localization for the Na+, K+-ATPase α2 subunit and the glutamate transporters GLAST and GLT-1 in the rat somatosensory cortex. *Cerebral Cortex*, 12(5): 515–525.

Cruz N.F., Dienel G.A. (2002). High glycogen levels in brains of rats with minimal environmental stimuli: Implications for metabolic contributions of working astrocytes. *Journal of Cerebral Blood Flow & Metabolism,* 22(12): 1476–1489.

Damier P., Kastner A., Agid Y., Hirsch E. C. (1996). Does monoamine oxidase type Bplay a role in dopaminergic nerve cell death in Parkinson's disease? *Neurology,* 46: 1262–1269

Deloulme, J. C., Raponi E., Gentil J., Bertacchi N., Marks A., Baudier J. (2004). Nuclear expression of S100B in oligodendrocyte progenitor cells correlates with differentiation toward the oligodendroglial lineage and modulates oligodendrocytes maturation. *Molecular and Cellular Neuroscience*, *27*, 453–465.

Dick F. D., De Palma G., Ahmadi A., Scott N.W., Prescott G.J., Bennett J., Semple S., Dick S., Counsell C., Mozzoni P., Haites N., Wettinger S.B., Mutti A., Otelea M., Seaton A., Söderkvist P., Felice A.; Geoparkinson study group. (2007). Environmental risk factors for Parkinson's disease and parkinsonism: the Geoparkinson study. *Occupational and Environmental Medicine*, 64: 666–72.

Dringen R., Kranich O., Hamprecht B. (1997). The γ-glutamyltranspeptidase inhibitor preserves glutathione released by astroglial cells in culture. *Neurochemical Research*, 22(6): 727–733.

Dringen R., Pfeiffer B. and Hamprecht B. (1999) Synthesis of the antioxidant glutathione in neurones: Supply by astrocytes of CysGly as precursor for neuronal glutathione. *J. Neurosci.* 19, 562–569.

Drögemüller K., Helmuth U., Brunn A., Sakowicz-Burkiewicz M., Gutmann D.H., Mueller W., Deckert M., Schlüter D. (2008). Astrocyte gp130 expression is critical for the control of Toxoplasma encephalitis. *Journal of Immunology,* 181(4): 2683-2693.

Duan S., Anderson C.M., Keung E.C., Chen Y., Chen Y., Swanson R.A. (2003). P2X7 receptor-mediated release of excitatory amino acids from astrocytes. *The Journal of Neuroscience*, 23(4): 1320-1328.

Ehrhart-Bornstein M., Treiman M., Hansen G.H., Schousboe A., Thorn N.A., Frandsen A. (1991). Parallel expression of synaptophysin and evoked neurotransmitter release during development of cultured neurons. *International Journal of Developmental Neuroscience,* 9(5): 463–471.

Eid T., Lee T.W., Thomas M.J., Amiry-Moghaddam M., Bjornsen L.P., Spencer D.D., Agre P., Ottersen O.P., De Lanerolle N.C. (2005). Loss of perivascular aquaporin 4 may underlie deficient water and K+ homeostasis in the human epileptogenic hippocampus. *Neuroscience*, 102(4): 1193-1198.

Endo F, Komine O., Fujimori-Tonou N., Katsuno M., Jin S., Watanabe S., Sobue G., DezawaM., Wyss-Coray T., Yamanaka K. (2015). Astrocyte-Derived TGF-β1 Accelerates Disease Progression in ALS Mice by Interfering with the Neuroprotective Functions of Microglia and T Cells. *Cell Reports*, 11(4): 592-604.

*Eng, L. F., Ghirnikar, S. R., Lee, L. Y. (2000). Glial Fibrillary Acidic Protein: GFAP-Thirty-One Years. *Neurochemical Resaerch*, 25, 1439-1451.

Faulkner J.R., Herrmann J.E., Woo M.J., Tansey K.E., Doan N.B., Sofroniew M.V. (2004). Reactive astrocytes protect tissue and preserve function after spinal cord injury. *The Journal of Neuroscience*, 24: 2143-2155.

Fowler J. S., Volkow N. D., Wang G. J., Pappas N., Logan J., Shea C., Alexoff D., MacGregor R. R., Schlyer D. J., Zezulkova I., Wolf A. P. (1996). Brain monamine oxidase A inhibition in cigarette smokers. *Proceedings of the National Academy of Sciences, USA93,* 14065-14069.

Gandhi G.K., Cruz N.F., Ball K.K., Dienel G.A. (2009). Astrocytes are poised for lactate trafficking and release from activated brain and for supply of glucose to neurons. *Journal of Neurochemistry*, 111:522–536.

Gegg M.E., Beltran B., Salas-Pino S., Bolanos J.P., Clark J.B., Moncada S., Heales S.J. (2003). Differential effect of nitric oxide on glutathione metabolism and mitochondrial function in astrocytes and neurones: implications for neuroprotection/neurodegeneration? *Journal of Neurochemistry*; 86: 228-237.

Gibb W. R. G., Lees A. J. (1989). The significance of the Lewy body in the diagnosis of idiopathic Parkison's disease. *Neuropathology and Applied Neurobiology*. 15(1), 27–44.

Goate A., Chartier-Harlin M.C., Mullan M., Brown J., Crawford F., Fidani L., Giuffra L., Haynes A., Irving N., James L., et al. (1991). Segregation of a missense mutation in the amyloid precursor protein gene with familial Alzheimer's disease. *Nature*, 349(6311): 704-706.

*Gordon, G. R. J., Mulligan, S. J., Vicar, B. A. M. A. C. (2007). Astrocyte Control of the Cerebrovasculature. *Glia*, 1221:1214–1221.

*Gordon, P. H. (2013). Amyotrophic Lateral Sclerosis : An update for 2013 Clinical Features , Pathophysiology , Management and Therapeutic Trials. *Aging and Disease, 4*(5), 295–310.

Greffard S., Verny M., Bonnet A.-M., Seilhean D., Hauw J.-J., Duyckaerts C. (2010). A stable proportion of Lewy body bearing neurons in the substantia nigra suggests a model in which the Lewy body causes neuronal death. *Neurobiology of Aging*, 31(1), 99–103.

Grosche J., Kettenmann H., Reichenbach A. (2002). Bergmann glial cells form distinct morphological structures to interact with cerebellar neurons. *The Journal of Neuroscience Research*, 68(2): 138-149.

Guastella J., Nelson N., Nelson H., Czyzyk L., Keynan S., Miedel M.C., Davidson N., Lester H.A., Kanner B.I. (1990). Cloning and Expression of a Rat Brain GABA Transporter. *Science*, 249: 1303-

1306.

Halassa M.M., Fellin T., Takano H., Dong J.H., Haydon P.G. (2007). Synaptic islands defined by the territory of a single astrocyte. *Journal of Neuroscience*; 27: 6473–77.

Harlan B.A., Pehar M., Sharma D.R., Beeson G., Beeson C.C. Vargas M.R. (2016). Enhancing NAD+ Salvage Pathway Reverts the Toxicity of Primary Astrocytes Expressing Amyotrophic Lateral Sclerosis-linked Mutant Superoxide Dismutase 1 (SOD1). *The Journal of Biological Chemistry*, 291(20): 10836-10846.

Haun S.E., Segeleon J.E., Trapp V.L., Clotz M.A., Horrocks L.A. (1996). Inosine mediates the protective effect of adenosine in rat astrocyte cultures subjected to combined glucose-oxygen deprivation. *Journal of Neurochemistry*, 67(5): 2051-2059.

*Heneka M. T., Rodríguez J. J., Verkhratsky A. (2009). Neuroglia in neurodegeneration. *Brain Research Reviews*, *63*(1–2): 189–211.

Higashi K., Fujita A., Inanobe A., Tanemoto M., Doi K., Kubo T., Kurachi Y. (2001). An inwardly rectifying K(+) channel, Kir4.1, expressed in astrocytes surrounds synapses and blood vessels in brain. *American Journal of Physiology- Cell Physiology*, 281: C922-C931.

Hsiao H.Y., Chen Y.C., Chen H.M., Tu P.H., Chern Y. (2013). A critical role of astrocyte-mediated nuclear factor-kB-dependent inflammation in Huntington's disease. *Human Molecular Genetics,* 22: 1826–1842.

Illarionova N.B., Brismar H., Aperia A., Gunnarson E. (2014) Role of Na, K-ATPase alpha1 and alpha2 isoforms in the support of astrocyte glutamate uptake. *PLoS One*, 9: 98469

Janzer R.C., Raff M.C. (1987). Astrocytes induce blood-brain barrier properties in endothelial cells. *Nature,* 325(6101): 253-257.

Jo S., Yarishkin O., Hwang Y.J., Chun Y.E., Park M., Woo D.H., Bae J.Y., Kim T., Lee J., Chun H., Park H.J., Lee D.Y., Hong J., Kim H.Y., Oh S.-J., Park S.J., Lee H., et al. (2014). GABA from reactive astrocytes impairs memory in mouse models of Alzheimer's disease. *Nature. Medicine*, 20: 886-896.

Kálmán M., Hajós F. (1989). Distribution of glial fibrillary acidic protein (GFAP)-immunoreactive astrocytes in the rat brain. *Experimental Brain Research*, 78(1): 147-163.

Kaneko N., Marín O., Koike M., Hirota Y., Uchiyama Y., Wu J.Y., Lu Q., Tessier-Lavigne M., Alvarez-Buylla A., Okano H., Rubenstein J.L., Sawamoto K. (2011). New neurons clear the path of astrocytic processes for their rapid migration in the adult brain. *Neuron,* 67:213–223.

Khasnavis S., Pahan K. (2014). Cinnamon treatment upregulates neuroprotective proteins Parkin and

DJ-1 and protects dopaminergic neurons in a mouse model of Parkinson's disease. Journal of Neuroimmune Pharmacology, 9:569–581.

*Kiernan, M. C., Vucic, S., Cheah, B. C., Turner, M. R., Eisen, A., Hardiman, O., Burrel J.R., Zoing, M. C. (2011). Amyotrophic lateral sclerosis. *The Lancet*, *377*(9769), 942–955.

Kim, J.M., Cha S.H., Choi Y.R., Jou I., Joe E.H., Park S.M. (2016). DJ-1 deficiency impairs glutamate uptake into astrocytes via the regulation of flotillin-1 and caveolin-1 expression. *Scientific Reports*, 6, 28823

Knopman D.S., Parisi J.E., Salviati A., et al. (2003). Neuropathology of cognitively normal elderly. *Journal of Neuropathology & Experimental Neurology,* 62: 1087–95.

Ko, Y.P., Lang, H.J., Loh, S.H., Chu, K.C.,Wu, M.L., (1999). Cl^--dependent and Cl^--independent Na+/ $HCO3^-$ acid extrusion in cultured rat cerebellar astrocytes. *Chinese Journal of Physiology*, 31: 237–248.

Kreda S.M., Seminario-Vidal L., Van Heusden C., Lazarowski E.R. (2009). Thrombin-promoted release of UDP-glucose from human astrocytoma cells. *British Journal of Pharmacology*, 153(7): 1528-1537.

Krum J.M., Mani N., Rosenstein J.M. (2002). Angiogenic and astroglial responses to vascular endothelial growth factor administration in adult rat brain. *Neuroscience*, 110(4): 589-604.

Larsen B. R., Assentoft M., Cotrina M. L., Hua S. Z., Nedergaard M., Kaila K., et al. . (2014). Contributions of the Na(+)/K(+)-ATPase, NKCC1, and Kir4.1 to hippocampal K(+) clearance and volume responses. *Glia,* 62: 608–622.

Lee M., Schwab C., McGeer P.L. (2011). Astrocytes are GABAergic cells that modulate microglial activity. *Glia,* 59:152-165.

Lee W., Reyes R. C., Gottipati M. K., Lewis K., Lesort M., Parpura V., Gray M. (2013). Enhanced Ca^{2+}-dependent glutamate release from astrocytes of the BACHD Huntington's disease mouse model. *Neurobiology of Disease,* 58: 192-199.

Lenart B., Kintner D.B., Shull G.E., Sun D. (2004). Na-K-Cl Cotransporter-Mediated Intracellular Na+ Accumulation Affects Ca2+ Signaling in Astrocytes in an In Vitro Ischemic Model. *Journal of Neuroscience*, 24: 9585-9597.

Lenhossek Mv. (1891). Zur Kenntnis der Neuroglia des menschlichen Ruckenmarkes. *Verh Anat Ges*.; 5:193–221.

Levy-Lahad E., Wasco W., Poorkaj P., et al. (1995) Candidate gene for the chromosome 1 familial Alzheimer's disease locus. *Science*, 269: 973-977

Li D., Hérault K., Isacoff E.Y., Oheim M., Ropert N. (2012). Optogenetic activation of LiGluR-expressing astrocytes evokes anion channel-mediated glutamate release. The Journal of Physiology; 590(4): 855-873.

Liddelow S.A., Guttenplan K.A., Clarke L.E., Bennett F.C., Bohlen C.J., Schirmer L., Bennett M.L., Münch A.E., Chung W.S., Peterson T.C., et al. (2017). Neurotoxic reactive astrocytes are induced by activated microglia. *Nature*, 541: 481-487.

Linstedt A.D., Mehta A., Suhan J., Reggio H., and Hauri H.P. (1997). Sequence and overexpression of GPP130/GIMPc: evidence for saturable pH-sensitive targeting of a type II early Golgi membrane protein. *Molecular Biology of the Cell,* 8: 1073–1087.

Logan S., Pharaoh G.A., Marlin M.C., Masser D.R. et al. (2018). Insulin-like growth factor receptor signaling regulates working memory, mitochondrial metabolism, and amyloid-β uptake in astrocytes. *Molecular Metabolism,* 9: 41-155.

Lunde L.K., Camassa L.M., Hoddevik E.H., Khan F.H., Ottersen O.P., Boldt H.B., Miry-Moghaddam M. (2015). Postnatal development of the molecular complex underlying astrocyte polarization. *Brain Struct. Funct*., 220: 2087 - 2101.

Mac M., Nałęcz K.A. (2002). Expression of monocarboxylic acid transporters (MCT) in brain cells. Implication for branched chain a-ketoacids transport in neurons. *Neurochemistry Int*ernational, 43: 305–309.

MacAulay N., Gether U., Klaerke D.A., Zeuthen T. (2001). Water transport by the human Na+-coupled glutamate cotransporter expressed in Xenopus oocytes. The Journal of Physiology, 530:367–378.

*Maccioni R. B., Muñoz P., Barbeito L. (2001). The Molecular Bases of Alzheimer's Disease and Other Neurodegenerative Disorders. *Archives of Medical Research, 32*, 367–381.

Makar T.K., Nedergaard M., Preuss A., Gelbard A.S., Perumal A.S., Cooper A.J. (1994). Vitamin E, ascorbate, glutathione, glutathione disulfide, and enzymes of glutathione metabolism in cultures of chick astrocytes and neurons: evidence that astrocytes play an important role in antioxidative processes in the brain. *Journal of Neurochemistry*; 62:45-53.

Marín-Padilla M. (1995). Prenatal development of fibrous (white matter), protoplasmic (gray matter), and layer I astrocytes in the human cerebral cortex: a Golgi study. *The Journal of Comparative Neurology,* 357:554–572.

Mawuenyega K.G., Sigurdson W., Ovod V., Munsell L., Kasten T., Morris J.C., Yarasheski K.E., Bateman R.J. (2010). Decreased clearance of CNS beta-amyloid in Alzheimer's disease. *Science*, 330:1774.

Mena M. A., de Bernardo S., Casarejos M. J., Canals S., Rodriguez-Martin E. (2002). The role of astroglia on the survival of dopamine neurons. *Molecular Neurobiology*, 25: 245–263.

Miller R. H., Raff C. M. (1984). Fibrous and protoplasmic astrocytes are biochemically distinct. *The Journal of Neuroscience, 4*(2): 585–592.

Miyake T., Kitamura T. (1992). Glutamine synthetase immunoreactivity in two types of mouse brain glial cells. *Brain Research*, 586: 53–60.

Morgello S., Uson R. R., Schwartz E. J., Haber R. S. (1995). The human blood-brain barrier glucose transporter (GLUT1) is a glucose transporter of gray matter astrocytes. *Glia*, 14(1): 43–54.

Morita K., Sasaki H., Furuse M., Tsukita S. (1999). Endothelial Claudin: Claudin-5/TMVCF Constitutes Tight Junction Strands in Endothelial Cells. *The Journal of Cell Biology*, 147(1): 185-194.

Morris J.C., Storandt M., Miller P., McKeel D.W., Price J.L., Rubin E.H., Berg L. (2001). Mild cognitice impairment represents early-stage Alzheimer disease. *Archives of neurology*, 58: 397-405.

Moura M.C., Casulari L.A., Novaes M.R.C.G. (2016). Ethnic and demographic incidence of amyotrophic lateral sclerosis (ALS) in Brazil: a population based study. *Amyotrophic Lateral Sclerosis and Frontotemporal Degeneration,* 17: 275-281.

Mulligan S.J., MacVicar B.A. (2004). Calcium transients in astrocyte endfeet cause cerebrovascular constrictions. *Nature*, 431(7005): 195-199.

Nagelhus E.A., Horio Y., Inanobe A., Fujita A., Haug F.M., Nielsen S., Kurachi Y., Ottersen O.P. (1999) Immunogold evidence suggests that coupling of K+ siphoning and water transport in rat retinal Muller cells is mediated by a coenrichment of Kir4. 1 and AQP4 in specific membrane domains. *Glia*, 26: 47-54.

Nagy J.I., Rash J.E. (2003). Astrocyte and Oligodendrocyte Connexins of the Glial Syncytium in Relation to Astrocyte Anatomical Domains and Spatial Buffering. *Cell Communication and Adhesion,* 10: 40-406.

*Newington, J. T., Harris, R. A., Cumming, R. C. (2013). Reevaluating Metabolism in Alzheimer's Disease from the Perspective of the Astrocyte-Neuron Lactate Shuttle Model. *Journal of Neurodegenerative Disease.*

Newman E.A., Frambach D.A., Odette L.L. (1984). Control of Extracellular Potassium Levels by Retinal Glial Cell K+ Siphoning. *Science*, 225(4667): 1174-1175.

Nolte C., Matyash M., Pivneva T., Schipke C.G., Ohlemeyer C., Hanisch U.K., Kirchhoff F., Kettenmann H. (2001). GFAP promotercontrolled EGFP-expressing transgenic mice: a tool to visualize astrocytes and astrogliosis in living brain tissue. *Glia,* 33: 72–86.

Oberheim N.A., Takano T., Han X., He W., Lin J.H., Wang F., Xu Q., Wyatt J.D., Pilcher W., Ojemann J.G., Ransom B.R., Goldman S.A., Nedergaard M. (2009). Uniquely hominid features of adult human astrocytes. *The Journal of Neuroscience: the official Journal of the Society for Neuroscience*, 29: 3276–3287.

Ogata K., Kosaka T. (2002). Structural and quantitative analysis of astrocytes in the mouse hippocampus. *Neuroscience,* 113(1).

*Ohno Y., Kinboshi M., Shimizu S. (2018). Inwardly Rectifying Potassium Channel Kir4.1 as a Novel Modulator of BDNF Expression in Astrocytes. *International Journal of Molecular Science,* 19: 3313.

Oliveira J. M. A., Jekabsons M. B., Chen S., et al. (2007). Mitochondrial dysfunction inHuntington's disease: the bioenergetics of isolated and in situ mitochondria from transgenic mice. *Journal of Neurochemistry*, 101: 241-249.

Ottersen O.P., Zhang N., Walberg F. (1992). Metabolic compartmentation of glutamate and glutamine: morphological evidence obtained by quantitative immunocytochemistry in rat cerebellum. *Neuroscience*, 46(3): 519-34.

Paísan-Ruíz, C., Jain, S., Evans, E. W., et al. (2004). Cloning of the gene containing mutations that cause PARK8-linked Parkinson's disease. *Neuron*. 44: 595–600.

Papanikolaou M., Lewis A., Butt A.M. (2019). Glial and neuronal expression of the Inward Rectifying Potassium Channel Kir7.1 in the adult mouse brain. *Journal of Anatomy*, 235: 984-996.

Parpura V., Basarsky T.A., Liu F., Jeftinija K., Jeftinija S., Haydon P.G. (1994). Glutamate-mediated astrocyte-neuron signalling. *Nature*, 369:744-747.

Pellerin L., Magistretti P.J. (1994). Glutamate uptake into astrocytes stimulates aerobic glycolysis: A mechanism coupling neuronal activity to glucose utilization. *Proceedings of the National Academy of Sciences of the United States of America*, 91(22): 10625-10629.

Pellerin L., Pellegri G., Bittar P.G., Charnay Y., Bouras C., Martin J.-L., Stella N., Magistretti P.J. (1998). Evidence Supporting the Existence of an Activity-Dependent Astrocyte-Neuron Lactate Shuttle. *Developmental Neuroscience,* 20: 291-299.

Phelps C.H. (1972). Barbiturate-induced glycogen accumulation in brain. An electron microscopic study. *Brain Research,* 39:225–234.

Philippe J.-M., Dubois J.-M., Rouzaire-Dubois B., Cartron P.-F., Vallette F., Morel N. (2002). Functional expression of V-ATPases in the plasma membrane of glial cells. *Glia,* 37, 365–373.

Pike C.J., Cummings B.J., Monzavi R., Cotman C.W. (1994). Beta-amyloid-induced changes in cultured

astrocytes parallel reactive astrocytosis associated with senile plaques in Alzheimer's disease. *Neuroscience*, 63: 517-531.

Pines G., Danbolt N.C., Bjørås M., Zhang Y., Bendahan A., Eide L., Koepsell H., Storm-Mathisen J., Seeberg E., Kanner B.I. (1992). Cloning and expression of a rat brain L-glutamate transporter. *Nature*, 360:464–467.

Price J.L., Morris J.C. (1999). Tangles and plaques in nondemented aging and "preclinical" Alzheimer's disease. *Annals of Neurology*, 45: 358–68.

Puri S., Bachert C., Fimmel C.J., Linstedt A.D. (2002). Cycling of early Golgi proteins via the cell surface and endosomes upon lumenal pH disruption. *Traffic*, 3: 641–653.

Quandt F.N., MacVicar B.A. (1986). Calcium activated potassium channels in cultured astrocytes. *Neuroscience*, 19: 29-41.

Rannikko E.H. (2015). Exogenous alpha-synuclein induces toll-like receptor 4 dependent inflammatory responses in astrocytes. *BMC Neuroscience,*16:57.

Rosen D.R., Siddique T., Petterson D., Figlewicz D.A., Sapp P., Hentati A., Donaldson D., Goto J., O'Regan J.P., et al. (1993). Mutations in Cu/Zn superoxide dismutase gene are associated with familial amyotrophic lateral sclerosis. *Nature*, 362: 59-62.

Rosenblatt A., Liang K.Y., Zhou H., Abbott M.H., Gourley L.M., Margolis R.L., Brandt J., Ross C.A. (2006). The association of CAG repeat length with clinical progression in Huntington disease. *Neurology*, 66: 1016–1020.

*Ross, C. A., Tabrizi, S. J. (2011). Huntington's disease : from molecular pathogenesis to clinical treatment. *The Lancet Neurology*, 10(1), 83–98.

Rothstein J.D., Dykes-Hoberg M., Pardo C.A., Bristol L.A., Jin L., Kuncl R.W., Kanai Y., Hediger M.A., Wang Y., Schielke J.P., Welty D.F. (1996). Knockout of glutamate transporters reveals a major role for astroglial transport in excitotoxicity and clearance of glutamate. *Neuron*, 16(3):675-86.

Rouach N., Koulakoff A., Abudara V., Willecke K., Giaume C. (2008). Astroglial metabolic networks sustain hippocampal synaptic transmission. *Science*, 322:1551–1555.

Rutledge E.M., Aschner M., Kimelberg H.K. (1998). Pharmacological characterization of swelling-induced d-[3H]aspartate release from primary astrocyte cultures. *American Journal of Physiology*, 274(6): C1511-C1520.

Schell M.J., Molliver M.E., Snyder S.H. (1995). D-Serine, an endogenous synaptic modulator: Localization to astrocytes and glutamate-stimulated release. *Proceedings of the National Academy*

of Sciences USA, 92: 3948-3952.

Schneider G.H., Baethmann A., Kempski O. (1992) Mechanisms of glial swelling induced by glutamate. *Canadian Journal of Physiology and Pharmacology*, 70:S334–S343.

*Selkoe, D. J. (2001). Alzheimer's Disease : Genes, Proteins, and Therapy. *Physiological Reviews*, *81*(2), 741–766.

Shain W.G., Martin D.L. (1984) Activation of beta-adrenergic receptors stimulates taurine release from glial cells. *Cellular and Molecular Neurobiology*, 4:191–6.

Shendelman S., Jonason A., Martinat C., Leete T., Abeliovich A. (2004). DJ-1 is a redox-dependent molecular chaperone that inhibits alpha-synuclein aggregate formation. *PLOS Biology*., 2: e362.

Sherrington R., Rogaev E.I., Liang Y., et al. (1995). Cloning of a gene bearing missense mutations in early-onset familial Alzheimer's disease. *Nature*, 375: 754-760.

Shibasaki K., Hosoi N., Kaneko R., Tominaga M., Yamada K. (2016). Glycine release from astrocytes via functional reversal of GlyT1. *Journal of Neurochemistry*, 140(3): 395-403.

Sibson N.R., Dhankhar A., Mason G.F., Behar K.L., Rothman D.L., Shulman R.G. (1997). In vivo 13C NMR measurements of cerebral glutamine synthesis as evidence for glutamate-glutamine cycling. *Proceedings of the National Academy of Sciences USA*., 94(6):2699-2704.

Silver J., Edwards M.A., Levitt P. (1993). Immunocytochemical demonstration of early appearing astroglial structures that form boundaries and pathways along axon tracts in the fetal brain. *The Journal of Comparative Neurology*, 328: 415-436.

Simard M., Arcuino G., Takano T., Liu Q.S., Nedergaard M. (2003). Signaling at the Gliovascular Interface. *The Journal of Neuroscience*, 23(27): 9254-9262.

*Simard M., Nedergaard M. (2004). The neurobiology of glia in the context of water and ion homeostasis. *Neuroscience, 129*, 877–896.

Skotte N. H., Andersen J. V., Santos A., Aldana B.I., Willert C.W., Norremolle A., Waagepetersen H.S., Nielsen M.L. (2018). Integrative Characterization of the R6/2 Mouse Model of Huntington's Disease Reveals Dysfunctional Astrocyte Metabolism. *Cell Reports*, 23: 2211–2224.

*Sofroniew M. V. (2009). Molecular dissection of reactive astrogliosis and glial scar formation. *Trends in Neuroscience,* 32(12).

Söllvander S., Nikitidou E., Brolin R., Söderberg L., Dag S., Lannfelt L., Erlandsson A. (2016). Accumulation of amyloid-beta by astrocytes result in enlarged endosomes and microvesicle-

induced apoptosis of neurons. *Moecular Neurodegeneration*, 11:38.

Song, H., Stevens, C. F., Gage, F. H. (2002). Astroglia induce neurogenesis from adult neural stem cells. *Nature,* 39–44.

Stelzmann R.A., Schnitzlein H.N., Murtagh F.R. (1995). An English translation of Alzheimer's 1907 Paper, " Über eine eigenartige Erlranliung der Hirnrinde". *Clinical Anatomy*, 8: 429-431.

Storck T., Schulte S., Hofman K., Stoffel W. (1992) Structure, expression, and functional analysis of a Na+-dependent glutamate/aspartate transporter from rat brain. *Proceedings of the National Academy of Sciences USA,* 89:10955–10959.Stout C.E., Constantin J.L., Naus C.C.G., Charles A.C. (2002) Intercellular Calcium Signaling in Astrocytes via ATP Release through Connexin Hemichannels. *Journal of biological Chemistry*,277(12), 10482-10488.

Straub S.V., Bonev A.D., Wilkerson M.K., Nelson M.T. (2006). Dynamic Inositol Trisphosphate-mediated Calcium Signals within Astrocytic Endfeet Underlie Vasodilation of Cerebral Arterioles. *Journal of General Physiology*, 128(6) 659-669.

Takano T., Kang J., Jaiswal J.K., Simon S.M., Lin J.H., Yu Y., Li Y., Yang J., Dienel G., Zielke H.R., Nedergaard M. (2005). Receptor-mediated glutamate release from volume sensitive channels in astrocytes. *Proceeding of the National Academy of Sciences USA*, 102: 16466-16471.

Takumi T., Ishii T., Hori Y., Morishige K.I., Takahashi N., Yamada M., Yamashita T., Kiyama H., Sohmiya K., Nakanishi S., Kurachi Y. (1995). A novel ATP-dependent inward rectifier potassium channel expressed predominantly in glial cells. *Journal of Biological Chemistry*, 270: 16339-16346.

Tanaka J, Toku K, Zhang B, Ishihara K, Sakanaka M, Maeda N. (1999). Astrocytes prevent neuronal death induced by reactive oxygen and nitrogen species. *Glia*, 28:85-96.

Tapiola T., Overmyer M., Lehtovirta M., Helisalmi S., Ramberg J., Alafuzoff I., Riekkinen P., Hilkka S. (1997).The level of cerebrospinal fluid tau correlates with neurofibrillary tangles in Alzheimer's disease. *NeuroReport*, 8: 3961- 3963.

Tong X., Ao Y., Faas G. C., et al. (2014). Astrocyte Kir4.1 ion channel deficits contribute to neuronal dysfunction in Huntington's disease model mice. *Nature Neuroscience.* 17(5), 694-703.

Valenza M., Leoni V., Karasinska J.M., Petricca L., Fan J., Carroll J., Pouladi M.A., Fossale E. Nguyen H.P., Riess O., et al. (2010). Cholesterol defect is marked across multiple rodent models of Huntington's disease and is manifest in astrocytes. *Journal of Neuroscience,* 30: 10844–10850.

Vaughn J. E., Pease D. C. (1967). Electron microscopy of classically stained astrocytes. *The Journal of Comparative Neurology*, 131(2), 143–153.

*Verkhratsky A., Parpura V. (2010). Recent advances in (patho) physiology of astroglia. *Acta Pharmacologica Sinica*, 1044–1054.

*Verkhratsky A., Rodríguez J. J., Parpura V. (2013). Astroglia in neurological diseases. *Future Neurology*, *8*(2).

*Verkhratsky, A., Sofroniew, M. V, Messing, A., Nihal, C. I., Rempe, D. (2012). Neurological diseases as primary gliopathies : a reassessment of neurocentrism. *ASN NEURO*, *4*(3).

Virchow R. (1858). Die Cellularpathologie in ihrer Begründung auf physiologische and pathologische Gewebelehre. Zwanzig Vorlesungen gehalten während der Monate Februar, März und April 1858 im pathologischen Institut zu Berlin. 1. Hirschwald; Berlin, 440.

Visser P.J., Verhey F., Knol D.L., et al. (2009) Prevalence and prognostic value of CSF markers of Alzheimer's disease pathology in patients with subjective cognitive impairment or mild cognitive impairment in the DESCRIPA study: a prospective cohort study. *The Lancet Neurology*; 8: 619–627.

Voskhul R.R., Peterson R.C., Song B., Ao Y., Morales L.B.J., Tiwari-Woodruff S., Sofroniew M.V. (2009). Reactive Astrocytes Form Scar-Like Perivascular Barriers to Leukocytes during Adaptive Immune Inflammation of the CNS. *The Journal of Neuroscience*, 29(37): 11511-11522.

Wada M., Miyakawa S., Shimada A., Okada N., Yamamoto A., Fujita T. (2005). Functional linkage of H+/peptide transporter PEPT2 and Na+/H+ exchanger in primary cultures of astrocytes from mouse cerebral cortex. *Brain Research*, 1044, 33–41.

Wallraff A., Odermatt B., Willecke K., Steinhäuser C. (2004). Distinct Types of Astroglial Cells in the Hippocampus Differ in Gap Junction Coupling. *Glia*, 48: 36-43.

Wanner I.B., Anderson M.A., Song B., Levine J., Fernandez A., Gray-Thompson Z., Ao Y., Sofroniew M.V. (2013). Glial scar borders are formed by newly proliferated, elongated astrocytes that interact to corral inflammatory and fibrotic cells via STAT3-dependent mechanisms after spinal cord injury. *The Journal of Neuroscience*, 33: 12870-12886.

Warr O., Takahashi M., Attwell D. (1999) Modulation of extracellular glutamate concentration in rat brain slices by cystine-glutamate exchange. *The Journal of Physiology*, 514: 783–93.

Wilhelmsson U., Faiz M., De Pablo Y., et al. (2012). Astrocytes Negatively Regulate Neurogenesis Through the Jagged1-Mediated Notch Pathway. *Stem Cells*, 30: 2320-2329.

Wu Z., Guo Z., Gearing M., Chen G. (2014). Tonic inhibition in dentate gyrus impairs long-term potentiation and memory in an Alzheimer's disease model. *Nature Communications*, 5: 4159.

Yang Y., Videnský S., Jin L. I. N., Jie C., Lorenzini I., Frankl M., Rothstein J. D. (2011). Molecular

Comparison of GLT1$^+$ and ALDH1L1$^+$ Astrocytes In Vivo in Astroglial Reporter Mice. *Glia*, 200–207.

Ye Z.C., Wyeth M.S., Baltan-Tekkok S., Ransom B.R. (2003). Functional hemichannels in astrocytes: a novel mechanism of glutamate release. *The Journal of Neuroscience,* 23: 3588-3596.

Zhao J., O'connor T., Vassar R. (2011). The contribution of activated astrocytes to Abeta production: implications for Alzheimer's disease pathogenesis. *Journal of Neuroinflammation*, 8: 150.

Zhou W., Zhu M., Wilson M.A., Petsko G.A., Fink A.L. (2006). The oxidation state of DJ-1 regulates its chaperone activity toward alpha-synuclein. *Journal of Molecular Biology*, 356, pp. 1036-1048.

(* = Review)